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Invited Paper

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Among his most significant awards are the Presidential Commendation in 1971, the Charles B. Dudley Award in 1978 for excellence in publications from the American Society for Testing and Materials and the Founder's Award of the Society for Environmental Toxicology and Chemistry in 1981. In August 1984, Dr. Cairns received the American Water Resources Association's Icko Iben Award for his many contributions in furthering interdisciplinary communications in various aspects of water resources. Most recently, in December 1984, he received the U.S. Department of Agriculture's Research Service B. Y. Morrison Medal at the International Chemical Congress of the Pacific Basin Societies. The B. Y. Morrison Memorial Lectureship was established in 1968 to recognize outstanding accomplishments in the environmental sciences and to stress the urgency of preserving and enhancing man's environment. This paper was the text for the Morrison Memorial Lecture and Luncheon held in conjunction with the award ceremonies.

EVALUATING THE OPTIONS FOR WATER QUALITY MANAGEMENT[1]

John Cairns, Jr.[2]

ABSTRACT: The agricultural revolution occurred because the unmanaged environment was not providing food in either the quantity or quality that society desired. The "environmental revolution" is developing because the unmanaged environment is clearly not capable of assimilating societal wastes without being seriously degraded. Effective environmental management will require regional and site-specific modification of general principles and practices that can be used at a national or international level. An environmental management system can be operated in the same manner as any industrial quality control system with three basic components: (a) sensors at appropriate locations, (b) rapid generation and feedback of information, and (c) a quality control group capable of taking immediate effective action when system performance is outside predetermined boundary conditions. This discussion focuses primarily on three areas: (a) management options available to regulate intrusion of societal wastes into natural systems, (b) types of methods available for predicting and validating effects on natural systems, and (c) modifications of present legislation that would permit the most flexibility in selecting from the various management options. Also considered are multispecies toxicity tests using species with cosmopolitan distribution in test systems with a high degree of environmental realism. Among the many values of such tests is the ability to exchange information from all parts of the world effectively because the test organisms are not restricted to a particular geographic region.
(KEY TERMS: water quality; hazard evaluation; water management; criteria; monitoring; legislation.)

INTRODUCTION

Ecological consciousness did not become a major feature of industrialized societies until about 15 years ago. The suddenness of the development appears to have been a consequence of exceeding the assimilative capacity of the environment for societal wastes in a variety of ways and locations, producing

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effects that could not escape attention. The similarity to the agricultural revolution, in which the unmanaged environment did not produce food in sufficient quantity or quality to meet the expectations of society, is instructive. The environmental revolution is the result of a variety of unmistakable signals from natural systems that a tolerance threshold for societal wastes has been crossed in sites in almost every part of the world. As Woodwell (1984) notes, the sudden surge in ecological awareness probably resulted from the widespread intensive use of DDT, which produced biologically dramatic effects and which could be associated with chemical concentrations in animal tissues. The unmistakable evidence of transfer of DDT through the food chain to humans drew the attention of people who might otherwise have ignored these events.

Unfortunately, the young but rapidly developing field of ecology was still in the observational phase of development and had only begun to enter the predictive phase. One could document effects of toxicants, but toxicity tests to predict their effects before use were not in general use. Furthermore, the sophisticated tests needed for accurate predictions could not be developed quickly. The challenge of predicting environmental response to a variety of anthropogenic stresses was not comparable to catching up with the Russians following the launching of Sputnik. As Slobodkin (1984) notes:

> In contrast to molecular genetics and biochemistry, which have at times had a relatively small number of empirical questions that needed answering before understanding could advance, central focal questions do not generally exist in ecology. There are no single obvious-next-questions. The problems of ecology span the full range of interactions among the earth's two million species of organisms and their environment. Production of a unitary, comprehensible, ecological theory is thus completely intractable. There is no present hope for deriving the mechanistic basis of, say, a tropical rain forest in the same way that one can begin to understand the mechanistic basis for nerve conduction, muscle contraction, or photosynthesis.

Slobodkin also notes that practical questions are not going to wait for ecologists to complete their "homework." This is quite evident if one examines the present system.

The management of water quality now depends on a curious patchwork of laws, methods, and treatment systems lacking a unifying theme. Such management might be characterized as an attempt to control the entry of societal waste into the environment based on indirect evidence of the probable effects. Given the lack of system and structure that has characterized pollution control efforts in the past, it is astonishing that results have been as good as they appear to be. These relatively good results may be due to one or both of two factors: (a) the assimilative capacity of natural systems is generally much greater than it was thought to be and/or (b) we have failed to detect all but the grossest ecosystem responses, such as fishkills and the like.

If environmental protection is viewed as a quality control problem, the unifying components become clear:

1. An explicit description of the environmental qualities to be protected. Statements such as "protection of fish, shellfish, and wildlife" are not adequate for this purpose. Specific functional and structural characteristics to be protected must be identified with sufficient precision that a quantitative determination can be made when these fall outside the boundaries of natural variability and could legitimately be characterized as a stress response (Odum, *et al.*, 1979). While some guidelines might be developed on a national scale for this purpose, an effective quality control system must depend primarily on regional and even site-specific criteria. As a consequence, an illustrative list of control parameters might be produced from which local or regional selections could be made with the recognition that each ecosystem is sufficiently unique that special parameters might be necessary for maintaining quality control. It is also worth noting that the protection of rare and endangered species will almost certainly require a different strategy from the protection of ecosystems. Ecosystems are characterized by successional processes and other types of change whereas the protection of rare and endangered species requires that optimal conditions, or as close to optimal as is possible for that species, be maintained in perpetuity.
2. A general agreement is needed on the methods and procedures necessary to determine how well the qualities selected are staying within acceptable ranges. A variety of both predictive and reactive (feedback loop) tests can be used for this purpose (Cairns, 1982a). Biological tests are essential because no instrument built by man can measure toxicity; however, chemical and physical tests must also be conducted because biological response alone makes determining the cause difficult. Desirable chracteristics of single species tests are now generally understood. Desirable characteristics of multispecies tests have been discussed by Tebo (in press), Loewengart and Maki (in press), and Cairns (1984). Among the most important desirable characteristics of tests are: (a) low frequency of false negatives or positives, (b) readily usable by a large variety of institutions and individuals, and (c) end point should be a discrete variable.
3. A rapid and effective response must follow whenever the quality control conditions are not within the predetermined parameters. This is the most drastic change for those accustomed to settling environmental problems in the courts when such problems really should be resolved by a professional management team charged with that responsibility.

In this discussion, I will first examine present management options for ensuring that the assimilative capacity of the environment is not exceeded. This includes an analysis of various options and the selection of the most desirable ones. Second, implementation of the options with present methodology is examined, as well as areas where further research is needed for fully effective management. Modifications of present legislation that will enable the regulatory implementation of the strategy are also discussed.

MANAGEMENT OPTIONS

The basic management responsibility is to optimize the benefits of a technological society without damaging natural life support systems. It is clear that many citizens of the world would not accept this minimal goal but would insist on preserving aesthetic and recreational benefits as well. The major difficulty in optimizing benefits is that neither the assimilative capacity of natural systems nor the output of the global technological society is constant. Furthermore, the discharge of anthropogenic wastes into the environment does not cycle in phase with various natural cycles. As a consequence, management must either discharge wastes into the natural receiving systems using limits developed for "worst possible case conditions" or establish a feedback loop from natural systems that will enable quick and effective tracking of changes in assimilative capacity. There are only three basic options available to regulate the intrusion of anthropogenic wastes into natural systems: (a) "pipe standards" – controlling both the quality and quantity of discharges into natural systems at the discharge pipe, (b) technology-based standards – installing the best practical or best applicable technology in hope of adequately protecting the environment, and (c) receiving system standards – using the condition or quality of the receiving system as a means of regulating waste discharges. Both pipe standards and technology-based standards have two serious weaknesses: (a) they do not use direct measurements of the impact on natural systems as a means of quality control, and (b) they ignore the well established fact that much environmental stress is the result of nonpoint source discharges. Receiving system standards and criteria also have two serious drawbacks: (a) as is the case for agricultural practices, regional climatic and environmental conditions are so different that a high degree of site-specificity is essential, and (b) there is no general agreement among environmental professionals about the end points or parameters essential to a quality control system based on receiving system criteria and standards.

The proceedings of the seminar *Development and Assessment of Environmental Standards,* published in 1983 by the American Academy of Environmental Engineers, lists other faults of both pipe and technology-based standards. Knowing the chemical concentration leaving a pipe does not ensure accurate prediction of its toxicity in a particular receiving system because environmental quality mediates the toxic response. Also, chemicals from several pipes can interact synergistically in the receiving system to increase toxicity markedly. An additional fault of technology-based standards is the failure to consider either the size of the receiving system or its environmental characteristics. Because of these deficiencies, receiving system criteria and standards are being re-examined. Perhaps ecologists will stop squabbling among themselves and recognize that if the informed professionals are not willing to endorse end points or parameters for water quality control then less informed people will do so or go back to pipe and technology-based standards. Since receiving system standards have worked rather well for thermal discharges (e.g., Section 316A, Public Law 92500) in the United States, they should be given a more extensive trial for other anthropogenic wastes.

TYPES OF METHODS AVAILABLE FOR PREDICTING AND VALIDATING EFFECTS OF ANTHROPOGENIC STRESS ON NATURAL SYSTEMS

Basically three different categories of tests are available to determine the toxicity or environmental stress of materials discharged into the environment or activities producing environmental disturbance. In broad, general terms these are: (a) single species laboratory toxicity tests using lethality or some other easily observed response as an end point; (b) more complicated tests that are conducted primarily, but not entirely, in the laboratory with systems having a higher degree of environmental realism (these are generally called microcosms) or with even larger systems, often called field units or designated as mesocosms (e.g., see the excellent article by Odum, 1984); (c) field studies at the site of discharge which usually involve an inventory of species in a reference station compared to stations exposed to the stress, but sometimes involve a functional assessment such as detritus processing or energy flow.

Single Species Tests

Sloof (1983) feels that single species toxicity tests for environmental protection are a direct outgrowth of the tests designed to protect human health. He notes that this is unfortunate because, while the tests are similar, the ways in which the tests are used are quite different. Single species tests are generally, but not entirely, carried out in relatively simple apparatus with a small number of organisms not greatly different in size or other easily measured characteristics. These organisms are generally required to be relatively free of easily observable diseases, and mortality in the control tank must be 10 percent or less during the test period. The simplest of single species tests are inexpensive, can be carried out in both mobile and stationary laboratories by personnel with relatively little scientific training, and appear to have worked rather well for the past 30 years. Their success is probably due to two factors: (a) results of a short-term test are usually multiplied by an application factor which very substantially reduces the response concentration to an estimated "safe" concentration; (b) sophisticated, scientifically justifiable validation of the predictions made with single species tests in natural systems by methods acceptable to professional ecologists has been exceedingly rare (National Research Council, 1981).

Microcosms and Mesocosms

The use of microcosms and mesocosms, though not a new approach, has become increasingly common in the last few years. The publication *Environmental Toxicology and Chemistry* of the Society for Environmental Toxicology and Chemistry (a relatively young but vigorous professional society) shows an extraordinary interest in these systems. Their chief advantage is that they have a higher degree of environmental realism than single species tests so that studies of chemical fate and transformation can be combined with environmental toxicology. The chief weakness of the systems is that even now only a relatively few research investigators in industry, regulatory agencies, and universities have extensive experience with microcosms and mesocosms. Some are no more costly than the moderately sophisticated single species tests, while others are quite elaborate, require highly trained personnel for successful operation, and may be an order of magnitude more costly than single species tests.

Field Studies

A large number of methods has been used for 30 years or more for field studies in aquatic systems. Most of these might be inelegantly characterized as "critter counting." Although many of the methods can be used in a wide variety of situations, a far higher degree of design adjustment is inevitably required by site-specificity than is necessary for either single species or microcosm tests. Since field tests are likely to be more costly than either of the other categories, skillful sampling design and interpretation of data are mandatory. A major drawback is that one cannot tell in many situations whether the stress is approaching a response threshold or whether it is quite far away. For the other categories of tests, one can set-up a graded series of concentrations of chemicals, temperatures, suspended solids, and the like to ensure that the observable effects response threshold is determined. In field studies, crossing this threshold is undesirable even for experimental purposes because the natural system will be damaged. This can partly be offset by doing studies directly at the outfall before the effluent is well mixed with the receiving water; assume then that if a response occurs it will be detected there first and that the damage, if any, will be fairly limited. This gives an even more precise determination of the natural response threshold than the laboratory tests. Another problem with field studies is distinguishing between the high degree of natural variability characteristic of most systems and a trend of degradation caused by a deleterious waste discharge. However, if field studies are accompanied by single species toxicity tests and/or microcosm and mesocosm tests, the array of evidence available should assist in overcoming this difficulty.

Hazard Evaluation Protocols

In recent years, the number of tests designed to estimate the hazard of chemicals and other anthropogenic stresses to aquatic life have so multiplied that these tests had to be organized into orderly sequences. However, the impetus that ensured the preparation of the protocols came from an entirely different need and that was the consent decree against the Environmental Protection Agency on the "65 hazardous chemicals." An examination of the evidence used to prepare criterion documents on these chemicals showed that no systematic generation of data occurred for the purpose of preparing these documents but rather data were taken almost entirely from scholarly journals and the "gray" literature that was not peer reviewed. Both were usually prepared for more limited purposes than estimation of environmental hazard. Thus, large gaps of information and sometimes conflicting evidence surfaced because a standard minimal data base was not generated. Although quite a variety of these protocols has been prepared (Dickson, *et al.*, 1979), no general agreement exists as to the methods that should be included or the sequencing of these methods. In 1973, Cairns and Dickson prepared a protocol for testing ammunition plant wastes that was subsequently published in an ASTM journal (Cairns and Dickson, 1978). This publication really had a laboratory protocol starting with the simplest laboratory toxicity test and proceeding to the most elaborate, plus a field protocol with the same gradient. Subsequently, Cairns (1981) decided that these protocols unintentionally followed the evolutionary development of laboratory and field testing with the most familiar tests done first and the least familiar last. A few years later, a more complete case was made for testing different levels of biological organization simultaneously (Cairns, 1983). The argument about protocol structure and content is still continuing and is unlikely to be resolved quickly; it deserves serious attention. The debate revolves around two major points: (a) How accurately can one predict from one part of the protocol to the other and from one level of biological organization to another? and (b) How much information redundancy occurs (i.e., how much information overlap) when one carries out a large array of tests? Thus, the data needed to resolve the issue about protocol structure and content are quite clear, but the amount of effort required to get the necessary information will be enormous.

LEGISLATION

Most present legislation is designed to prevent or restrict intrusion of societal wastes into natural systems. When legislation does focus on environmental quality directly, it is in such a general way as to be virtually meaningless. Most recently, much has been heard about "fishable, swimmable waters." Such statements avoid being explicit regarding the qualities necessary to protect ecosystem health and do not state whether one should be satisfied with catching an occasional carp (which may or may not have abnormal growths) or whether a quality sport fishery is intended and, if so, how one determines this. Legislation is needed that will facilitate local and regional decision making on criteria and standards but which will take over when local stewardship fails. Criteria for systems of national importance (e.g., such as those containing rare and endangered species, national parks and forests, and the like) might have guidelines provided by a federal agency and a management team representing the federal government. Additionally, certain types of environmental effects (e.g., acid rain) will require negotiation between nations such as Canada and the United States and ultimately should be regulated by some form of world government. Details for implementing this are beyond my competence, for I have no legal training, and beyond the scope of this discussion even if I did. However, it is clear that quality control and other management practices must be based on direct evidence from the system being protected. In some cases, the system will extend over national political boundaries.

Who Pays for These Quality Control Systems?

Not too many years ago, we were accustomed to thinking of water, air, and the environment in general as a "free good" or a "common ground." Common ground is, of course, something that is not owned by a particular person, such as the envelope of air surrounding the earth, and is, therefore, open to use by all. Only relatively recently has some thought been given to protecting the rights of others using the common ground and even the rights of the non-human biota occupying it. The idea of a use tax is resisted vigorously, partly because the determination of charges will not be easy and because people resist paying for something that once was free. Nevertheless, someone will have to pay for the management system protecting environmental quality, and what better way to do so than to apportion the charges for this among all users, including individuals who drive automobiles, heat their homes, etc. These are small but collectively important mobile and stationary emission sources. Fortunately for us, the acid rain problems in Europe will probably force some action there from which we can learn and devise our own methods. While all of these actions may seem difficult or impossible, the Arab oil embargo forced a number of energy-saving measures, particularly in industry, that were retained even when the cost of oil dropped. "Unthinkable" measures become "thinkable" when the incentives are sufficiently powerful.

Developing Quality Control Criteria

The restoration of the Thames and Clyde Rivers in the United Kingdom, Lake Washington in the United States, and a number of other aquatic ecosystems elsewhere in the world demonstrates that present methodology and technology properly used can work wonders. Some effects (e.g., egg shell thinning in birds caused by DDT) are undoubtedly beyond our present and near future predictive capabilities and will have to be detected by biological monitoring of natural systems. Even the crude monitoring systems in place a few years ago detected the egg shell thinning, and corrective measures reduced the magnitude of the problem in a few years. Almost certainly as our monitoring and predictive capabilities improve, the number of such incidents will be markedly reduced and those that occur will be detected quickly. Detection of more subtle effects, such as minor changes in energy flow and detritus processing in natural systems, will require years to perfect.

THE NEED FOR A WORLD-WIDE STANDARD TOXICITY TEST

Most of the disagreement about the effects of various kinds of toxicants is the result of using different species under different test conditions and different end points. Additionally, some countries have less information about the effects of potentially toxic materials than others, and some have virtually none at all. These differences make general use difficult. The question of transferability of results has also been hampered by differences in climatic conditions of ecosystems and the like. An ideal standard test for world-wide use should have the following characteristics: (a) uses organisms with a cosmopolitan distribution; (b) generates quantitative information that is easily communicated; (c) uses inexpensive apparatus that is easily obtainable; and (d) provides information about acute and chronic effects in less than one month. One might also add the following qualifications: (a) that the end points or parameters be accepted by professionals as important or significant; (b) that laboratory results be readily transferable to the field (i.e., high probability of field validation of predictions based on laboratory results); (c) that replicability be quite high and response variability precisely documented or low; (d) that the organisms used be ecologically important or significant; and (e) that the no-observable-effects level (i.e., the concentration at which no adverse effects are observed) be less than, equal to, or not greatly above commercially important species, such as fish, shell fish, etc. A candidate test uses protozoans, a group of organisms with a cosmopolitan distribution. The scientific understanding for the end points used has appeared in scholarly journals for a long time (Cairns, 1969, 1982b). The use for toxicity testing has also passed peer review in scholarly journals (Cairns, 1980; Niederlehner, *et al.*, 1985), and field testing has been extensive (Cairns, *et al.*, 1979; Henebry and Cairns, 1984; Plafkin, *et al.*, 1980). In addition to testing in the United States, Bermuda, and the Bahamas, Dr. Shen Yun-fen of the Institute of Hydrobiology, Academia Sinica, Wuhan, People's Republic of China and her colleagues have been testing the method for approximately 2½ years with considerable success. For purposes of calibration, Dr. Shen spent approximately a half-year at the University Center for Environmental Studies, Virginia Polytechnic Institute and State University, carrying out some collaborative research. The method using protozoans has also been field tested in Antarctica (Cathey, *et al.*, 1982) and has proven successful. Additional evidence indicates that the method will work well in Australia, the Amazon River, the United Kingdom, and Europe. Given the variety of conditions already tested, it seems likely to be as useful throughout the world as any other method. Optimal use would require that identifications be made by a trained taxonomist. Our studies had good results using students with no prior taxonomic experience but who were good at discriminating differences in shape, form, and pattern. In terms of the reliability of the information for toxicity testing purposes, there was no statistically significant difference between a newly trained person and a skilled taxonomist. This is not to say that the skilled taxonomist did not provide additional information but, rather, that the information was generally not used and the primary information could be obtained by a less skilled person. Professional organizations could provide an enormous service by developing a world-wide information pool on pollutional and environmental measurements and by providing a widely used standard test that could be a reference for translating data obtained from indigenous species with limited distribution to those with cosmopolitan distribution. Equally important, a standard test would provide some means of plotting environmental water quality on a world-wide basis.

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The Case for Simultaneous Toxicity Testing at Different Levels of Biological Organization

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ABSTRACT: In the past few years, there has been widespread acceptance of the idea that a scientifically justifiable estimate of the hazard of a chemical to aquatic life can be obtained in a cost-effective way from data systematically generated for that purpose. One cannot assume that data generated for research purposes other than hazard evaluation will be adequate to meet this need, since such data have been demonstrably inadequate for producing criterion documents for the 65 chemicals named in the consent decree imposed on the U.S. Environmental Protection Agency (EPA) to stimulate the production of these documents. Since the EPA has had insufficient time to generate a substantial body of its own evidence, data were obtained from the open literature. Although the evidence was, in most cases, exemplary in terms of the original purpose for which it was intended, it was inadequate in the aggregate for the purpose of hazard evaluation. The Toxic Substances Control Act provided the primary impetus for the development of hazard evaluation protocols that require the systematic generation of data. Although these were designed primarily for new chemicals, the need to use them for some existing chemicals has been recognized.

Although the structure and organization of hazard evaluation or toxicity testing protocols are quite varied, the most familiar ones espouse sequential testing. That is, simple inexpensive range-finding tests involving single species are used at the outset, and one proceeds through tiers or phases in which the tests increase in complexity, sophistication, cost, and, frequently, duration. It is generally recognized that the amount of evidence needed to make a sound estimate of hazard will differ from chemical to chemical, and, therefore, provision has been made for terminating testing at multiple points in the sequence. One expects that only a few of the more dangerous or persistent chemicals will require every test in the series, since the large volume of new chemicals being generated and the scarce resources for testing now prohibit a complete series of tests on all chemicals.

Justification for sequential testing is as follows:

1. More expensive, sophisticated tests can be carried out more efficiently if the results from simpler toxicity tests are available when the complex tests are designed.
2. Carrying out tests in sequence is most likely to ensure that an adequate amount of

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data is made available for the estimate of hazard without markedly overshooting the point at which a sound evaluation of hazard can be made.

3. In situations in which one is examining an array of chemicals that are all intended for the same purpose (to select the one with the least environmental impact), sequential testing is likely to identify the most suitable candidate chemical at the least cost.

A strong case can also be made for simultaneous testing at different levels of biological organization:

1. No compelling evidence exists that single-species tests can be used to predict multispecies, community, or ecosystem responses accurately. Additionally, the ability to predict sublethal effects, such as altered behavior, growth, reproductive success, and the like, from short- or long-term tests on lethality has not been exemplary. If such a prediction were adequate, the need for additional tests beyond the range-finding test would not be so crucial. Since the information from toxicity tests at the beginning of the sequence is not demonstrably correlated with responses in the latter parts of the sequence, the ability to use information from the first part of the sequence in the design of subsequent toxicity tests is questionable.

2. If the time involved in collecting sufficient information to make an adequate estimate of hazard is important (because of the financial outlay in developing a new chemical and the like), delay in reaching a critical mass of information for a sound decision is a major cost factor. Even if money were saved in the sequential testing procedure, this might be overbalanced by the amount lost elsewhere as a consequence of the additional delay. This is particularly true in view of the fact that short-term tests are carried out early in the sequence and longer-term tests are postponed until the short-term tests are completed.

3. The sequential arrangement of tests from the simple to the more complex possibly reflects, in a general way, the historic development of the field. Therefore, toxicity tests—with which there is a long familiarity—are placed early in the sequence and more recent and more sophisticated tests, which are still in the experimental stage of development, are placed last. Because our awareness of the need for additional information has evolved in this fashion (that is, from simple, short-term crude tests to more sophisticated tests), it does not mean that this evolution of thinking has to be repeated in the hazard evaluation process.

The belief that multispecies, community, and ecosystem toxicity tests are second-order tests that can only be carried out after single-species tests is an assumption that deserves serious attention. There is no compelling evidence that single-species tests can be used to predict reliable responses at more complex levels of organization. Development of suitable tests at higher levels of organization than single species may well have been impaired because of the views just discussed, since both funding and research priorities almost certainly have been influenced by them. Although tests at higher levels of organization may be expensive, many are less expensive than or comparable in cost to long-term, continuous-flow exposure tests of single species. If toxicity testing at different levels of organization is to be simultaneous instead of sequential, much more attention needs to be given to increasing the array of suitable test methods for multispecies testing than has been the case in the past.

KEY WORDS: toxicity testing protocols, hazard evaluation, multispecies toxicity tests, ecosystem toxicity tests, aquatic toxicology, hazard assessment

Single-species tests have been the workhorse of the toxicity testing field for many years. There are both practical and emotional reasons for this. Single-species tests are essential for obtaining information on concentrations and durations of exposures to chemicals that produce changes in survival, production, physiology, biochemistry, and behavior of individuals within a particular species. Single-species tests range from those measuring acute effects, where the major concern has been rapid mortality, to highly sophisticated tests of chronic effects. The types of observations possible in chronic toxicity tests include long-term survival rates; growth rates; changes in reproduction; pharmacokinetic responses; determination of the mechanism of toxicity; pathological, biochemical, and physiological changes; and mutagenic, teratogenic, and carcinogenic rates. In recent years, increasing emphasis in acute toxicity testing has been given to the use of life history stages [*1*][2] and more sensitive end points than lethality. Simpler tests at least are reasonably priced and deliver certain kinds of information relatively rapidly. However, as one moves to more sensitive parameters and longer periods of exposure, together with improvements in the environmental realism of a test, such as continuous flow and other more realistic conditions, the cost increases may be an order of magnitude or more.

We also tend to be somewhat emotionally attached to responses with which we identify. All the things that happen to other species in single-species tests can also happen to us. Death is an aspect we understand better than disequilibrium in an ecosystem. I have been carrying out single-species toxicity tests since 1948, am doing so now, and expect to continue to do so for the remainder of my professional career. In addition, in 1973 a colleague and I produced a protocol for laboratory toxicity testing that started with single-species tests. This was eventually published [*2*] after it had been utilized for a number of years with reasonable success.

Evidence examined since the first protocols were produced and tested has convinced me that sequential testing proceeding from a single-species test to higher levels of organization is not the best strategy and that simultaneous testing at different levels of biological organization is more scientifically justifiable and even sounder economically. I am not against sequential testing *per se*, but I am against sequential testing that proceeds from lower to higher levels of biological organization (for example, from single species to populations, communities, and ecosystems). In short, the most generally accepted assumption is that simple inexpensive range-finding tests involving single species should be used only at the outset of the sequence and that one should then proceed through tiers or phases in which the tests increase in complexity (that is, higher levels of biological organization), sophistication, cost, and, frequently, duration. Sequential testing is essential if one assumes that the amount of evidence needed to make a sound estimate of hazard will differ from chemical to chemical, and, therefore, one should make provisions for terminating testing at multiple points in a sequence. One would expect that only a few of the more dangerous or persistent chemicals would require every test in the series, since a large number of new chemicals appear annually [*3*], and the scarce resources for testing now prohibit a complete series of tests on all chemi-

[2] The italic numbers in brackets refer to the list of references appended to this paper.

cals. I still support this assumption and believe it is both scientifically and economically defensible. *I only question the assumption that single-species tests should be the only ones carried out in the first tier or phase of the sequence*. Perhaps the most expeditious way to explore my change in position is to start with the assumptions which originally led me to believe that laboratory toxicity testing should start with single-species tests:

1. More expensive, sophisticated tests can be carried out more efficiently if the results from simpler toxicity tests are available when the complex tests are designed.

This statement assumes that substantive predictive value can be obtained from short-term to chronic tests with single species and applied to higher levels of biological organization. MacArthur [*4*] has aptly diagnosed a basic problem: "Scientists are perennially aware that it is best not to trust theory until it is confirmed by evidence. It is equally true, as Eddington pointed out, that it's best not to put too much faith in facts until they have been confirmed by theory." The facts that we have been gathering are the various responses of single species to various chemicals under reasonably well controlled conditions. The assumption is that we can use these facts to predict "safe" concentrations with low probable risks to communities and ecosystems using single-species tests. I know of no substantial body of scientifically justifiable evidence that supports this hypothesis. Nevertheless, although this hypothesis is rarely so explicitly stated, it is an underlying assumption in much of our regulatory and enforcement practices on toxic substances and other environmental pollutants and is also responsible for directing research funds in pollution assessment predominantly toward single-species tests.

This assumption is worthy of detailed examination because of its pervasiveness in water pollution assessment. Failure to state this assumption explicitly may be due to a general feeling that it is so well supported by evidence that the statement is platitudinous, or to a fear of causing a loss of confidence in the few biological tests now generally accepted. If the former is the reason, there certainly has been no rigorous gathering of evidence or analyses of this evidence to check the assumption, and, if the latter, we should be comforted by the fact that single-species toxicity testing has an irreplaceable role in water pollution assessment. Perhaps, however, it is time we recognize that single-species tests will be of even greater value if used in combination with tests that can provide data on population interactions and ecosystem processes.

Taub [*5*] has given an example of predator-prey interaction that illustrates the inadequacy of single-species tests for predicting the outcome of multispecies interactions. If a chemical introduced into the natural environment reduces the growth rate of a prey population so that both birth and death rates are effectively lowered by the establishment of an older age structure, the relative population size could remain the same, but the flow of biomass available in the system might be reduced. A predator population that relies on this particular flow of biomass could then lose a substantial source of food. If that source were critical to the survival of the predator (that is, no other suitable sources were available), the population could become extinct or markedly reduced, although no change would be observed in the size of the prey population. Single-species tests that use either the predator or the prey could not have been utilized to predict this outcome, although the indirect effect could have been as severe as direct chemical toxicity. A single-species laboratory test might also indicate that the effect on *that species* would be more severe than would actually occur in nature.

In the following hypothetical example, the chemical might adversely affect test Species A and produce increased mortality in a single-species laboratory test. However, in an ecosystem, the chemical could affect a predator of Species A by inhibiting its reproduction. Although the size of A was reduced in laboratory tests, reduction in predation in the natural environment might well compensate for this effect, thus resulting in a minimal change in its population size in the "real world." This effect might theoretically have been predicted accurately from individual tests on each of the species involved, but only if one had a very substantial working knowledge of the particular ecosystem in which they lived and interacted.

I will postpone for the moment a discussion of other considerations, such as whether tests that will furnish such evidence are now available or whether we can afford to carry them out if they are. At this point, it is sufficient to indicate that single-species tests are not likely to be useful predictors of effects, particularly indirect effects, at higher levels of biological organization. As a consequence, information obtained from them early in a testing sequence is not likely to make subsequent tests at higher levels of biological organization more efficient or better designed.

2. Carrying out tests in sequence is most likely to ensure that an adequate amount of data is made available for the estimate of hazard without markedly overshooting the point at which a sound evaluation of hazard can be made.

The danger of overshooting, in terms of gathering an evidence base in excess of that necessary to estimate hazard reliably, is clearly overshadowed by the possibility of gravely underestimating or overestimating hazard as a consequence of stopping at that point in the sequence where only single-species tests have been carried out. One should be concerned with both quality and quantity of data generated. If the predictive value of data at one level of organization for response at another level of biological organization is not great, then an array of tests at different levels of biological organization is essential in order to make an informed and accurate estimate of hazard. Simultaneous testing at different levels of biological organization need not preclude prudence in determining how much evidence to gather at each level. Simultaneous testing at different levels of biological organization is no more likely to overshoot the mark in terms of gathering an excess of data to make an informed estimate of hazard than single-species testing alone and is more likely to ensure that the estimate will be confirmed in the "real world."

3. In situations in which one is examining an array of chemicals that are all intended for the same purpose (to select the one with the least environmental impact), sequential testing proceeding from the lowest to the highest levels of biological organization is likely to identify the most suitable candidate chemical at the least cost.

This statement is based on the assumption which I now think to be scientifically unjustifiable—that differences in response will remain in the same relationship from one level of biological organization to another. There is little direct evidence to either confirm or invalidate this assumption, and the indirect evidence suggests that this is not likely to be true. Even within a single species, the relative sensitivities of different life history stages of a single species are not likely to remain constant from one chemical to another, particularly if these chemicals are markedly different in structure. Therefore, if different life history stages vary in response from one chemical to another, it seems highly probable that different levels of biological organization will also vary in the same way and, therefore, that the predictive value from one level of biological organization to another, even in this regard (that is, in determining the chemical with the least biological impact out of an array), will be minimal. At the very least, sufficient evidence to make a scientifically justifiable decision is essential. Until this assumption is either verified or discredited, it would be prudent to assume that it is not valid.

One reviewer of this paper correctly noted that "toxicity is not the only mechanism of deciding whether extensive testing on a chemical should be done or not." Although this paper focuses on toxicity testing, I reaffirm a position held since 1948 that a mixture of different types of evidence including biological, chemical, and physical are needed to make a sound decision. Selection of chemicals for testing is based on considerations of use, including frequency of use, geographic distribution, and intensity of use, as well as biological considerations—a position clearly stated in *Principles for Evaluating Chemicals in the Environment* (Ref 6, Tables 10 and 11, p. 227), produced by a committee which I chaired. In addition to high or moderate toxicity to representative single species, one should consider the persistence of a chemical or its continuous introduction into the ecosystem as well as conditions of use, environmental partitioning, and the like. These aspects are fully discussed in publications in which I had a part [*6-11*]. The strategy espoused, particularly in the "Pellston series" [*7,9,10,12*], is that the amount of data gathered should be determined by the proximity of the environmental concentration of the chemical and the concentration below which no adverse biological effects are noted.

Case for Simultaneous Testing

A strong case can also be made for simultaneous testing at different levels of biological organization (that is, single species, multispecies, community, and ecosystem).

Predictive Value of Single-Species Toxicity Tests

Single-species toxicity tests can provide useful information on the concentrations and durations of exposures to chemicals that result in changes in the survival, reproduction, physiology, biochemistry, and behavior of individuals within particular species. Single-species toxicity tests can range from tests of acute effects, where the major concern has traditionally been rapid mortality, to highly sophisticated tests of long-range or chronic effects with more subtle end points. The types of observations possible in chronic toxicity tests include long-term survival rates; growth rates; changes in reproduction; pharmacokinetic responses; mechanisms of toxicity; pathological, biochemical, and physiological changes; and mutagenic, teratogenic, and carcinogenic rates.

Relatively little evidence is available to determine if the response of test species in natural situations is similar to that predicted from laboratory tests. The limited evidence that is available includes all three possibilities: (1) correspondence between the laboratory and field response is quite good (that is, the laboratory predictions were validated), (2) the response in the field is less than that predicted by laboratory tests, and (3) the field response is greater than that predicted by laboratory tests. Cairns et al [*13*] found excellent correspondence between laboratory and field responses for a sizable number of fish species when chlorine and temperature preference and avoidance of a steam electric power plant discharge were concerned. It may be that this excellent agreement between field and laboratory responses was the result of using a mobile laboratory, which was stationed on the bank of the river being studied and which used test water pumped directly from the river. In addition, since both laboratory test water quality and temperature were determined by ambient conditions in the river, a degree of environmental realism was achieved that would normally be impossible with the use of standard laboratory reference waters and test conditions. Replication of test conditions in this series of tests would be exceedingly difficult for another laboratory situated elsewhere, but replication of the test methodology would not be. In contrast, although the results of median lethal concentration (LC_{50}) tests for aquatic invertebrate species indicated low toxicity for polychlorinated biphenyls, subsequent field work and multispecies tests revealed a decrease in diversity of invertebrate populations [*14*]. Laboratory tests may also indicate that a material is much more toxic than it appears to be in the field. For example, data from laboratory tests with a standard set of aquatic invertebrates suggested that mirex was highly toxic to the test species. However, the field data failed to corroborate these findings [*15*]. Similar results obtained with subchronic tests using several aquatic invertebrate species exposed to methoxychlor provided evidence that some of the species used were affected adversely at concentrations of 0.2 μg/L [*16*]. However, a one-year field study investigating exposure in streams yielded additional information on population effects and interactive responses. Only very subtle changes were detected in individual species at 0.2

μg/L in the stream environment, and multispecies interactions, such as predator-prey relationships, appeared unaffected [17].

Although more literature exists in which laboratory and field responses of single species have been compared, it is astonishing how seldom attempts are made to corroborate the evidence gathered in laboratories from the field response of species. When this is done, the correspondence may be good, or the laboratory response may be markedly greater or less than the response under field conditions. This brief discussion was included for two purposes: (1) to show that if the predictive value from single species in the laboratory to single species in the field is somewhat questionable, at least in certain cases, one might assume it to be eminently reasonable that there would be a considerably lower predictive value from single species to multispecies and to higher levels of biological organization; and (2) to demonstrate that the predictive value of single-species tests appears to be greatest in situations in which there is a high degree of environmental realism. Improving the environmental realism of a single-species test will diminish its advantages over tests at higher levels of biological organization because it will increase the complexity of the test and, therefore, the difficulty in carrying it out. This will, in turn, markedly increase the cost. As a consequence, the primary justifications for using single-species tests to predict responses at higher levels of biological organization, namely, simplicity and low cost, are severely impaired if one incorporates sufficient environmental realism into the test to ensure excellent corroboration with the field results.

Time

In 1973, when I first considered the problem of arraying test methods to optimize the time of information generation and costs per unit of information generated, it seemed quite reasonable to place first short-term, crude single-species tests using lethality as an end point. The question I thought should be asked was "How toxic is this chemical in relation to other chemicals?" A more appropriate question now seems to be "What toxicological information will enable me to make a reliable estimate of the consequences of introducing this chemical into a complex environment?" If the latter is the most appropriate question, then the time required to generate data might well be shortened if several levels of biological organization were tested simultaneously. The question of both time and amount of data required to make an accurate estimate of hazard is influenced very strongly by the degree of predictive value of data gathered early in a sequence for results in tests further on in the sequence. One way to illustrate this point is to use a diagrammatic representation of the process.

The first diagrammatic representation of a sequential hazard assessment procedure demonstrating increasingly narrow confidence limits for estimates of the no-biological-effect concentration and the actual expected environmental concentration of a chemical is given in Fig. 1. Note that the confidence

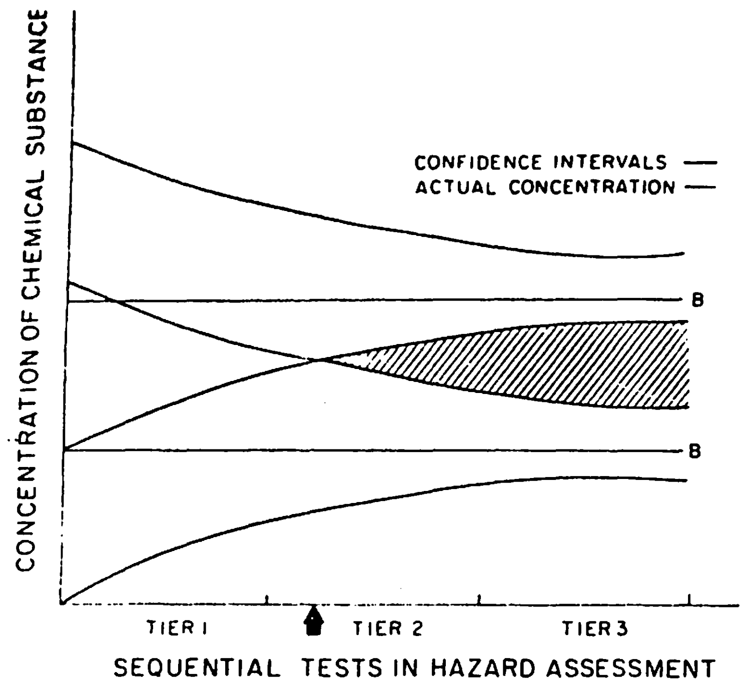


FIG. 1—Diagrammatic representation of a sequential hazard assessment procedure demonstrating increasingly narrow confidence limits for estimates of no-biological-effect concentration and the actual expected environmental concentration. Modified from Cairns et al [7] with the kind permission of the American Society for Testing and Materials.

interval boundaries depicted by dashed lines are not absolutely straight, which suggests that some tests furnish information of predictive value about the tests to follow. There is a further suggestion that the degree of predictive value decreases as one proceeds through the tiered system. It is also interesting that the confidence interval line slopes for both the no-adverse-biological-effects concentration and the environmental concentration of the chemical are displayed as being roughly identical. At the time that these were drawn at a workshop on hazard evaluation held at Pellston, MI [7], it seemed intuitively reasonable to characterize these relationships in that way. The emphasis at that workshop was on coupling environmental concentration information and biological effects information. Although the sequencing of tests was important at the early stages of thinking this problem through, predictive value from one tier to another, although recognized as an important characteristic, was not discussed in detail or with precision. Even now nearly four years after that workshop, there is no substantive body of data generated in a systematic fashion for the purpose of hazard evaluation to enable one to determine the predictive value of information generated in one tier for the type of response likely to occur in the next tier in the series.

Assuming, for the moment, that the slopes of the confidence interval lines are roughly comparable for both the no-adverse-biological-effects concentration and the environmental concentration of the chemical, one could have two other strikingly different alternatives. One of these is that there is no predictive value from one part of the tiered sequence to another or from one test to another and that each test furnishes totally different knowledge of roughly comparable quality, which then enables one to proceed incrementally toward the goal of reducing uncertainty to an acceptable level. This situation is represented diagrammatically in Fig. 2. Another alternative is that the predictive value at the early stages of the sequential or tiered testing system is extremely high and, therefore, uncertainty is reduced almost precipitously; however, after the initial tests are carried out, additional tests do little or nothing to reduce the uncertainty further. This situation is depicted diagrammatically in Fig. 3. Other alternative slopes are depicted in Figs. 4 through 7.

Of course, it seems intuitively reasonable that the slopes might not be the same for these two quite dissimilar types of information. If one assumes that the slopes might be totally different for the confidence intervals depicting the determination of the environmental concentration and the no-adverse-biological-response concentration, one could assemble a number of other alternative figures. If they are not the same, one can obtain a substantial number of other figures which I leave to the reader's imagination.

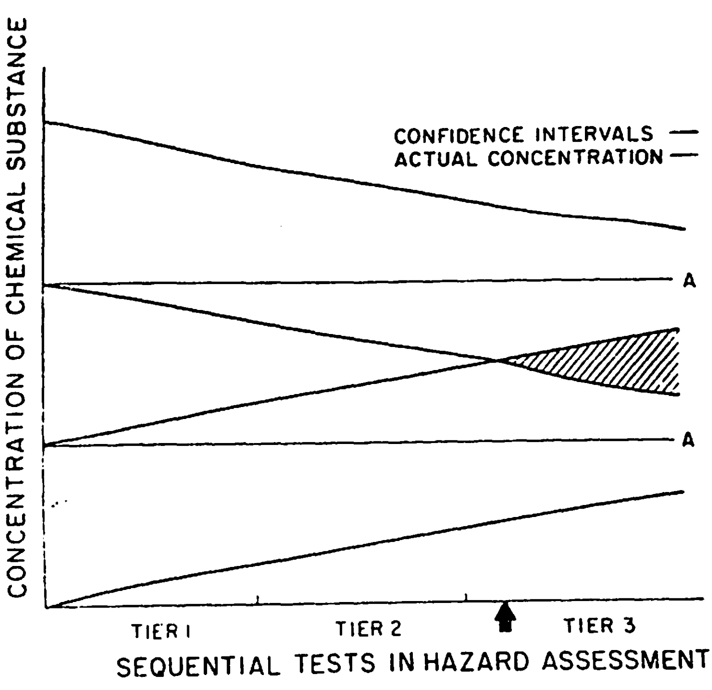


FIG. 2—*Diagrammatic representation assuming little or no predictive value for any information. As a consequence, reduction in uncertainty is uniform and incremental.*

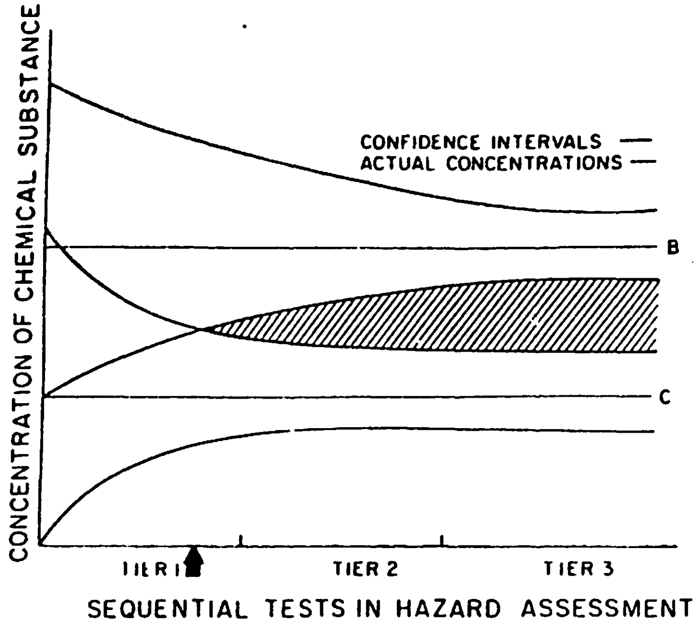


FIG. 3—*Diagrammatic representation assuming a very high predictive value of information for the early stages of the sequential hazard assessment procedure and little change in confidence thereafter because of the complex and highly variable nature of the ecosystems.*

Historic Development of the Field

The sequential arrangement of tests from the simple single species to the more complex ecosystem tests possibly reflects, in a general way, the historic development of the field. Therefore, we place toxicity tests—with which there is a long familiarity—early in the sequence and place last the more recent and more sophisticated tests which are still in the experimental stage of development. The fact that our awareness of the need for additional information has evolved in this fashion (that is, from simple, short-term crude tests to more sophisticated tests) does not mean that we should repeat this evolution of thinking in the hazard evaluation process. There are a number of alternative explanations for the arrangement just mentioned, but the striking similarity between the historic development of the field and the arrangement of some protocols deserves attention.

Discussion

One might speculate that no test is necessary at other levels of biological organization in instances in which single-species tests show that the no-adverse-biological-effects concentration is markedly above the estimated environmental concentration of a chemical. However, it is quite possible for pre-

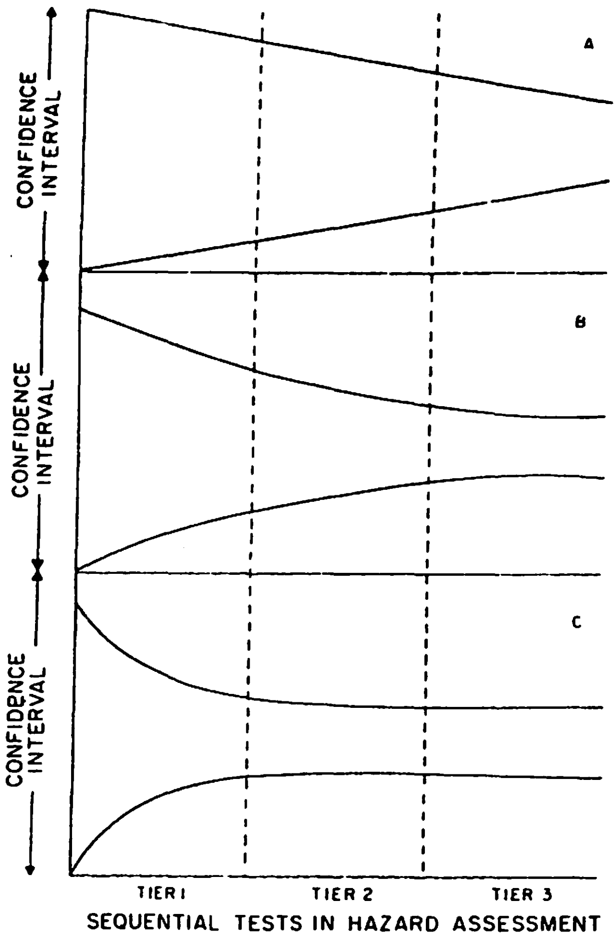


FIG. 4—*Comparison of alternative possibilities for biological evidence:* (A) *the information has low predictive value—gradual reduction of uncertainty as one proceeds through Tiers 1 through 3;* (B) *information has some predictive value—more rapid reduction of uncertainty;* (C) *high predictive value—rapid reduction of uncertainty.*

liminary estimates of either concentration to be markedly different from the real-world concentration. Therefore, situations in which preliminary results indicate that the no-adverse-biological-effects concentration is far above the estimated environmental concentration may show, on more detailed examination, that the two concentrations are quite close together. Conversely, it is also possible that the preliminary results will indicate the two concentrations are quite close and subsequent examination will show that they are very far apart, with the no-adverse-biological-effects concentration being well above the estimated environmental concentration. Therefore, those who advocate use of single-species tests alone in situations in which there appears to be no danger

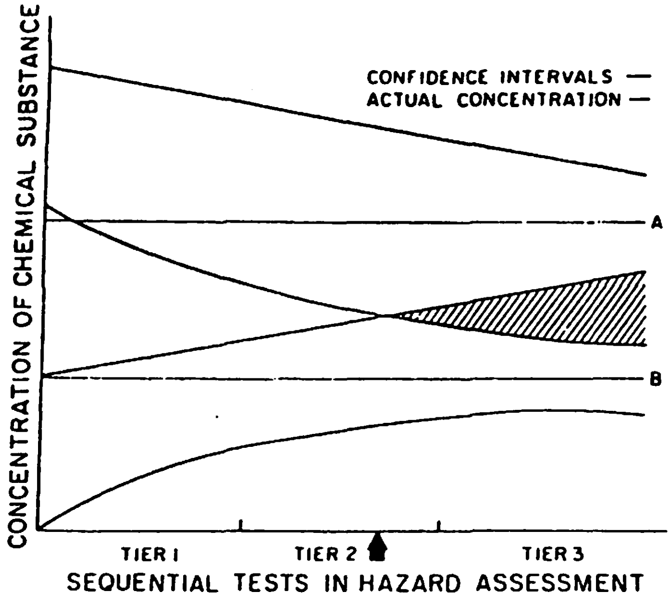
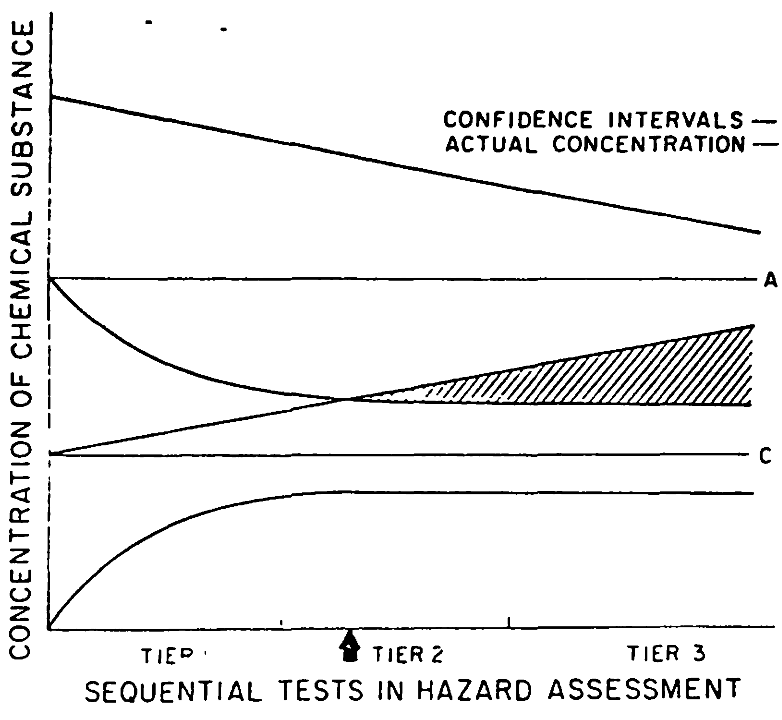


FIG. 5—*Comparison of alternative possibilities: the biological evidence has low predicitive value* (A)*: the environmental concentration of chemical evidence has moderate predictive value* (B).



. 6—*Comparison of alternative possibilities: the biological evidence has low predictive* A)*: the environmental concentration of chemical value has high predictive value* (C).

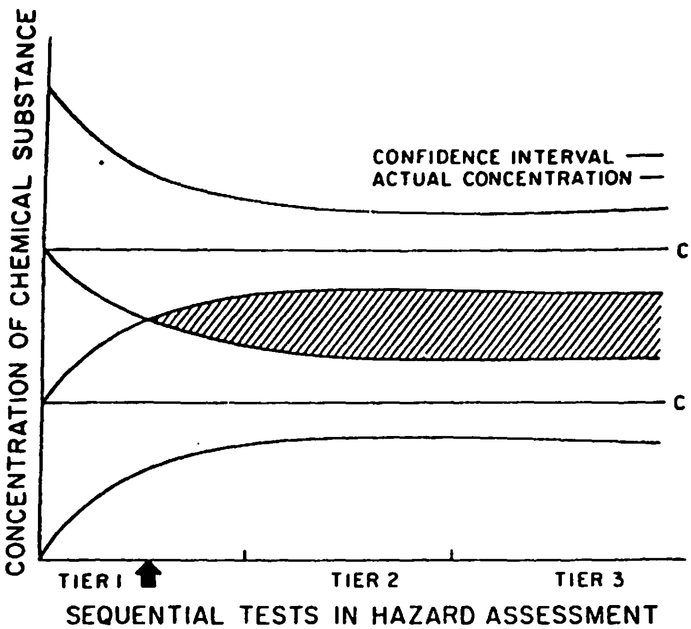


FIG. 7—*Comparison of alternative possibilities: both types of evidence have high predictive value* (C).

because the two concentrations are well apart and the no-adverse-biological-effects concentration is well above the estimated environmental concentration are using scientific judgment to obviate any additional tests. Since scientific judgment is required at almost every step of the hazard evaluation process (including which tests to use, how to sequence the tests, the degree to which extrapolations can be made from these tests to responses not included in the test series, the biological and ecological significance of the responses being measured, and the like), there is no reason why judgment should not be exercised at the very outset. However, it seemed useful to remind practitioners that apparent relationships based on preliminary information may, in fact, be quite misleading.

In terms of sequencing, if the slopes of the lines are as depicted in Fig. 2, the type of sequencing used would not seem to be particularly important because all the types of information generated would be increasing one's confidence equally in the estimate of the no-adverse-biological-effects concentration. The assumption that all types of biological information generated are of equal value seems intuitively unreasonable. If the slopes in Fig. 3 are correct, only the early parts of the sequencing are of any importance because not much new information is added after the initial test series. This seems to be, if one judges actions as a basis for determining how people feel about these three sets of figures, the assumption that best depicts our current stance because we base most of our decisions on a relatively few tests. Whether this is for economic or scientific reasons is not clear.

The most important question now facing those preparing protocols is which form of sequencing to use. It is clear that all tests do not provide information of comparable value and, therefore, it seems reasonable to array tests so that the most useful information in terms of cost, time required, and (of course, most important) scientific merit is generated first. One of the important criteria in determining scientific merit should be the ability to predict responses in succeeding tests in the sequential protocol. Many protocols start with a single-species toxicity test and include no other levels of biological organization in the early stages in the sequence. This is done despite the lack of substantive evidence that one can accurately predict the response at higher levels of biological organization from the single-species tests and also rather general agreement among ecologists that such predictions from one level of biological organization to a higher level of biological organization are not scientifically justifiable.

Although it is seldom explicitly stated, single-species tests are usually carried out with the assumption that the results are useful in protecting ecosystems. Although there is very little direct scientifically justifiable evidence to validate or negate this assumption, what evidence is available suggests that sometimes the evidence derived from single-species tests alone can be either underprotective or overprotective of ecosystems. The same thing is true for predictions of higher levels of biological organization, such as multispecies and communities. Therefore, the practice of sequential testing starting from single species and progressing toward ecosystems appears scientifically unjustifiable. Protocols designed for estimating hazard should almost certainly require simultaneous testing at the outset of several different levels of biological organization. The facts that single-species tests are more widely used and generally accepted and that simple, inexpensive screening tests at higher levels of biological organization are infrequent and sometimes costly are probably more a function of perceived need than a function of the difficulty of carrying out such tests. There are multispecies and community-level tests now in the literature that can be carried out with equipment no more complex than that now used for single-species tests at costs that are not very different [*18,19*]. My recommendation in this paper is that the matter is too important to be neglected and that the predictive value of tests at one level of biological organization for responses at other levels needs to be determined in a scientifically justifiable way. One of the best means of doing this is to develop some testing protocols that require simultaneous testing at different levels of biological organization. The efficacy of these, using all the standard criteria for judgment, such as information content, cost, and time required for information generation, can be used to determine whether the present practice of starting almost entirely at one level of biological organization (the single-species test) is preferable to starting protocols with tests at several levels of biological organization.

This is too important a question to leave to anecdotal evidence or absence of evidence or anything other than a rigorous scientific examination of the question. We will be doing a disservice to the field of toxicity testing if substantive definitive evidence is not obtained on this question.

Acknowledgments

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REGULATING HAZARDOUS CHEMICALS IN AQUATIC ENVIRONMENTS

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> "It is the mark of an instructed mind to rest satisfied with the degree of precision which the nature of the subject permits and not to seek an exactness where only an approximation of the truth is possible." Aristotle

I. Introduction

Dr. Joshua Lederberg, president of Rockefeller University and Nobel Laureate scientist, addressed the urgent need for new approaches to testing toxic chemicals in a speech made in February, 1981, at the World Environmental Center.[1] He felt that finding ways to assess and control the environmental health risks posed by toxic substances is one of society's major scientific challenges. Serious questions about the efficacy of both our scientific and regulatory approaches exist; consequently, the quote from Aristotle is particularly appropriate in this context because we must now make regulatory decisions with an inadequate scientific base. The economic benefits of producing a new chemical or technology (e.g., a power plant) may be quite clear, but the indirect costs, in terms of hazard to human health and the environment, are not.

Most of the earlier regulatory standards for discharge of potentially hazardous chemicals into the environment allowed fixed concentrations that were not to be exceeded. This strategy proved inappropriate for several reasons. (1) Some chemicals produce adverse biological effects at concentrations below present analytical

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1. Anon, *Improve Toxic Testing Nobel Scientist Urges*, in CHEMECOLOGY (1981).

capabilities. (2) Environmental quality parameters, such as water hardness, temperature, pH, and dissolved oxygen concentration, mediate the toxic response—the same concentration of zinc would produce a different toxicological response in the hard water of the Guadaloupe River in Texas than in the soft water of the Savannah River between Georgia and South Carolina. (3) Toxic chemicals may act differently in combination than they do individually.

Unfortunately, there is no instrument devised by man that will measure toxicity. Only living material can be used for this purpose. This immediately produces both scientific and regulatory difficulties because living material is complex, regionally differentiated, often highly variable, and may act differently in laboratory test containers than in natural systems. This paper examines current regulatory and scientific approaches to the presence of hazardous substances in an aquatic environment. Implementation of a specific hazard evaluation process is recommended to ameliorate the inadequacies of present approaches.

II. Regulating Toxic Substances

Although most toxic substance regulations are designed to protect human health and the environment against deleterious concentrations of chemicals, they differ strikingly in protection strategy, statement of goals, allocation of costs, and responsibility for generating appropriate data and means of implementation. Unfortunately, the scientific underpinnings for almost all regulatory objectives are inadequate. Although concern about this problem had been growing for years, it was probably first crystalized by the water quality criterion documents for sixty-five classes of pollutants that were designated as toxic in section 307(a)(1) of the Clean Water Act of 1972[2] and next by the premanufacture testing policy enunciated under section 5 of the Toxic Substances Control Act of 1976[3] (TSCA). The scientific problems inherent in toxic substance regulations were matters of concern and discussions.[4]

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2. Originally Federal Water Pollution Control Act of 1972 as amended in 1977, the Clean Water Act, 33 U.S.C. §§ 1251-1376 (1976 & Supp. IV 1980).
3. 15 U.S.C. §§ 2601-2629 (1976 & Supp. IV 1980).
4. Cairns, Jr. & Maki, *Hazard Analysis in Toxic Materials Evaluation*, 51(4) J. Water Poll. Control Fed. 666-71 (1979); Deland, *EPA "Policy" for Testing Toxics*, 15(4) Envt'l Science & Tech. 385 (1981); Schaeffer, Park, Kerster, & Janardan, *Sampling and the Regulatory Maze in the United States*, 4(6) Envt'l Mgt. 469-81 (1980); Christman, *Clear Water Goals*, 15(3) Envt'l Science & Tech. 283 (1981).

TSCA gives the Environmental Protection Agency (EPA) control over the manufacture of chemicals that may or may not prove to be toxic well before they are likely to enter the environment; TSCA thereby differs from earlier legislation such as FIFRA,[5] which regulates substances that were designed to be toxic, and the Clean Water Act, which regulates the discharge of toxics into the environment. The administrator of the EPA has the authority under TSCA to prohibit or restrict the use of any chemical that may present an unreasonable risk to human health and/or the environment. In Section 2(b) of TSCA, Congress places responsibility for providing scientifically justifiable evidence of the probability of harm to organisms on the producers of these chemicals. If the evidence presented is inadequate, the EPA has the authority to require additional toxicity testing. Congress also indicated that the EPA must use its regulatory authority "in such a manner as not to impede unduly or create unnecessary economic barriers to technological innovation while fulfilling the primary purpose of this Act to assure that such innovation and commerce in such chemical substances and mixtures do not present an unreasonable risk of injury, to health or the environment"[6] (Section 2(b)TSCA).

Within this context it is important to define terms such as "risk" and "concentration."

> *Risk* is the probability of harm from an actual or predicted concentration of a chemical in the environment. *Safe concentrations* are those for which the risk is acceptable to society. As a consequence, the assessment of hazard requires both a scientific judgment based on evidence and a value judgment of society and/or its representatives. Evidence for a scientific judgment must cover (a) toxicity—the inherent property of the chemical that will produce harmful effects to an organism (or community) after exposure of a particular duration at a specific concentration, and (b) *environmental concentration*—those actual or predicted concentrations resulting from all point and nonpoint sources as modified by the biological, chemical, and physical processes acting on the chemical or its byproducts in the environment.[7]

The balancing of risk and benefit in environmental law was addressed relatively recently by Ricci et al.,[8] who emphasized that con-

5. Federal Insecticide, Fungicide and Rodenticide Act, as amended by the Federal Environmental Pesticide Control Act of 1972, 7 U.S.C., § 135 (1972 & Supp. IV 1980).

6. Toxic Substances Control Act of 1976, 15 U.S.C. §§ 2601-2629 (1982).

7. Cairns, Jr., *Estimating Hazard*, 30 (2) BIOSCIENCE 101-07 (1980).

8. Ricci & Moltan, *Risk and Benefit in Environmental Law*, 214 (4525) SCIENCE 1096-1100 (1981).

sideration of the technical complexity of the assessment of health risks leads to understandable judicial caution about interposing legal judgments on these unresolved scientific issues.

The determination of adequacy of scientific evidence for estimating hazard is a difficult problem. Evidence indicates that commonly used toxicity tests do not provide adequate data for estimating hazard to the environment. For the moment, consider a situation in which an industry feels it has provided scientifically adequate evidence and the EPA does not. Section 5 of TSCA does not require that particular environmental tests be documented on all new chemical substances before submission of premanufacture notices.[9] A recommended data base is set, for which the ecotoxicity data are based entirely on short-term single species laboratory toxicity tests. These ecotoxicity data are not mandatory but are for "guidance." Yet when industry uses a multispecies laboratory toxicity test or actual field evidence and the EPA disapproves, significant conflict results. Estimation of hazard to man and his environment is clearly a highly technical question which the courts alone are not qualified to decide, and there is no impartial "science court" of highly qualified experts specially charged with this responsibility (although the National Academy of Sciences might serve in this capacity). Since ecotoxicology is a very new and rapidly developing field, only a well qualified expert will have the necessary background to judge the scientific validity of the evidence provided. Since, at the very least, toxicological information must be coupled with information about the environmental fate and partitioning of chemicals and both assessed for statistical reliability, a panel of experts will be needed.

Biological evidence is essential to estimate hazard to the environment. Alternative ways of abating pollution have been tried, however, reliance solely on chemical/physical measurements was not scientifically justifiable for the reasons already mentioned. Technology-based standards, such as Best Applicable Technology (BAT) and Best Practicable Technology (BPT), were also employed, based on the assumption that the "practical approach" was the best way to abate pollution.[10] Problems occur with this approach.[11] From

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9. *See* 15 U.S.C. §§ 2601-2629 (1976 & Supp. IV 1980); 44 Fed. Reg. 8986 (1981).

10. *See* 33 U.S.C. §§ 301-304 (1976 & Supp. IV 1980).

11. Cairns, Jr., *Comment on "Desirable Characteristics of Environmental Quality Standards and General Considerations Involved in Their Development,"* in PROCEEDINGS OF THE SEMINAR ON DEVELOPMENT AND ASSESSMENT OF ENVIRONMENTAL STANDARDS (1983).

an ecological standpoint, technology-based standards ignore: (1) the size of the receiving system; (2) the well-established fact that environmental quality (e.g., pH, water hardness, temperature, etc.) may markedly influence toxicity; and (3) that the total impact of all stresses on natural systems must be considered, not just the impact of a single discharge. From an industrial standpoint, use of BAT and BPT may be unsuitable because: (1) a small industry on a large river may be forced to spend money on technological improvements which produce no demonstrable biological or ecological benefits; (2) long-range financial planning is difficult when the rate of technological development is difficult to predict (but would undoubtedly accelerate if this law were enforced); and (3) operators with new equipment that they cannot use properly may produce poorer quality effluents than they would with old equipment they understand. Biological evidence must be combined with chemical/physical measurements to produce an effective hazard evaluation process.

A. Biological Evidence

Since the primary objective of environmental legislation is to prevent harm to the biota (including humans), the most reliable estimates of hazard should be based on direct measurements of living organisms rather than indirect chemical/physical measurements from which biotic condition is inferred. As previously mentioned, no instrument will measure toxicity—this can only be done with living material. Yet, without chemical/physical data, determining what caused the biological response in the living material is difficult or impossible. Therefore, a scientifically justifiable estimate of hazard requires a mixture of biological/chemical/physical data.

The intent of environmental regulation is to prevent harm to the environment rather than to document the cause and extent of damage after an ecological perturbation (although this is undeniably important). Predictive tests carried out in surrogates of natural systems are essential to accomplish this purpose. In designing such test systems, a conflict or tension exists between the desire for *environmental realism* that incorporates both the complexity and variability of natural systems and the need for *replication* (ability to reproduce results) that is most easily achieved in simple systems with only one variable. This tension is presently relieved by providing four steps in the hazard evaluation process: (1) screening tests; (2) predictive tests; (3) confirmative tests; and (4) monitoring. Screening toxicity tests are designed to determine quickly, inexpensively, and simply whether or not a chemical substance is very toxic or less so relative to other chemicals. The predictive tests are generally more sophisticated laboratory toxicity tests also normally carried out with single species. There is considerable concern that single species toxicity tests cannot be used to accurately predict responses at higher levels of biological organization.[12] The basis for this concern is that new important properties are evident at higher levels that cannot be studied at lower levels of biological organization (cell-tissue-individual-population-multispecies-community-ecosystem). This is merely a restatement of the old phrase "the whole is more than the sum of its parts." Because most estimates of hazard are based on single species laboratory tests (screening or predictive tests) lacking in environmental realism, confirmative tests are recommended as well.[13] To ensure that potentially dangerous situations do not go undetected, surveillance must be carried out with a variety of methods.[14] These various biological tests provide the necessary basis for any effective hazard evaluation process.

B. *Implementation*

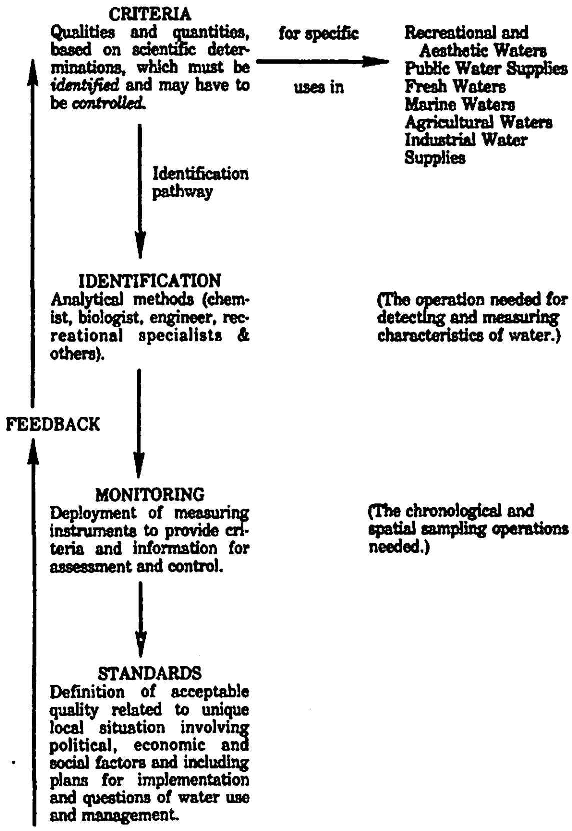
To implement a hazard evaluation process one needs: (1) professionally endorsed parameters representing key responses; (2) formal identification of the methods most suitable to measure these parameters; and (3) certification of either individuals or laboratories capable of making the measurements accurately. Unfortunately, although the use of biological responses to predict and assess pollution is both scientifically justifiable and plausible to the layman, the means of implementing fully this course of action are not in place. Of course, both scientists and laymen agree that fish should not die. But they may not agree on other desirable parameters such as the ability of fish to spawn.[15] *Detroit Free Press* staff writer Thomas BeVier quotes William Gregory, President of Edison Sault Electric Co., as saying that trying to establish spawning beds is impractical. "We can plant fish instead and all have a damn good time."[16] Conversion

12. Cairns, Jr., *Guest Editorial: Beyond Single Species Testing*, 4 MARINE ENVT'L RES. (1980). NAT. RES. COUN., TESTING FOR EFFECTS OF CHEMICALS ON ECOSYSTEMS xii (1981).
13. Kimerle, *Aquatic Hazard Assessment—Concepts and Application*, Workshop On Hazard Assessment, Int'l Jt. Comm'n 221-230 (1979).
14. Cairns, Jr. & van der Schalie, *Biological Monitoring, Part I: Early Warning Systems*, 14 WATER RESEARCH 1179-96 (1980).
15. BeVier, *It's Fish vs. Electricity on the St. Mary's River*, Detroit Free Press, Aug. 1, 1982, at 1 col. 2.
16. *Id.*

of scientifically justifiable criteria into legal standards should consider a number of non-scientific parameters (Figure 1).[17] Even if there were general agreement on the scientific component, there would be disagreement on these. Yet, even among scientists there is no widespread strong endorsement of a multispecies, community, or ecosystem parameter to assess pollution[18] or of underlying ecological principles.[19] While statistically sound ecological comparisons by respected ecologists do exist,[20] it is unlikely that community and/or ecosystem parameters will be frequently used by regulatory agencies and industry until they acquire formal professional endorsement.

FIGURE 1

Conceptual framework for developing standards from criteria.



17. NAT. ACAD. OF SCIENCES, WATER QUALITY CRITERIA OF 1972 (1973).

18. Cook, *Quest for an Index of Community Structure Sensitive to Water Pollution*, 11 ENVT'L POLL. 269-88 (1976).

19. Gilbert, *The Equilibrium Theory of Island Biogeography: Fact or Fiction?* 7 J. BIOGEOGRAPHY 209-35 (1980).

20. Green, *Multivariate Approaches in Ecology: The Assessment of Ecological Similarity*, 11 ANN. OF REVISED ECOLOGY AND SYSTEMATICS 1-14 (1979).

Although it would be best to endorse parameters and methods to measure them separately, formal endorsement of a standard method is an indirect endorsement of the parameter it measures as well. This may be accomplished by consensus developed through publications such as *Standard Methods for the Examination of Water and Wastewater*[21] or by a group of experts representing a group of professional societies such as the American Society for Testing and Materials, Philadelphia, Pa.. The standard methods formally endorsed in this way have so far been limited to single species toxicity tests. At the 1981 annual meeting of The Society for Environmental Toxicology and Chemistry, it was asked of the plenary session attendees (about 600) if anyone knew of a standard method for toxicity testing at a higher level of biological organization than single species—there was no response. Although directly assessing the health of the biota in a "receiving system" (the one into which wastes are discharged or other anthropogenic stresses occur) is the most plausible approach for preserving environmental quality, and although methods have been available for years to study a variety of environmental parameters, biologists have to date formally endorsed only parameters and methods for single species toxicity tests. The report by the Committee to Review Methods for Ecotoxicology of the National Research Council, the operating arm of the National Academy of Sciences and the National Academy of Engineering, clearly stated: "Single species tests, if appropriately conducted, have a place in evaluating a number of phenomena affecting an ecosystem. However, they would be of greatest value if used in combination with tests that can provide data on population interactions and ecosystem processes."[22] In short, the legislation is in place in TSCA and the scholarly journals have contained a significant number of methods for years, adding to them at an impressive rate. Nevertheless, professional ecologists have not formally endorsed either parameters or methods particularly suited for hazard evaluation and pollution abatement.

In addition to endorsed parameters and measurement methods, professional certification is a necessary element to an adequate hazard evaluation process. A number of societies now have some form of professional certification (e.g., The American Fisheries

21. AM. PUB. HEALTH ASS'N, AM. WATER WORKS ASS'N & AM. FED. OF WATER POLL., STANDARD METHODS FOR THE EXAMINATION OF WATER AND WASTEWATER (14th ed. 1976).

22. NAT. RES. COUN., TESTING FOR EFFECTS OF CHEMICALS ON ECOSYSTEMS xii (1981).

Society and The Ecological Society of America). The certification requirements, however, do not include a substantial number of publications on toxicity testing or any other substantive evidence of skill in this field. This is by no means a denigration of either of these reputable organizations or their certification processes; rather it is a recognition that present certification does not explicitly require proficiency in the most common formally endorsed standard methods for pollution assessment and hazard evaluation—single species toxicity tests. Perhaps the next phase in the development of professional certification will include more explicit indications of capabilities. Environmental Science and Ecology are such diverse fields that it is unlikely that one person could be proficient in all areas.

Gloyna, et al.[23] have pointed out the need to determine the kinds and numbers of specialists required to implement environmental legislation. Quality control systems of all kinds are only effective when a continuous monitoring system is in place—environmental quality control is not an exception to this rule. The process of hazard evaluation or risk analysis should be based on an adequate data base generated for that purpose rather than whatever can be obtained from the open or semi-open literature. These articles were almost always designated to fill other, often quite different, needs. The cost of reducing risk, including monitoring, can then be estimated and an informed decision made about the acceptable level of toxic substances to be discharged into the environment.

III. Conclusion

It is my conviction that we are now in a major transitional phase comparable to the agricultural revolution. The latter resulted from society's belief that the unmanaged environment was not capable of producing food in either the quantity or quality desired. Now there is unmistakable evidence that the unmanaged environment cannot always assimilate or recover from the anthropogenic stresses including toxic wastes, surface mining, and acid rain. Our attitudes and actions are more attuned to a frontier society which no longer exists than to a society where moving on is no longer a solution to

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23. Gloyna, E. F., R. McGinnis, L. Abron-Robinson, P. R. Atkins, M. S. Baram, J. Cairns, Jr., C. W. Cook, H. H. Folk, J. H. Ludwig, M. T. Morgan, J. D. Parkhurst, E. T. Smerdon & G. W. Thomas, 5 Manpower for Environmental Pollution Control 427 (1977).

problems. Solutions to all of the major problems of our time require the collaboration of a diverse array of disciplines that are typically isolated from each other and unaccustomed to substantive interactions. A major factor in this impasse is the university where each department is an independent entity with a different approach to problem solving. Young faculty wishing to engage in interdisciplinary research do so at considerable peril since tenure and promotion committees often credit only those contributions cast in a particular disciplinary mode. Interdisciplinary articles generally are not welcomed by traditional journals, and articles in the new interdisciplinary journals will probably not be given much weight. The reemergence of integrative science characteristic of the period before the era of specialization is essential. At the same time, we must relearn how to communicate the essence of this information to laymen, use it to make decisions, and convert these, where appropriate, to useful regulation.

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The Perspective of an Aquatic Ecologist

John Cairns, Jr.

INTRODUCTION

It is almost platitudinous to state that ecological systems are complex. Yet this basic fact of complexity is frequently ignored when environmental quality standards are developed primarily or entirely from single-species tests. It has been stated *ad nauseum* that a biological community or a natural ecosystem is more than the sum of its individual parts; however, this truism is flagrantly ignored when environmental quality standards are developed and based on "the most sensitive species" toxicity tests or more arbitrary technology-based standards with the assumption that these will inevitably protect ecosystems. Theoretical ecologists have not been helpful in the development of toxicity tests for levels of complexity above single species. Although ecologists in general deplore the impact of an industrial society on the environment, they have been remarkably unwilling to endorse professionally a standard method at the community or ecosystem level that can be used in the development of environmental quality standards. The situation is not entirely hopeless because a number of potentially useful methods at levels of biological organization higher than single species have been in the literature for a sufficient length of time so that a number of individuals have become familiar with their weaknesses and strengths. These need only be prepared as standard methods in the form suitable for environmental quality control. The benefit to industry from generating additional ecological information with these methods should be the privilege of making better nondegrading use of some receiving systems beyond the level permitted by present legislation where the level can be demonstrated as being vastly overprotective. Lest this be misinterpreted as approval of the "right to pollute," I affirm that this is not the intent of the statement "nondegrading use of assimilative capacity." The intent is to permit more flexibility of use at concentrations of chemicals below the "no adverse biological response threshold" as evidenced by data from various levels of biological organization from single species to community and ecosystem.

Adaptability to Regional Conditions

If the U.S. Department of Agriculture were to give identical advice to farmers in Alaska and Florida, the farmers would probably turn to other sources. Ecosystems in Florida and Alaska have considerably greater differences in their characteristics than do farms in those two states. Despite these recognized differences, attempts have been made to apply a single rigid standard for zinc and other potential pollutants equally to all parts of the country without sufficient regard for the modification of the expression of toxicity by regional characteristics. Why has this irrational and scientifically unjustifiable position been tolerated? Possibly because attention has not been given to the environmental qualities that need protection. Nor does a group of persons exist who are charged with managing this protection in specific ways. Some notable exceptions to this exist, of course, but management of the environment in general consists principally of protecting it from certain kinds of insults rather than keeping it in a particular condition.

Explicit Statement of Environmental Qualities Being Protected

Such general statements as maintenance of "fishable, swimmable waters" or protection of "fish, shellfish and wildlife" are too general for proper implementation. In addition to being subjective, these statements provide no indication of what specific parameters are useful in measuring these qualities. Ecologists need to endorse professionally specific qualities of ecosystems, both structural and functional, that deserve protection. These qualities should be stated explicitly. For example, it is a *sine qua non* that without normal energy and nutrient flow and processing, ecosystems and biological communities would be significantly altered. To be more specific, it is possible to measure how detritus is processed in headwater streams and to determine whether that processing is following expected patterns. Other rate functions can be stated and measured explicitly with available methodology. Structural characteristics of natural communities, such as diversity, autotrophic:heterotrophic ratios, species evenness, etc., can also be measured, and significant trends beyond normal variability can be established. Ecosystems are certainly more complex than a human being, and criteria for determining human health are more explicitly stated and rigorously measured than criteria now used for environmental health. Furthermore, by forcing explicit statements regarding the qualities being protected, the scientific justification for doing so must also be more carefully stated. This should produce enormous benefits. Development of a sound biological monitoring program will be enhanced if the desirable qualities are identified with more precision. Without question, all levels of biological organization should be included in the statement of environmental qualities being protected. The present unstated implication is that a water quality standard that results in a 96-hr LC50 for the fathead minnow may be used to protect ecosystem well-being. The scientific justification for this position is weak. A more explicit statement of the qualities presumed to be protected by a single-species fish toxicity test would probably spotlight the unsuitability of such tests for protecting ecosystem structure and function.

Standards Should Be Validated

In no standard published in the *Federal Register* or elsewhere have major efforts been made to validate the accuracy, suitability, and effectiveness of single-species testing. If a single-species test is used to protect ecosystems, validation should include some study of systems more complex to see if the extrapolation to the more complex systems is valid. In addition, if an application factor is applied to an LC50 or some other threshold so that the "no-adverse-response concentration" can include other characteristics than those directly measured in the test, the justification for doing so should be supported with specific suitable evidence. This has been the case only rarely, and what validation has been done could not be considered exemplary.

Recovery Time Following Damage

It is exceedingly well established that different ecosystems have different recovery times once displaced from their nominative state (the normal state including variability). Some ecosystems may recover quite rapidly (a matter of a few years) from severe displacement, and others may require orders of magnitude of time longer. While recovery time cannot be predicted with precision, distinctions can be made between those systems that are resilient and those that are not. There is convincing evidence that certain ecosystems are perturbation-dependent[1] and that fire, flood, or other seemingly catastrophic events must occur at intervals to maintain the genetic and species diversity characteristic of the system. Some pines, for example, require fire before their seeds will germinate. I hasten to add that no evidence exists to support the assumption that spills of hazardous chemicals will substitute for natural perturbations to which systems have adjusted. Nevertheless, there is general agreement that some localities have more opportunistic species, capable of recolonizing an area following a catastrophic event, than do others. There is also a rapidly expanding base of case histories on recoveries from oil spills, spills of hazardous chemicals, and the like. As this information base expands and is analyzed, it will be possible to draw ever more precise distinctions between those ecosystems likely to recover rapidly from displacement and those that are not. Even in the present relatively primitive state of knowledge in this area, distinctions of this kind can be made with sufficient accuracy to show significant relative differences between alternative sites for construction of new industrial facilities. Such distinctions also can be made for existing industrial plants. From a management standpoint, the significance of this information is that identical spills in two different ecosystems might put one out of commission for only a few years and the other for 30 to 100 years. In short, in one case, society would only be deprived of whatever amenities were provided by the ecosystem for a few years, and, in the other case, a significant portion or all of the lifetime of many of the residents of the area would be involved. Industries with the foresight to build plants, particularly new plants, in more robust ecosystems deserve some recognition for this effort. Those who do not might reasonably be expected to implement above-average protective measures, including biological monitoring.

GENERAL CONSIDERATIONS INVOLVED IN STANDARDS DEVELOPMENT

Evidence for Development of Criteria and Standards Should be Generated Systematically

The usage, in *Water Quality Criteria of 1972*[2], of the words "criteria" and "standards" is followed in this manuscript recognizing that an important distinction exists between the two. Development of an appropriate scientifically justifiable criterion presumably precedes production of a standard through the process described in Figure 1 of the *Water Quality Criteria of 1972*. Criteria in the Freshwater Aquatic Life and Wildlife section of *Water Quality Criteria of 1972* were developed almost entirely from information in the literature. Almost without exception, this literature was produced for purposes other than criterion development. Most research is undertaken to gain understanding of a process rather than to implement a regulatory or policy decision. This is in no way a denigration of the research effort, but rather an indication that the information that was finally used to develop criteria had actually been generated to serve other purposes. As a consequence of the unsystematic way in which the information was produced, the information base for two heavy metals, for example, might be quite different because of differences in test species, duration of testing, end points and life history stages, and (occasionally) levels of biological organization (e.g., single species, multispecies, community). Very often the information base was quite inadequate. In few cases was the information well-balanced in terms of the requirements of a criterion document. Recognition of this deficiency has resulted in the development of field and laboratory protocols.[3,4] The purpose of these protocols was to sequence the information-gathering process. Consequently, judgment of the adequacy of the information base for making a decision to use or not use the chemical in question could be made at intervals in the information-gathering process. This protocol also ensured that the information was gathered in a uniform manner for different chemicals and addressed a number of other considerations as well. These considerations are discussed at length in Cairns *et al.*[5] and Dickson *et al.*[4] Despite the availability of these protocols, most standards are still developed primarily from whatever information is available in professional journals and to a lesser extent from "in-house" or limited-distribution, non-peer-reviewed documents. Even for two chemicals with relatively similar formulation, the mix of evidence is likely to be quite dissimilar. More important, because information has not been gathered in a systematic way, critical gaps often exist in the information base. Some good examples of this are provided by the criterion documents prepared a few years ago in response to the consent decree against the U.S. EPA.

Prediction of Estimated Environmental Concentration of the Chemical Should Be Explicitly Stated

For a number of years publications (including journal articles and books) have been available on environmental partitioning, transformation, and persistence of chemicals.

Fugacity equations and other means of determining these characteristics are well known. However, rarely is this information coupled with generation and analysis of biological toxicity testing information. For example, if a chemical partitions into the sediments, it seems abundantly clear that it would be more realistic to select a test organism that lives in the sediments than one that lives in the water column. There is little indication that this is being done in a major way. On the contrary, criteria and standards are based primarily on a few well-established laboratory toxicity-testing species. It is less common to find that the organisms used for testing were selected from the environmental compartment into which the chemical in question is most likely to partition. Similarly, greater attention should be given to transformation products that, in some cases, might be of significant concern. Finally, the persistence of the chemical should be a major determinant of the length of test carried out in the laboratory and the types of monitoring used in the field. These are just a few of the ways in which chemical-fate information could be used in enhancing the value of toxicity testing both in the design of the test and in the predictions made from it. Surely, the coupling of these two important types of information on a widespread basis is long overdue.

Prediction from Effects on One Level of Biological Organization to Another

There is abundant evidence,[6] that it is not a scientifically sound practice to predict community or ecosystem response from single-species tests. As one progresses upwards from one level of biological organization to another (cell, tissue, individual, population, community, and ecosystem), new properties are added that cannot be studied at lower levels of biological organization. For example, it is impossible to study energy flow or nutrient cycling with a single species. Yet these are very important system properties. As a consequence, the assumption that establishing a standard from the response threshold of the "most sensitive species" will inevitably protect the system in which it resides is totally unjustifiable. In the first place, the low probability of selecting the most sensitive species from a meager array of organisms suitable for laboratory testing almost ensures that this will not be done. More important, compelling evidence exists that community and ecosystem processes can be impaired at chemical concentrations that will not produce single-species test responses using present criteria for end points. It is more probable, however, that due to various types of redundancy in complex systems, the single-species laboratory tests in a system lacking environmental realism will be overprotective than underprotective. In neither case, however, is the scientific justification for extrapolating from one level of biological organization to another supported by evidence in the literature. The durability of the single-species toxicity test as virtually the sole means of determining environmental hazard of chemicals is surely primarily due to the inability of professional ecologists to endorse a single parameter at a level of biological organization higher than the single species or to produce a standard method to measure such a parameter.

Fortunately, the Society for Environmental Toxicology and Chemistry and several other organizations such as the biological components of the American Society for Testing and Materials (e.g., parts of Committee D-19) have shown a strong interest in this area, and ecologists in these organizations might ultimately both identify useful parameters at levels of biological organization higher than single species and produce standard methods for measuring them. In the meantime, the usual arguments against tests at higher levels of biological organization than single species—(a) that higher level tests are more costly, (b) that they are difficult to replicate, (c) that they are more difficult to interpret—persist. All are false. Some multispecies and community tests with microorganisms can be carried out as cheaply as tests with single species. Furthermore, the argument that single species tests are less expensive becomes much less persuasive when they are considered *in the aggregate,* namely that as many as seven different single species tests may be required to make a judgment of hazard at the present time. A much larger number of species can be tested collectively in one community-level test. Furthermore, as more community toxicity tests are developed, their cost will certainly decrease. As interest in this area of research grows, it is almost certain that more cost-effective tests will be produced. Finally, it is true that a single-species test is easy to analyze only if no attempt is made to extrapolate from it to the complex systems that exist in the real world. In short, single species tests are very quantifiable, generate single numbers (e.g. LC50s), and have a number of other endearing characteristics. However, when the analysis is extended to serve the intended use of the information, its ease of interpretation can by no stretch of the imagination be considered simple. It is much easier to project from a community or multispecies toxicity test to the real world than from a single-species test, particularly if the single species is not one residing in the area in question.

Mandatory Statistical Analysis

Statistical analyses showing confidence limits, soundness of data on which standards are based, and other analyses designed to show the statistical validity of the assumptions should be included in all criterion documents. An examination of early documents reveals a regrettable inattention to statistical evaluation of information, and even recent documents are not exemplary in this regard. Surely such statements as this should be platitudinous, but they will not be so until practice matches the current state of knowledge.

Identification of the Producers of Criterion Documents

All stages in the production of a standard call for professional judgment. Criterion documents that precede the production of a standard require exceptional professional judgment and integrity because they require the analysis of a diverse array of information and the synthesis of this information into a criterion. The criterion should be scientifically justifiable. Therefore, it is regrettable that the names of those who produce particular criterion documents are not published in the documents. The credentials of the persons making the professional judgments required for the production of criterion documents is as important as the credentials of those producing evidence upon which the documents are based. It is a *sine qua non* that scientific professionals be held responsible for publications containing their ideas. This is true of the material appearing in the professional journals on which criterion documents are presently based and should be true of the production of criterion documents as well. In short, accountability is as important in production of a criterion document as it is elsewhere in science. Anonymity serves no useful purpose. The entire U.S. EPA does not produce each criterion document but rather a few individuals within the agency. There is no reason why skill and competence should not be acknowledged by associating the names of the producers with the document nor is there any reason why ineptness and shoddy work should not be chastised as well. Direct criticism is a hallmark of the scientific process. A persuasive reason does not exist for not identifying individuals responsible for preparing particular criterion documents.

Identification of Literature used in Criterion and Standards Production

All literature examined in preparing a criterion or standards document should be included *even if a decision was made not to use that information!* At the present time, one does not know (when examining a federal criterion document that is almost certainly going to be transformed into a standard by the states) whether a publication has been missed, or examined and discarded. When some colleagues and I reviewed the criterion documents prepared in response to the previously mentioned decree, we found that many publications available in the literature were not cited in the criterion document in the *Federal Register.* It was not always clear to us whether this literature had been missed or whether it had been discarded and, if discarded, why. Presumably all of the literature initially thought to be relevant was probably listed at one time in the document preparation. This literature listing should be included in the final draft to show the breadth of coverage. Presumably there is also no reason why the fact that it was ignored or discarded could not be mentioned. Such a comment would not necessarily be unfavorable to the persons who published the unused information, which might be perfectly sound scientifically but unsuitable for the purposes of the document. Scientists must, in their own publications, regularly evaluate the published work of their colleagues or be accused of missing some of the relevant literature. Even if such a practice did cause some offense, there is no reason why these same principles should not apply to criterion documents. The amount of space required for this particular exercise should be relatively small and the information would reduce considerably the uncertainty about the competence of those who prepared the document. Identifying whether a publication was peer-reviewed (regardless of whether or not it was used in the criterion document) would also be helpful. This would not require additional space but merely an identifying asterisk associated with the citation.

Endorsement by the U.S. EPA Science Advisory Board

Documents prepared in response to the consent decree were not invariably endorsed by the U.S. EPA Science Advisory Board. Since the SAB presumably should be responsible for examining such documents, an indication of its approval or disapproval should be provided. Even a statement of whether the SAB had reviewed the document would be a useful addition for those who read the *Federal Register.* Perhaps the reasons for SAB disapproval might also be included in the *Federal Register,* when this is the case. The endorsement of the U.S. EPA Science Advisory Board should be a prerequisite to the appearance of the criterion document as a final draft in the *Federal Register.* Until the document has acquired this endorsement, it should be considered provisional. Endorsement does not mean that the criterion document need not be validated, but merely that the document is scientifically justifiable in terms of the information available at a particular time.

It is abundantly clear that methodology used to produce criteria and standards documents lags seriously behind methodology available in peer-reviewed literature. Some suggestions made in this brief analysis seem self-evident, but despite this, are not being implemented. It is equally unfortunate that professional ecologists who decry environmental degradation have not given comparable attention to the need for professional endorsement of parameters and standard methods useful in resolving these problems.

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FRESHWATER BIOLOGICAL MONITORING: KEYNOTE ADDRESS

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ABSTRACT

The field of biological monitoring has advanced quite rapidly in the past 20 years. However, fuller use of existing methodology is hampered by doubts in both industrial and regulatory agencies regarding both the precision of the tests and the ways in which the information will be used. Issues related to these points are listed and discussed. None are insurmountable, but most will require sound scientific judgment.

KEYWORDS

monitoring; pollution; toxicity; assimilative capacity; application factors; regulatory standards.

INTRODUCTION

Hellawell (1978) defines surveillance as "a continued programme of surveys systematically undertaken to provide a series of observations in time" and biological monitoring as "surveillance undertaken to ensure that previously formulated standards are being met." Since it is our goal to protect natural systems before they are injured, two types of tests are needed (Cairns, 1982): (a) predictive (to estimate probability of harm) and (b) reactive (to correct errors in the predictions or validate them). Kimerle et al. (1978) propose four stages of studies in the hazard evaluation process: (a) screening, (b) predictive, (c) confirmative, and (d) monitoring. The first two are likely to be laboratory toxicity tests and the last two field studies, although numerous exceptions may occur. This discussion focuses on the screening and predictive tests because the confirmative and monitoring tests will follow only if the first two are widely accepted. They will be widely accepted only if certain criteria are utilized. These criteria will be the primary focus of this discussion.

THE RE-EMERGENCE OF BIOLOGICAL MONITORING

Over 2000 years ago, Aristotle placed freshwater animals in seawater and observed their response, presumably to answer the question how does this material affect this organism? In so doing, he carried out an aquatic toxicity test. Subsequent efforts of using biological monitoring to detect potentially adverse effects involved the king's wine taster and canaries in coal mines. However, the advent of the industrial revolution and the increased environmental stress from anthropogenic sources did not produce a vast expansion of the field of toxicity testing of nonhuman organisms. This neglect of biological testing of toxic effects was possibly due to a combination of factors that included the reluctance of classical biologists to become involved with nasty industrial problems and the fact that biology was not then regarded as sufficiently quantitative to cope with the problem. As a consequence, chemists and engineers engaged in industrial production were also charged with regulating the intrusion of industrial wastes into the environment. Most early regulatory standards for discharge of potentially hazardous chemicals into the environment allowed fixed concentrations that were not to be exceeded. This strategy proved inappropriate for several reasons.

(1) Some chemicals produce adverse biological effects at concentrations below existing analytical capabilities.

(2) Environmental quality parameters, such as water hardness, temperature, pH, and dissolved oxygen concentration, mediate the toxic response - the same concentration of zinc would produce a different toxicological response to sunfish in the hard water of the Guadaloupe River in Texas than in the soft water of the Savannah River between Georgia and South Carolina.

(3) Toxic chemicals may act differently in combination than they do individually.

The second approach was to use technology-based standards, such as Best Applicable Technology (BAT) and Best Practicable Technology (BPT), which replaced the "pipe standards" approach of the first method. The strategy was to utilize the best technology available and assume that it would protect the environment. From an ecological standpoint, technology-based standards ignore:

(1) size of the receiving system.

(2) well-established fact that environmental quality (e.g. pH, water hardness, temperature, etc.) may markedly influence toxicity.

(3) total impact of all stresses on natural systems, not just the impact of a single discharge, must be considered.

From an industrial standpoint, use of BAT and BPT may be unsuitable because:

(1) small industry on a large river may be forced to spend money on new technology that produces no demonstrable biological or ecological benefits.

(2) long-range financial planning is difficult when the rate of technological development is difficult to predict.

(3) operators with new equipment that they cannot use properly may produce poorer quality effluents than they would with old equipment they understand.

Since both effluent-based standards and technology-based standards have proven at least partially inadequate, society and its representatives have belatedly recognized that biological evidence must be combined with chemical/physical measurements to evaluate hazard to human health and the environment effectively. In short, there is no instrument devised by man that can measure toxicity; only living material can be used for this purpose. This immediately produces both scientific and regulatory difficulties because living material is complex, regionally differentiated, usually highly variable, and may act differently in laboratory test containers than in natural systems. By default, the "ball is back in the biologist's court," and we are getting a second chance to show that biological evidence is pivotal. However, we must remember that biological evidence is not the only kind required in the hazard evaluation process. This time we must not fail to produce useful biological monitoring methods!

Perhaps, an examination of the criteria that "users" would apply to determine whether a toxicity test or other measurement of environmental stress is useful would be appropriate at this point in the discussion. Some of the desirable qualities follow.
(1) The most appropriate end points (e.g. lethality) must be identified and endorsed by professionals in the field of toxicity testing.
(2) Standard methods (or, alternatively, widely-endorsed methods) must be available to measure the end points.
(3) The tests must be required in specific regulations because most toxicity testing is carried out in response to regulatory requirements.

Regulators use results of toxicity tests for three major purposes:
(1) Rangefinding (e.g. screening) tests
Because these are frequently used by a very diverse group of organizations, some with few financial resources and staff trained for other purposes, they should be simple, rapid, inexpensive, and have wide applicability. Test organisms should be easily available and not difficult to maintain in the laboratory. The response end point should have a high sensitivity to stress to reduce the probability of false negatives.
(2) Establishing limitations
Tests should be of known precision with exposures that simulate environmental exposures and should be applicable to a wide range of site-specific situations.
The response used as an end point should be directly related to environmental hazard, easily interpreted, and meaningful to the public and courts. Alternatively, a "science court" might be established that has the confidence of the public to determine the validity of hazard assessments based on methodology too complex to be easily understood.
Results should be directly translatable into specific decision criteria.
The end point should be a discrete variable (e.g. death) to reduce the possibility of different interpretations. If the end point is not a discrete variable (a likely possibility for toxicity tests at higher levels of biological organization than single species), decision criteria used for selection should be explicitly stated.
(3) Monitoring
Tests should be rapid, inexpensive, and of known precision.
Response should be sensitive and relevant to the type of limitation imposed.

PREDICTIVE CAPABILITY

It is a sine qua non that biological monitoring must be eith able to detect deleterious biological effects quickly in a natura system or predict them before they occur. Since it is our goal to p event injury to natural systems, the predictive capability of any quality control system is extremely important. Most environmental scientists and engineers would characterize this statement as platitudinous. This makes the failure to check vigorously our predictions of response of natural systems to chemicals and other stresses all the more curious. Of course, this has been done, but the collective effort can hardly be characterized as persuasive or scientifically justifiable. An outline of some factors that should be considered follows.

(1) Reliability of Extrapolations from Test Systems to Natural Systems

It is extremely important to determine more definitively whether we can extrapolate from single species laboratory tests to responses of the same species in natural systems. Some evidence exists (e.g. Cairns and Cherry, 1983) that with a modest amount of environmental realism the correspondence between the laboratory response and the response in the field is remarkably similar, even when the laboratory test system differs in many ways from the natural system. However, this particular series of tests used the same species in the laboratory from the natural system as well as test water from the river upstream of the plant discharges. The fish were obtained from an area not affected by plant discharges and were, as a consequence, thoroughly acclimated to the chemical and physical characteristics of the water used in the test system. The correspondence between responses measured in the laboratory and in the field does not appear to be nearly as great when extrapolations are made from one species to another, even in the same taxonomic group. When extrapolations are made from *Daphnia* to fish, the correspondence may be even more reduced, although the evidence for this is not extensive. A good summary of this problem may be found in Kenaga (1982) and Kenaga and Moolenaar (1979). There is no significant body of evidence on the precision with which one can extrapolate from responses of single species toxicity tests or biological monitoring involving single species to the response end points of more complex systems such as communities and ecosystems (e.g. National Research Council, 1981). Since the ability to extrapolate from one response threshold to another using single species is not always precis it seems intuitively reasonable that extrapolation from a response end point of a single species test to a response end point of a complex system would be considerably less precise. If one uses response end points characteristic of higher levels of organization than single species, this means that such a particular end point is not measurable with single species, and, therefore, one would think that the probability of extrapolation with any degree of precision would be very low. Even if these speculations are not correct, the matter is of sufficient importance to justify the generation of an adequate body of evidence to determine the accuracy of the various kinds of extrapolations just discussed.

(2) Interpretation

What does the laboratory response mean in terms of environmental hazard? Lethality of a chemical concentration to fishes is easily understood by both scientists and laymen. More important, the environmental hazard is unmistakable. However, concentrations of a chemical that affect protozoan colonization processes (e.g. Cairns et al., 1980) are neither well understood nor widely accepted. Most people do not even know what protozoans are, and even knowledgeable toxicologists are not likely to be as influenced by results of such tests as they are by fish tests. Nevertheless, abundant evidence shows that freshwater protozoans are important regulators of bacteria in natural systems and contribute in a variety of other ways to important nutrient spiraling and energy transfer processes. I deliberately chose some of my own research as an illustration so that I can comment, without offending anyone, that I am not yet at all certain what this information means in terms of overall environmental hazard. In short, the method may be satisfactory from both a cost and reliability standpoint, the end point (in this case, colonization rate) may be one that is regarded as important by microbial ecologists, but the exact hazard resulting from a change in this process is still unclear. Thus, ecologists will not only have to develop more tests at higher levels of biological organization than single species than are now available but will also have to reach a consensus on the meaning of the response in terms of environmental hazard.

(3) Sensitivity

From a regulatory standpoint, it is desirable to have the response or end point sufficiently sensitive to avoid excessive false negatives. The precise level of sensitivity desired is a function of an array of factors, including: (a) number and kinds of alternative measurements possible, (b) objectives of the study, and (c) proximity of the environmental concentration of the chemical (or other environmental stress) to a critical threshold. It is also worth reaffirming that the response being utilized must be of ecological and/ or biological significance.

(4) Variability

If the response being measured is a discrete variable (e.g. lethality), the precision of the measurements being made can be more easily documented than is possible for non-discrete variables (e.g. nutrient spiraling). Nevertheless, a correlation seems to exist between the relative sensitivity of the tests (for example, behavioral changes occur before lethality) and the degree of variability encountered. Thus, the attempts to optimize both sensitivity and reduced variability will probably be fruitless, and a compromise between the degree of sensitivity and reduced variability will be required in many cases.

(5) Replicability

For regulatory purposes, toxicity tests and other methods used for biological monitoring must be sufficiently simple and standardized so that they can be carried out by governmental, academic, and private laboratories that have widely varying capabilities. Ideally, quality control aspects must be sufficient to obtain consistent results with acceptable inter-laboratory and intra-laboratory levels of precision. Again, an obvious conflict occurs when one attempts to obtain adequate environmental realism in a test and, simultaneously, a high degree of replicability following the criteria just mentioned. In some cases, replicability should be sacrificed, or at least muted, to obtain a greater degree of environmental realism, and the requirement that the method be suitable for utilization in a wide variety of laboratories may not be possible.

Resolution of these often conflicting requirements is only possible when the specific objectives of the study are explictly stated. When this is done, the various criteria just discussed should be considered, and the reasoning used in assigning priorities to them should be stated along with the objectives of the study.

SELECTION OF RESPONSE PARAMETERS

The response parameters used for single species tests are extremely well known and include such end points as lethality, growth rate, reproductive success, and behavior. Those for tests at other than single species and at higher levels of biological organization, such as multispecies, microcosms, community, or ecosystem tests, are less well known. Examples of response parameters for these complex systems are productivity, nutrient cycling, bioaccumulation, predator/prey interactions, diversity, colonization rate, decomposition, trophic balance, microbial functional attributes, and energy transfer. Virtually any parameter that can be measured can be used for biological monitoring, but obviously not all would be appropriate; the most appropriate would vary from one situation to another depending on the objectives of the study. Perhaps the best solution to the implementation of regulatory requirements involving such a vast array of end points is to follow the practice of Section 316a, Public Law 92-500, and put the burden of selecting methods and parameters upon the person or institution wishing to make use of the environment so that maximum flexibility is available in the selection of end points and methods. The proposal would then be reviewed by a competent group of scientists who would judge the adequacy of the end points and data base in view of the objectives of the study.

RELEVANCE AND ACCEPTANCE OF BIOLOGICAL MONITORING

Biologists are, in a sense, being given a "second chance" to show that the methodology they are producing has relevance in pollution abatement and hazard assessment. This renewed opportunity is due not so much to the perceived relevance of biological monitoring as it is to the notable failure of regulations based on "pipe standards" or technology-based standards (e.g. best applicable technology). The environmental impact statements, required in the United States for a number of years, have done nothing to enhance the role of ecology and biological monitoring and may have done considerable harm. The major reason for this is that almost all the statements consisted primarily of species lists with very little analysis of data, predictions based on the information gathered, or even an explicit and precise statement of the probability of h.. resulting from the proposed course of action. Even the most charitable scientist would be unlikely to label such documents as good science. The cost was generally enormous for biological measurements, and the useful information was miniscule. Only an extremely small fraction of the effort resulted in publications in the peer-reviewed literature. Although this was not the primary purpose of environmental impact statements, they were supposed to result in extrapolations and the ability to predict and validate the predictions of the responses of complex systems, if carried out in a scientifically justifiable way, would certainly be of enormous interest to the profession. In order to enhance acceptance of biological monitoring for environmental quality control and pollution abatement, those of us in the field must include decision criteria for the development of methods we expect to be used by engineers and others involved in pollution abatement and environmental quality control. Simultaneously, we must document their relevance to the general public and the decision makers in regulatory agencies, industry, and various levels of government. An illustrative checklist of such decision criteria and their relevance follows.

1. Technical Relevance

Does the end point or response parameter being used represent a realistic measure of population, community, or ecosystem level impact? Using this information, can one provide margins of safety based on objective criteria?

2. Social Relevance

Is the response meaningful to the public and the courts? The latter may not apply in other countries to the degree that it does in the United States where the legal system is the primary arena in which these matters are settled rather than in the scientific and engineering system.

3. Legal Relevance

The understanding and support of the general public is equally important because at the time this manuscript is being prepared, the federal deficit in the United States has reached staggering levels and the prospects for major reductions in an election year appear minimal. As a consequence, any activity that increases expenditures of federal and state funds will not be viewed as benignly as a comparable measure would have been in the 1960s. We must demonstrate that the response (end point) is useful for establishing limitations on the discharge of a substance or in the study of nonpoint source discharges. If the response is a continuous variable, is there an objective means of establishing a limiting exposure concentration to avoid hazard? The "no-effect" level, in terms of mortality as determined in single species toxicity tests, may be the only presently available discrete variable resulting from toxicity tests.

4. Cost and Timing

The enormous increases in energy and labor costs together with foreign competition have made industries extremely cost conscious in recent years. Federal deficits and pressure on state tax funds have created comparable pressures in regulatory agencies. As a consequence, one must provide convincing evidence that the cost is reasonable in terms of test objectives. Cost is, of course, largely a function of the time necessary to perform tests, space and facilities required, and the level of professional competence necessary to get results and interpret them. The decisions regarding acceptable costs are driven primarily by the degree of certainty required in the results.

Advocates of biological monitoring will be interested in a brief article entitled "Biomonitoring: A Useful Tool for Industry, Government" in Chemecology (p. 6, April, 1984) published by the Chemical Manufacturers Association. The following quote (representing roughly half the article) probably represents the position of United States industry.

"CMA commends the Environmental Protection Agency's efforts to integrate biological assessment techniques into the nation's water quality management program, but believes that there are limits on the use of this tool.

CMA believes that biological assessment is useful to screen plant effluent for potential harmful effects and to determine whether additional studies of receiving waters are needed.

Biomonitoring results also may show whether additional pollution controls are necessary to prevent environmental harm.

CMA, however, has serious technical objections to using biological assessment to set effluent limits, since not enough is yet known about its precision or accuracy."

Clearly two efforts are needed: (a) a comparison of the precision of alternative methods used to set effluent limits with biological monitoring and (b) a more precise evaluation of the accuracy of biological monitoring in terms of explicitly stated quality control objectives. With regard to the first point, chemical/physical measurements may be more precise in that they often have low variability, but they cannot predict toxicological responses as well as biological evidence. I believe CMA has confused the precision of the measurements with the precision of the predictions or extrapolations that should be "to promote a clean environment." In any case, they are justified in calling for more information about the precision of biological assessment to set effluent limits.

In the United States, and probably elsewhere in the world, few legislators and the general public they represent place a high value on protecting the qualities valued by ecologists, such as nutrient spiraling, energy flow, ecosystem stability, and the like. Present water quality goals of society are clearly oriented toward protection of important recreational and commercial species, and biological monitoring methods designed to protect them will almost certainly get the highest priority. If we wish other methods to be accepted, we must either show their relevance in terms of society's present goals or educate the public so that an appreciation of the subtleties and values of other environmental qualities are appreciated. Clearly, most ecologists hope that the environmental quality goals of society will evolve to consider protection of ecosystem structure and function, but these qualities are not appreciated by the general public and their representatives.

THRESHOLDS AND APPLICATION FACTORS

For many years, a relatively low key discussion has existed regarding the existence of any thresholds at the ecosystem level (e.g. Woodwell, 1975) or whether multiple thresholds exist (e.g. May, 1977). It is extremely improbable that theoretical ecologists will unanimously endorse a particular position on the existence of thresholds at levels of biological organization higher than single species in the next 20 or so years. Since the concept of zero discharge (forever containing societal wastes) and zero risk have fallen into disfavor for a variety of reasons and because the economic and technological resources are not available to achieve zero discharge even if it were possible, it is clear that some societal wastes will continue to be discharged directly into the environment. Although precise data are difficult to obtain, it is quite likely that an equal or greater amount of discharge will enter streams and other parts of the environment from nonpoint sources. For practical and regulatory purposes, there seems to be a threshold concentration below which no deleterious or adverse ecological effects occur. Even if this assumption is false and damage occurs below the level at which detection is possible or if the activity or chemical is considered essential to society, the amount that can be introduced into the environment must be determined. In these and other cases, arbitrary thresholds would be used even if real ones did not exist. As a consequence, two basic questions arise: (a) If one assumes there are multiple thresholds that are either real or artifacts of the measurement process, which one or ones should be selected for biological monitoring and hazard evaluation? and (b) Can these thresholds be measured directly or should some other threshold be used and the critical thresholds be extrapolated by using an application factor? An examination of the evolution of application factors will help in answering these questions.

Toxicity tests on industrial wastes in the late 1940s and early 1950s were commonly crude (by today's standards), short-term batch tests with fish that used lethality as an end point. Even the first method (Hart et al., 1945) that was widely accepted for toxicity testing of industrial wastes in this country clearly recognized that adverse biological effects were likely to occur at concentrations lower than those producing no response in that specific test. Variability on either side of the results might be due to the fact that only a small portion of a population was used in the test, that other untested organisms might differ in sensitivity, that unconsidered environmental factors might alter the response threshold, or that the duration of exposure was not adequate to detect fully the expressions of toxicity. In short, the fact that organisms might be more sensitive than the test indicated was of greater concern than the fact that some might also be more tolerant. As a consequence, Hart et al. (1945) included an equation to determine the "biologically safe concentration." This equation used an arbitrary application factor of 0.3 coupled with the slope-squared [determined by using two different LC50s (then called TL_m)] obtained at different time intervals. The history of the development of application factors is fairly well known to most readers. The important feature is that application factors moved from 0.1 of the 96-h LC50 to 0.001 of the 96-h LC50 as the information base about toxicity expanded. This was an inevitable consequence of using the "worst possible case" approach in developing application factors. The corollary to this is overprotection in most cases. This is attractive to environmentalists but not to cost conscious industry.

The basic purpose of an application factor is to extrapolate from various types of information, including LC50s, to estimate the no-adverse-biological-effects concentrations for permanent exposure. When the field of toxicity testing was in its infancy, the no-adverse-biological-effects threshold could not be measured directly because the number of tests available was relatively limited and generally involved fairly crude end points such as lethality. Confirming estimates of "safe concentrations" in the real world were rare, and evidence of success was primarily anecdotal and circumstantial. In short, the absence of highly visible catastrophies gave some reassurance that the application factors in use were reasonably sound. However, most application factors were essentially arbitrary, and their justification from a scientific standpoint was relatively weak. The rapidly developing field of hazard evaluation (e.g. Cairns et al., 1978) recommends a testing sequence of (a) screening tests, (b) predictive tests, (c) confirming tests, and (d) monitoring. The requirement that predictions be confirmed represents a major form of error control not previously present in the development of application factors, test methods, etc. Preceding the development of a hazard evaluation frame of reference was the development of a large series of life history stage tests and other toxicity tests using far more sensitive parameters than lethality. The entire life cycle of some macroinvertebrates can be carried out in a matter of 7 or more days. The development of multispecies toxicity testing procedures (Cairns, in press) provides means of evaluating responses of more complex systems than single species. This combination of available tests for the most sensitive stages in the life cycle of an organism and the possibility of measuring various functional attributes of more complex systems suggests that attempting to measure the no-adverse-biological-effects concentration directly is better than continuing to extrapolate to it as has been done in the past. If the attempt to measure the threshold directly is accompanied by confirmation in either microcosms, mesocosms, or field situations, errors that appear can be quickly and skillfully corrected. Furthermore, attempts at direct measurements focus attention on the deficiencies in the field of toxicity testing in a way that did not occur when scientists had more faith in the efficacy of application factors.

Application factors have served a very useful purpose in the development of the field and undoubtedly offered a degree of protection for natural systems that would otherwise have been lacking. However, recent developments in the field of toxicity testing, including arraying the information in a systematic fashion for the purpose of evaluating or estimating hazard (e.g. Dickson _et al_., 1979), make direct measurement of response thresholds more acceptable than estimated thresholds. This is not to advocate the total abandonment of application factors in the near future, but rather to urge that some consideration be given to the alternative suggested. (This discussion on application factors is reprinted by permission of the _Journal of the Water Pollution Control Federation_.)

The Hart _et al_. (1945) publication was ultimately adopted as a standard method by the American Society for Testing and Materials and also received wide attention when it was subsequently refined by Doudoroff _et al_. (1951). Both publications included an application factor designed to take care of variability not included in the test itself (such as changes or differences in water hardness, temperature, and the like) and also factors, our knowledge of which even the most charitable person would concede could only be described as substantial. It is clear that W. B. Hart had serious reservations about using laboratory data without field validation because he was the person who urged Ruth Patrick to form a field team consisting of a variety of specialists in aquatic ecology to determine the effects of toxicants on natural aquatic systems. Since this field team began operations in the spring of 1948, Hart undoubtedly realized the problems then associated with application factors. This is probably why the term _biologically safe_ that is used in the application factor equation is not more explicitly defined.

If one's objective is to protect a system rather than an aggregation of literally hundreds of species (the numbers likely to be found at a particular spot), using system attributes for toxicity tests rather than attributes of single species seems more plausible. A definite, highly predictable relationship may exist between the response to toxicants of single species and system attributes, but until this has been demonstrated in a scientifically justifiable way, assuming that such a relationship exists is not justified. If a direct measurement can be made of the quality being protected, then performing such a measurement directly is more reasonable than extrapolating from some other quality presumed to have a relationship to the quality being protected.

Industry has often mistakenly tried to dispose of its wastes by relying entirely on single species acute toxicity tests plus the currently acceptable application factor. In short, many have chosen what appears to be the cheapest biological test that will satisfy regulatory requirements. The regulatory agencies have chosen the same test and application factor because they are easily administered. However, the application factor is a substitute for direct information that is either thought to be unmeasurable or more costly. In no other area of industrial management would such poor information be tolerated! Lack of information has consistently been proven costly, and the consequences of using inadequate information for management decisions about toxicity is just now being seen. Regulatory agencies are equally guilty of substituting an administratively attractive and legally defensible method for one that is more scientifically justifiable. If a method is scientifically justifiable, it is almost certainly less legally defensible since courts commonly let legal procedures take precedence over scientific judgment. The scientific community has not been particularly helpful in getting beyond this unsatisfactory stage in hazard evaluation because of the refusal to endorse professionally some of the methods that might be useful in making these measurements. Scientists should be asking the question "Are the more complicated methods of direct measurement more satisfactory in view of the ultimate objective than the present methods being used?" rather than "Are there any faults that we can find with the alternative methods?" Theoretical ecologists also often fail to distinguish between what is essential and what is desirable.

ASSIMILATIVE CAPACITY

A basic assumption in all toxicity testing is the existence of a no-adverse-biological-effects threshold for both single species and higher levels of biological organization, such as communities or ecosystems. Introduction of new material, whether natural or synthetic, into an ecosystem will cause change, even though this change may not be detectable with present methodologies because of the enormous natural variability that occurs in most ecosystems. A key issue in validating the efficacy of this assumption (i.e., there is a concentration below which no deleterious effects occur) is the ability to differentiate between deleterious changes and non-deleterious changes. No agreement has been reached on the methodology to determine whether deleterious changes have occurred or whether the same thresholds are important for all ecosystems. Those who have not followed this controversy might find an exchange between Ian Campbell and me interesting. Campbell (1981) attacked some of the suggestions made in two of my articles on assimilative capacity (Cairns, 1977,a,b) as well as those of Westman (1972). Both Westman (1981)and I (1981a) responded, pointing to some of the weaknesses in Campbell's arguments. The publications just cited also contain citations of other literature relevant to this discussion that scientists might find useful.

If all anthropogenic introductions into natural systems have deleterious effects, we are indeed in trouble! This is another way of saying that natural systems and an industrial society cannot co-exist. In order to reduce, if not eliminate, our impact on natural systems, the human population should be reduced to millions from billions and returned to the hunter-gatherer stage of societal development from the agro-industrial stage. This could not be achieved very quickly, even in the unlikely event that there was general agreement to do so. Therefore, regardless of whether true biological response thresholds to chemical concentrations and other societal impacts exist, we must make some quality control decisions. If there is no assimilative capacity, we would still need to determine the degree of impact caused by various concentrations and determine the acceptable level. We are limited in making these determinations by the sensitivity of the methodology available and, therefore, whether the thresholds are real or illusory (i.e., a function of the sensitivity of the methodology); we can only make management decisions based on effects that are detectable. Therefore, while the existence of thresholds (and, therefore, assimilative capacity) may not be as well documented as a science court would wish, present limitations of science force us to make decisions as if they were.

WHOLE EFFLUENT TOXICITY TESTING

In the United States in the 1940s and 1950s, much attention was given to whole effluent toxicity testing and field studies in mixing zones below discharges (now called allocated impact zones). However, this approach fell into disfavor and single species laboratory toxicity tests with pure chemicals became the primary means of setting discharge limitations. However, in view of the strikingly expanded array of methods and information compared to the 1940s, 1950s, and even 1960s, the biologically defined treatment adequacy of whole effluent testing is justified. Furthermore, such an approach provides the single best opportunity to accomplish a number of objectives:

(a) to relate criteria and standards based on single species, single chemical laboratory toxicity tests to responses of single species to effluents and other mixtures of chemicals.

(b) to confirm or validate the correspondence of responses predicted on the basis of laboratory studies to those actually occurring in the field.

(c) to determine if the prediction of no-observable response based on single species toxicity tests is valid for other levels of biological organization, such as communities and ecosystems.

(d) to determine if an allocated impact zone below an industrial waste discharge (an area of discernible adverse biological effects) is an ecologically justifiable strategy.

(e) to develop a series of _in situ_ receiving system assessments (biological, physical, and chemical) related to the pressing problems of nonpoint source discharges and the means of coping with them.

CONCLUSIONS

A few years ago, I wrote an article entitled "Biological Monitoring: Future Needs" (Cairns, 1981b). I see no reason to make any substantive changes in those recommendations. However, some needs not mentioned in that article are in this discussion, namely the relationship to regulatory and industrial concerns. There will probably never be any instrument devised by man that will measure toxicity, and we will always be required to use living material for this purpose. It is a sine qua non that biological monitoring must be accompanied by chemical/physical monitoring in order to be effective. Nevertheless, the failure of alternative methods to produce satisfactory results forces other disciplines to look to biologists and ecologists for the ultimate answers about concentrations producing no-deleterious biological effects and the like. They obviously do this with mixed feelings because they have suspicions about the reliability of biological monitoring information, its cost effectiveness, its utility in making predictions and catching errors in these predictions, and probably, most important, the understanding of legislators and the general public for its value. We must simultaneously develop methods that meet at least some of the requirements discussed in here and in the 1981 article, and, when this is not possible, give explicit reasons why they could not be met and why the additional expenditure of funds and effort are justified in order to get more sophisticated information. Despite the formidable complexity of the problems, there is ample justification for believing they can be resolved.

ACKNOWLEDGEMENTS

I am deeply indebted to the participants of the Multispecies Toxicity Testing Symposium jointly sponsored by the Ecological Society of America and the Society for Environmental Toxicology and Chemistry. Although I naturally take full responsibility for the ideas expressed in this discussion, I wish to express particular thanks to Lee Tebo, Gorden Loewengart, Alan Maki, and Frank Golley whose presentations at the symposium just mentioned and the chapters to appear in the proceedings of that symposium stimulated my thoughts for substantive portions of this discussion. The proceedings of the multispecies symposium are being processed by the Editorial Board of the Society for Environmental Toxicology and Chemistry and should be published in 1984 or early 1985.

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7

Freshwater Protozoan Communities

J. Cairns, Jr.

1. INTRODUCTION

Protozoan communities are ecologically fascinating systems that provide a magnificent opportunity to investigators interested in testing various ecological hypotheses. In many instances, these communities fit models developed for higher organisms. The small size of protozoan communities and the speed with which events occur are simultaneously advantageous and frustrating. In a small space and a short time, research can be accomplished that would take many years for larger organisms. However, rapid movement of individuals and turnover of species make individual interactions difficult or impossible to detect and quantify, unless the components are studied in isolation from the remainder of the community.

Although the literature on freshwater protozoan communities is not large, it is still too large to cover adequately in a single chapter. The majority of readers probably do not have extensive first hand research experience in this area but may wish to carry out such studies or at least be aware of their significance. As a consequence, the objectives of this chapter are:

(1) to illustrate the utility of protozoan communities for both theoretical and applied studies;
(2) to provide references so that the reader can easily acquire additional information on protozoan communities, and
(3) to acquaint the reader with the use of artificial substrates which are now commonly used in a variety of studies of protozoan and other microbial communities.

Much of the early literature on ecology has been summarized by Noland and Gojdics (1967), a good general text is Sleigh (1973), and one of the important older papers on protozoan community structure is Picken (1937). Unfortunately, studies of spatial relationships within a protozoan community are rare and such information is badly needed. Picken (1937) was one of the first to note that an assemblage of protozoans is a complex community of herbivores, carnivores, omnivores, and detritus feeders which form a closed social structure. He also analysed food chains in protozoan communities that had notably different microbial associated communities of diatoms, cyanobacteria, and bacteria. Much attention has been given to the protozoans in sewage treatment systems where they have important roles to fulfill (Lackey, 1925; Barker, 1946; Curds, 1966, 1973) and to the degradation of organic compounds in natural waters (Bick, 1971). The passive dispersal of protozoans which is important in both colonization and succession has been studied by Gislen (1948), Schlichting (1961, 1964, 1969), Maguire (1963), Milliger *et al.* (1971), and Blanchard and Parker (1977). Succession has been studied by Woodruff (1912), Eddy (1928), Unger (1931), Cooke (1967), and Yongue and Cairns (1971). Ecological factors affecting protozoan distribution have been studied by Hausman (1917), Stout (1956, 1974), Kitching (1957), and Webb (1961). Interactions with other microorganisms have been studied by Hairston *et al.* (1968) and competition between protozoan species by Evans (1958). Maguire (1977) studied colonization processes with particular attention to changes in the autotroph to heterotroph ratio during this process. Brooks and Dodson (1965), Hrbacek (1977) and others have shown that predation may alter community composition of the plankton, including protozoan component. The importance of protozoans as grazers of natural bacterial populations has been discussed by

Bick (1958) and Fenchel (1977). A good summary of the literature on the importance of surface films may be found in Parker and Barson (1970). Ruggiu (1969) discusses the benthic ciliates in the profundal of Lake Orta, Northern Italy. Other studies of benthic protozoans include Moore (1939), Cole (1955), and Goulder (1974). Wang (1928) discusses the seasonal distribution of protozoans.

Various functions, e.g. respiration and production, have been studied by Ganf (1974) and Finlay (1978). Quantitative assessment methods have been developed by Sramek-Husek (1958), Borkott (1975), and Finley *et al.* (1979). Colonization of artificial, uninhabited substrates by microorganisms has been discussed by Butcher (1946), Cooke (1956), Grzenda and Brehmer (1960), and Spoon and Burbanck (1967).

In studies of natural communities, theoretical possibilities must be distinguished from what can be measured. Many years ago the poet Francis Thompson wrote ". . . Thou canst not stir a flower without troubling of a star." Later Hardin (1969) illustrated the validity of this hypothesis by lifting a flower in a vase and pointing out that he does indeed "trouble a star" because Newton's Law states "every body attracts every other body with a force that . . ." and so on. As Hardin lifted the flower, literally every star in the universe, even those beyond the reach of the most powerful telescopes, had its position and motion altered by virtue of the law of universal gravitation. However, although the validity of this assumption is recognized, it is ignored because it is practically of no importance. Thus, some theoretical effects are quantitatively beyond our ability to measure them and, therefore, of no operational utility. Similarly, although every alteration in environmental quality or community composition triggers a chain of events within the community, these are often beyond our capacity to measure.

The long evolutional history of protozoan communities, the high probability of cosmopolitan distribution for many of these species, and their relative morphological stability (keys produced by Kahl (1930) and Pascher (1913–1927) to mention only two that are still as effective as they were when they were written) raise the tantalizing possibility that assemblages of protozoan species may have a reasonably constant structure as well as synergistic, functional relationships. All of the statements just made about protozoans are also probably equally valid for diatoms. In addition, aggregations of diatom species have the advantage of being more readily preserved and, therefore, provide a greater opportunity for detailed counts of both the species diversity and the number of individuals per species (evenness). Such analyses are much more difficult for protozoans since preservation of a community consisting of a large number of species (many in extremely low numbers mixed with the inevitable detritus) is virtually impossible. Some of these problems have been discussed in detail by Cairns (1965) and will not be repeated here. However, since the similarities just noted exist between protozoans and diatoms, it is advantageous to use information generated with diatoms when comparable information for protozoans is exceedingly difficult to obtain. One of the most fascinating of these data sets is the curves originally proposed by Preston (1948, 1962) and confirmed by Patrick (1949) with diatoms, illustrated in Fig. 1. Patrick, frequently in association with various colleagues, has published a number of papers on this subject; many are discussed in Patrick (1977). The fascinating implication of this distribution which occurs in a wide variety of temperature zone streams at all seasons, is that not only is the species richness remarkably constant (considering the number of potential colonizing species) but also the numbers of individuals per species are arrayed in a predictable fashion. In short, a very high probability exists that a certain number of species with two to four individuals per species as well as a certain number with four to eight individuals per species

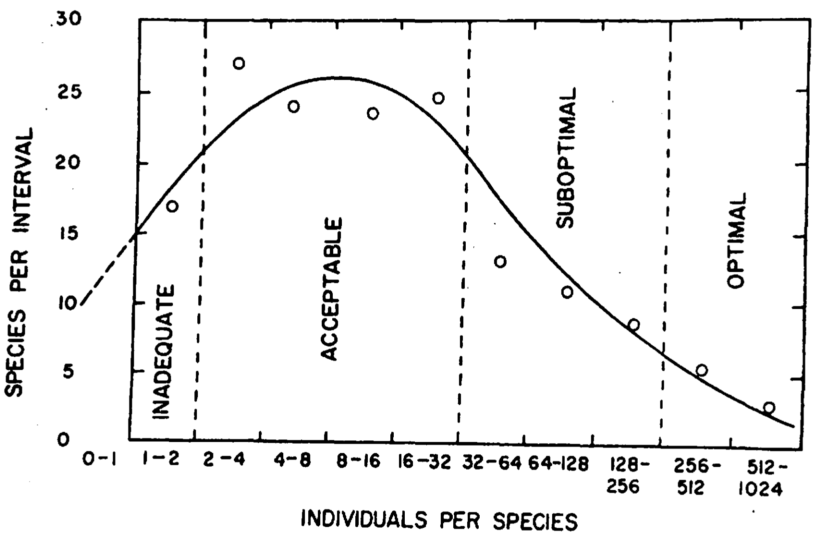


Fig. 1. Model for the truncate normal curve distribution hypothesized by Preston (1948), confirmed by Patrick *et al.* (1954), and modified by Cairns (1971).

and so on will occur. Protozoans quite likely follow the same distributional system because crude estimates of abundance show that low density species are by far the most numerous (Cairns *et al.*, 1969) and high density species are few. This follows in a very general way from the more detailed analysis possible with diatoms. The fact that protozoan communities appear to be organized in distributional patterns, similar to those of diatoms and even birds, e.g. the MacArthur-Wilson model, shows that such communities deserve careful attention and study.

Cairns and Yongue (1977) offered two hypotheses (not mutually exclusive) for the truncate normal curve found by Patrick and her coworkers. First, the chances of finding optimal conditions by organisms which are passively transported are slim but, if they do, they will flourish (i.e. low number of species—high number of individuals per species). The chances of finding sub-optimal conditions are greater, but the species that do so will not flourish and so on. The second hypothesis assumes a limited number of functional roles with any one of an array of species filling the role at a particular time (Fig. 2).

A necessary caveat at the conclusion of this introduction is that protozoans should not be considered in isolation from other members of the aquatic microbial community, such as algae, bacteria, and fungi, as well as the smaller metazoans, such as rotifers and gastrotrichs.

2. SIMILARITIES OF PROTOZOAN COMMUNITIES TO THOSE OF HIGHER ORGANISMS

A fallacious belief is widely held that protozoan communities have characteristics quite different from communities of higher organisms. A few illustrations will demonstrate that protozoan communities are structured by the same principles as those that shape communities of higher organisms.

2.1 Species—Area Curves

A species–area curve is developed when a comparatively homogeneous area (in terms of the habitat) is sampled starting with a relatively small area and increasing the area geometrically. This usually provides a clear demonstration that:

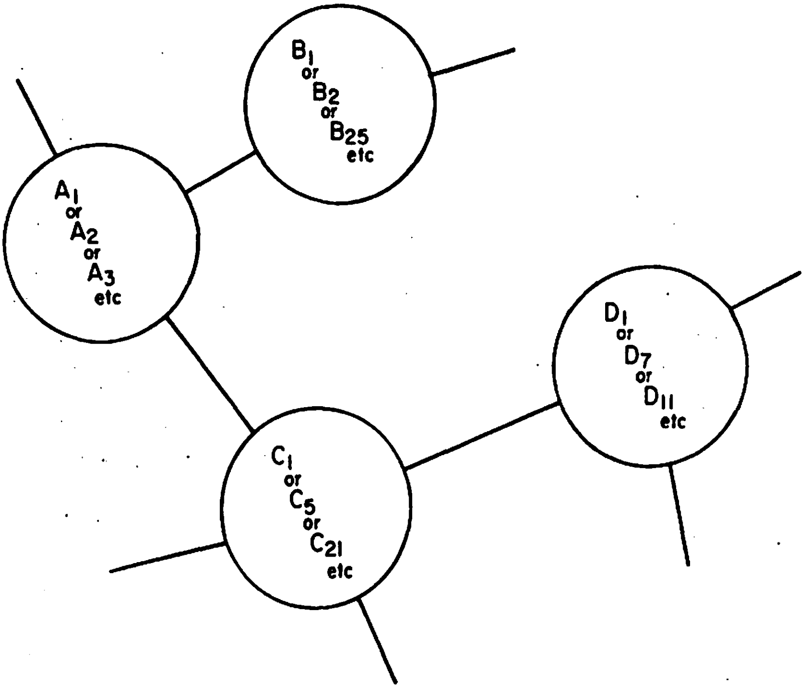


Fig. 2. Role A might be temporarily filled by a species designated A_1 or, as far as species temporarily filling role B is concerned, A_7. Thus community structure would be relatively stable despite species replacing each other because the "role relationships" remain constant. Division of role A among species might be determined by differential tolerances to pH, temperature, etc. The number of roles would, of course, greatly exceed those depicted in this simplified diagram (from Cairns, 1977).

(1) when the initial area is small, increasing the size of the area sampled results in a marked increase in the number of species found;
(2) but further increases generally result in a diminished number of species in proportion to the increase in area sampled.

A comparable curve for protozoans is given in Fig. 3 (Cairns and Ruthven, 1970).

2.2 Colonization—The MacArthur-Wilson Model

Simberloff (1974) has pointed out that "any patch of habitat isolated

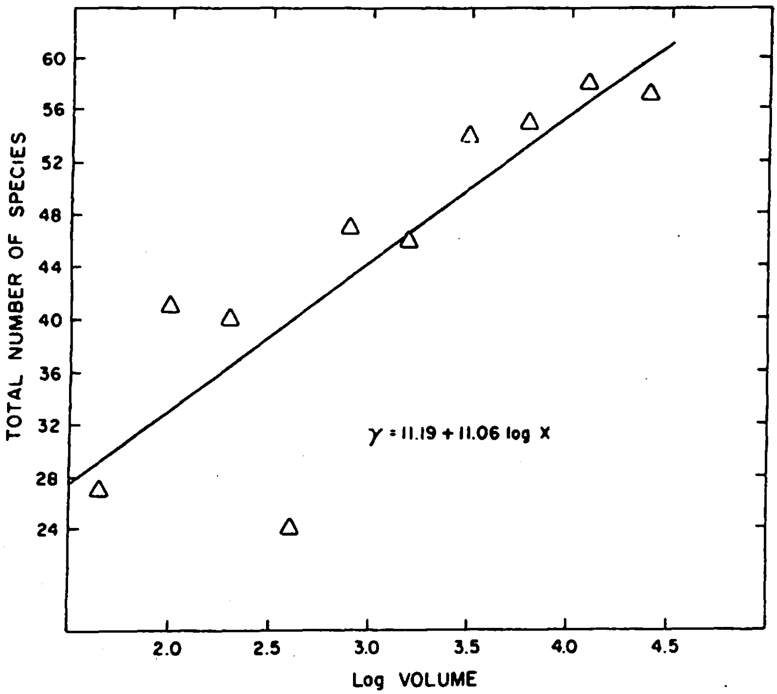


Fig. 3. Relationship between average number of species of freshwater protozoans and log substrate size.

from similar habitat by different, relatively inhospitable terrain traversed only with difficulty by organisms of the habitat patch may be considered an island". Thus a rock in a river, a mud flat, submerged log, or artificial substrate may be considered an ecological island (i.e., most micro-habitats).

Initial island colonization is a non-interactive process primarily influenced by the dispersal capacities and extinction potentials of the colonizing organisms (MacArthur and Wilson, 1963). With the establishment of an equilibrium (asymptotic) species number (Figs. 4 and 5), interactive processes, such as competition and predation, take precedence in determining the island's biotic composition. The island assemblage soon manifests the characteristics of an integrated community capable of maintaining a degree of autonomy with respect to the

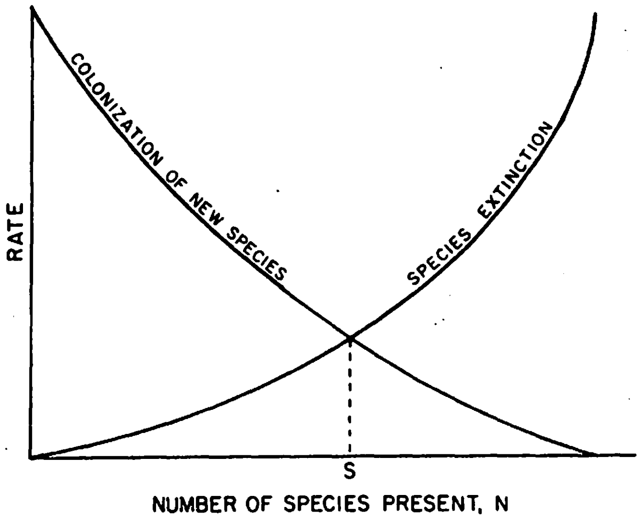


Fig. 4. Basic MacArthur-Wilson model with the equilibrium point(s) of a relatively constant number of species being reached when the colonization rate equals the extinction rate (from MacArthur-Wilson, 1963).

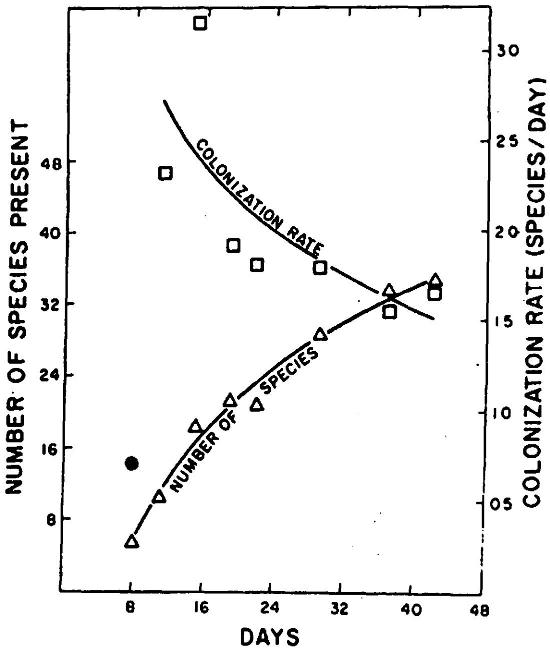


Fig. 5. Colonization of artificial substrates by Protozoa. Notice that the form of the curves is consistent with the predictions of the MacArthur-Wilson equilibrium model. The colonization rate declines as the number of species rises (from Cairns *et al.*, 1969).

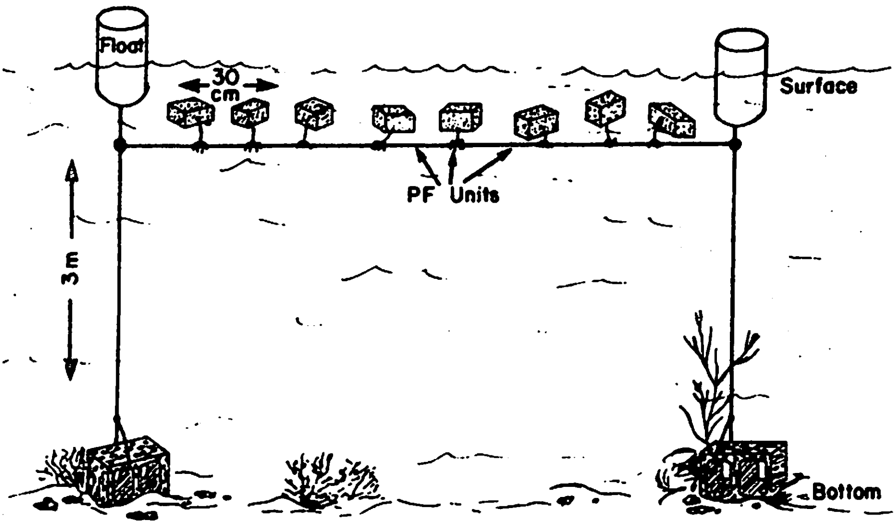


Fig. 6. Typical artificial substrate placement for surface water sampling in a lake or pond (from Cairns *et al.*, 1979).

surrounding environment. Thus, during the non-interactive phase of colonization, artificial substrates or uninhabited natural substrates (e.g. a rock that falls into a stream) act as sampling devices which passively collect organisms from the natural community (Fig. 6). After establishing an equilibrium, the substrate ceases to function directly as a sampling device and the associated species assemblage begins to evolve its own characteristic composition. If an artificial substrate is used as a species sampling device, it should be retrieved from the environment just before or soon after it acquires an equilibrium species number. The appropriate immersion time varies under different environmental conditions.

The question of the influence of time on colonization became especially important following an earlier study (Cairns *et al.*, 1976a) which indicated that location of substrates in a lake was of little importance in determining the outcome of colonization.

The chemical-physical upheaval associated with lake overturn (and presumably other episodic events, such as floods) apparently disrupts the stability of protozoan communities in terms of species richness (Cairns *et al.*, 1976b). It is possible that ecological space again becomes available with the breakdown of competitive mechanisms and non-interactive colonization dynamics take precedence in determining the species' numbers. All species are qualitatively equated with respect to their capacities to occupy the newly available space. Species already inhabiting a particular substrate have an advantage over pioneer occupants. Occupation of the remaining space is determined by a random selection from the available species pool (Gilroy, 1975). Rapid changes in composition have also been observed to occur concomitantly with the spring circulation and other catastrophic perturbations (Jassby and Goldman, 1974).

The disruption of competitive interactions also profoundly affects the species assemblages of the littoral and sublittoral sediments. Unlike the pelagic zone, the primary mechanisms of passive dispersal between the various areas of the sediments include differential sedimentation and turbulent mixing. The latter is probably the only important force affecting dispersal in the open water and is equivalent at all depths, particularly following overturn. Sedimentation, however, does tend to deposit materials (and presumably species) differently within the littoral and sublittoral sediments (Davis and Brubaker, 1973). Therefore, if potential colonists from other areas of the lake are also deposited in greater abundances in these zones, sediment-associated protozoan communities would be expected to exhibit quantitative differences in species numbers, but become somewhat more similar to all other areas after the overturn phase. This appears to be the case.

Of the sediment assemblages, those at the 5 m location most resemble those of the pelagic zone. In fact, they are normally included within the overall compositional cluster for the open water. The implication from pre-overturn that the 5 m location exhibits characteristics of both the pelagic and benthic environments is confirmed. Finally, these conclusions imply that the profundal sediments are inundated with a large number of species, primarily from the pelagic zone which had been previously excluded.

Drastic environmental perturbation, such as lake overturn, causes the partial or complete breakdown of interactive mechanisms (Cairns *et al.*, 1976b). Simple functions of non-interactive colonization assume primary importance in determining the nature of particular assemblages.

Henebry and Cairns (1980a) studied the colonization of artificial islands in a closed laboratory ecosystem using epicentres (source of species) colonized in natural systems. After seven days, the islands with the smallest area or closest to the epicentre had the highest species number. The latter fits the MacArthur and Wilson (1963) model but the former appears contradictory. However, the islands had not reached equilibrium which was the main point of the original hypothesis. Islands exposed to epicentres of intermediate maturity (half-way to equilibrium) had significantly greater species richness than islands tested with mature (at MacArthur-Wilson equilibrium) epicentres. Evidence gathered in this study strongly suggests that the kinds of species present during different periods of colonization are responsible for differences in species richness on islands exposed to epicentres of different maturities. In addition Henebry and Cairns (1980b) supported the hypothesis that colonization rates onto artificial islands were influenced by the maturity of source pools of species and the proportion of pioneer species in the source pool communities. Plafkin *et al.* (1980) found the acquisition of a stable equilibrium number to be more rapid for artificial islands drawing colonists from a species pool in a natural system (mostly lakes) stressed by organic enrichment.

2.3 Succession and Seasonal Change

Succession in protozoan communities is a well established phenomenon (Patrick *et al.*, 1967). Changes in dominance and composition may result from changes in organic loading (Fig. 7, McKinney and Gram,

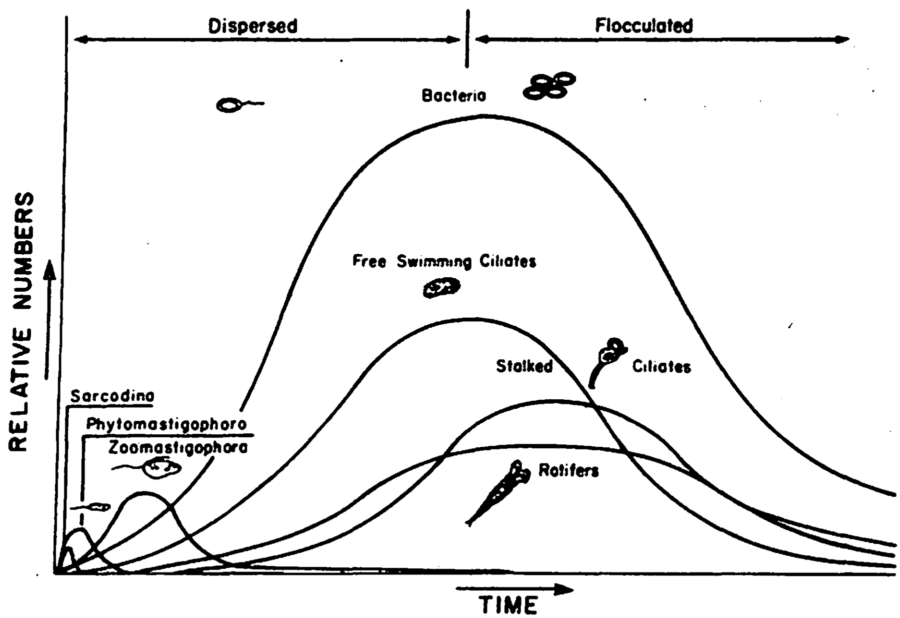
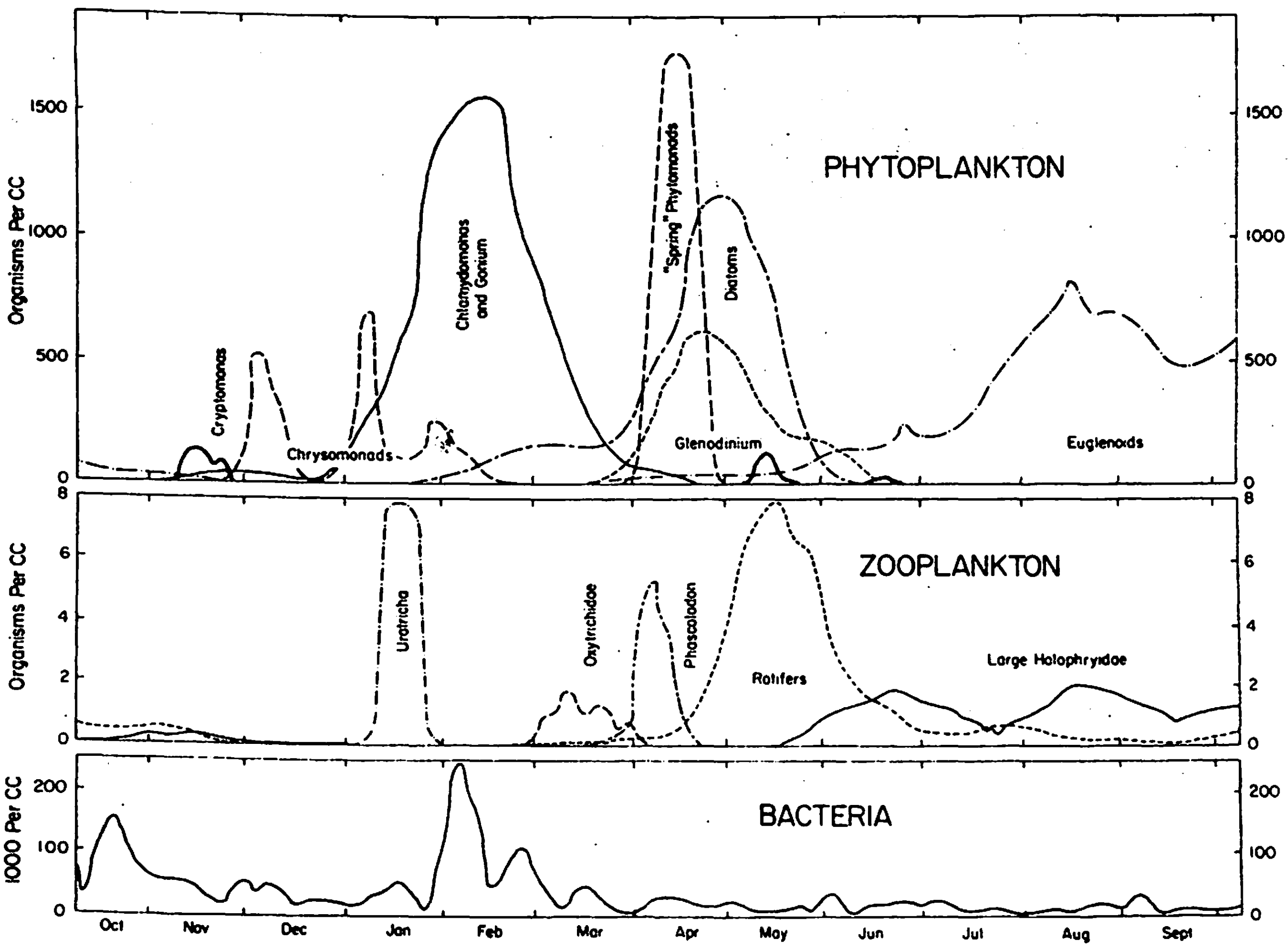


Fig. 7. Relative predominance of microorganisms in activated sludge systems.


PHYTOPLANKTON
Organisms Per CC
1500
1000
500
0
Cryptomonas
Chrysomonads
Chlamydomonas and Gonium
"Spring" Phytomonads
Diatoms
Glenodinium
Euglenoids
ZOOPLANKTON
8
6
4
2
Urotricha
Oxytrichidae
Phascolodon
Rotifers
Large Holophryidae
BACTERIA
1000 Per CC
200
100
Oct
Nov
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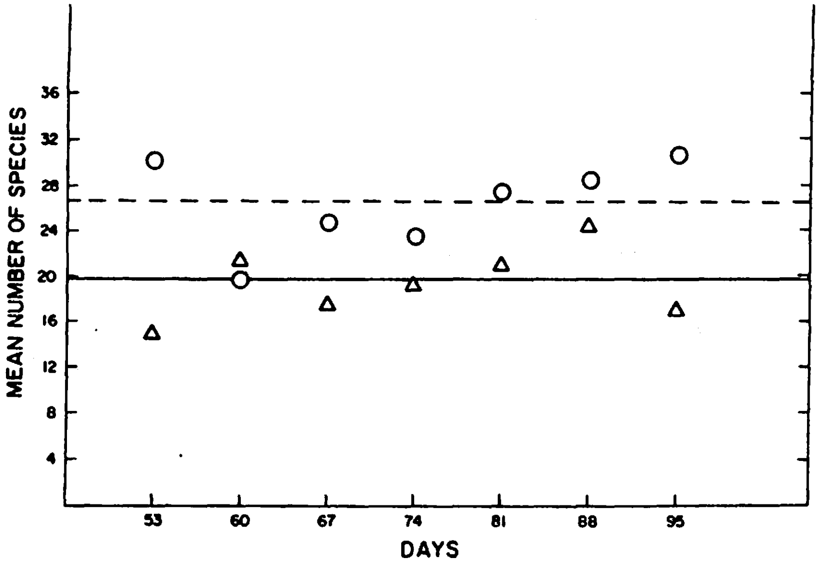

Fig. 9. Weekly mean number of species oscillating about the mean number of species for all substrates which were not previously squeezed (i.e. harvested) (△) and weekly mean number of species oscillating about the mean number of species for all substrates which were squeezed once previously (○).

1967) or in the planktonic community in a small pond as a consequence of seasonal change (Fig. 8, Bamforth, 1958). Flagellates make up the major portion of the early pioneer community becoming established on bare substrates and reaching equilibrium much earlier than the other taxonomic groups (Yongue and Cairns, 1978). However, the number of species or substrates may typically oscillate about a mean once the MacArthur-Wilson equilibrium point has been reached (Cairns *et al.*, 1971a). A segment of a long-term study from the paper just cited illustrates this point (Fig. 9).

◀ **Fig. 8.** Seasonal variation and succession of dominant organisms in a small artificial pond. The curves are smoothed by 15-point moving averages (after Bamforth, 1958).

3. ANALYSIS OF NATURAL POPULATIONS

3.1 Analysis of Protozoan Communities

Often problems arise in identifying species in protozoan communities before the community becomes seriously altered through reproduction, death, and encystment. There is a difference between obtaining the correct Latin name on each species and accurately determining how many taxons are represented. Of course, precision in identification is always desirable but when precision in identifying one species beyond any reasonable doubt means that the community composition of other species is altered before other members of the community are identified, it is not a desirable practice for effective community analysis. That is, precision in identification of a few "difficult" species may mean loss of precision in analysing the community structure. In case this is taken as a license for inaccurate taxonomy, I hasten to add that it is possible occasionally to have the best of both worlds by a combination of preparatory identifications and the use of technniques based on the principles of aeroplane recognition. The method consists of spending a week or so with a more leisurely identification of the species characteristic of the sampling area just before the main analysis begins. Although successional processes and species turnover does occur, many of the species identified in the more leisurely and, therefore, more precise preparatory investigation, will be found for quite a few days thereafter, and perhaps even for the entire period of the investigation. As a consequence, a substantial percentage of the identifications are confirmations of previous identifications and the number of totally new identifications are normally a minority.

Although the practice of fractionating a microbial community into the commonly accepted taxonomic subcomponents, such as protozoans or diatoms, has been deplored, it is necessary to discuss protozoan communities, primarily in a restricted taxonomic sense, by ignoring interactions with other microbial taxonomic groups since this is the standard procedure adopted. It is also worth noting that higher organisms, such as pulmonate snails, can markedly affect attached microbial community structures by selective cropping of species (R. Patrick, personal communication). For planktonic microbial communities, the classic work of Brooks and Dodson (1965) has shown that higher organisms, such as the alewife (an American fish), also affect community structure significantly by particle size discrimination in planktonic feeding. The study of subdivisions of a true microbial community, such as a protozoan community, is not without scientific justification, however, since predictions can be made based on this limited taxonomic array that can be verified and are remarkably consistent.

There is also justification for limiting the scope of a study by the use of the definition due to Whittaker (1975), that a community is "a system of organisms living together and linked together by their effects on one another and their responses to the environment they share". Whittaker (1975) defined an ecosystem as "a community and its environment treated together as a functional system of complementary relationships and transfer and circulation of energy and matter". Patrick (1949) discussed means of assessing the effects of pollution on aquatic communities using sections of streams from bank to bank and several hundred feet along the stream, by collecting samples from all of the common habitats within the system. The well-established species in the samples are identified and the results from all the samples combined to determine the community composition of each of the major groups of aquatic organisms, including algae and protozoans. Since the original study was concerned with pollution effects, breaking the system into discrete study areas seemed a satisfactory procedure to delimit a community. Alternatively, river microbial communities might be viewed as a continuum since it is theoretically possible that the actions of organisms on one bank of a river might have some effect upon a microorganism on the other bank. It is unlikely, however, that such effects could be demonstrated. A protozoan community thus defined (i.e. from bank to bank in a river) provides a useful means of detecting gross pollution effects (Cairns, 1965) but not the more subtle effects. The amount of work involved is substantial since frequently six or more subsamples require identification. One also has to assess the weight given to the subsamples, according to the percentage of the total habitat they are presumed to represent, or whether to measure qualitative changes in species richness without regard to the percentage of habitat represented by the sample from which the determinations were made. This in turn depends on whether interest centres on the qualitative or quantitative degree of the environmental impact under study.

3.2 Response to Changes in Water Quality

Interpretation of the response of protozoan communities to changes in water quality is enhanced by obtaining information on other taxonomic groups of aquatic organisms from the same area and collected at the same time. This is illustrated clearly by the histograms from Patrick (1949) who was one of the first to show the utility of an array of taxonomic groups in analysing pollution effects in freshwater ecosystems (Fig. 10). Each of the histogram columns represents a different number of species, despite the fact that they are nearly the same height in the healthy station. For example, the histogram for fish might represent approximately 35 species at 100%, which would be the average value for a number of healthy stations on unpolluted streams. The insect column that had 100% might represent over 80 species of aquatic insect larvae established at the same number of stations on unpolluted streams. Therefore, the 100% value represents the typical species richness (i.e., number of species present) in unpolluted systems. Note that for the semi-healthy station, the percentage of protozoan species remained about the same as that found in the healthy station, while the percentage of fish species declined substantially. However, the pollution tolerant worms rose by a substantial percentage. For the polluted station the changes were more dramatic: both the pollution tolerant algae and rotifers, and pollution tolerant worms rose significantly. In the case of the pollution tolerant algae and rotifers there was a doubling in numbers compared to the healthy station and the tolerant worms increased by 50%. The fish disappeared entirely and the taxonomically higher animals (rotifers, clams, insecta, and crustacea) almost disappeared. Finally, in the very polluted station, the percentage of aquatic organisms compared to the healthy station was greatly diminished.

It appears that pollution, particularly organic pollution, favours the lower organisms and, for some species, this is true. However, the effect displayed in the histogram series (Fig. 10) was probably due to the result of reduced predation on the lower organisms which permitted the expansion of certain populations, coupled with a direct benefit for some species from the substance causing the pollution. That the loss of the predator species is at least partly responsible for the increase in the columns representing the lower organisms is supported by the progression exhibited from the healthy to the polluted station. Note the rather dramatic loss in the very polluted station of all species, indicating that the pollutants were detrimental to almost all forms of life (except for a relatively few species) when at a high concentration. Analyses of the histograms shows that the number of protozoan species alone may not provide an early indication of pollutional effects as well as for some of the other aquatic organisms (e.g. fish).

In situations, such as the one just mentioned, where the number of species remains constant, there may be a shift in the numbers of individuals per species so that the individuals of a few species could become exceedingly abundant (this would change the diversity index without changing the species richness). A shift in dominance from

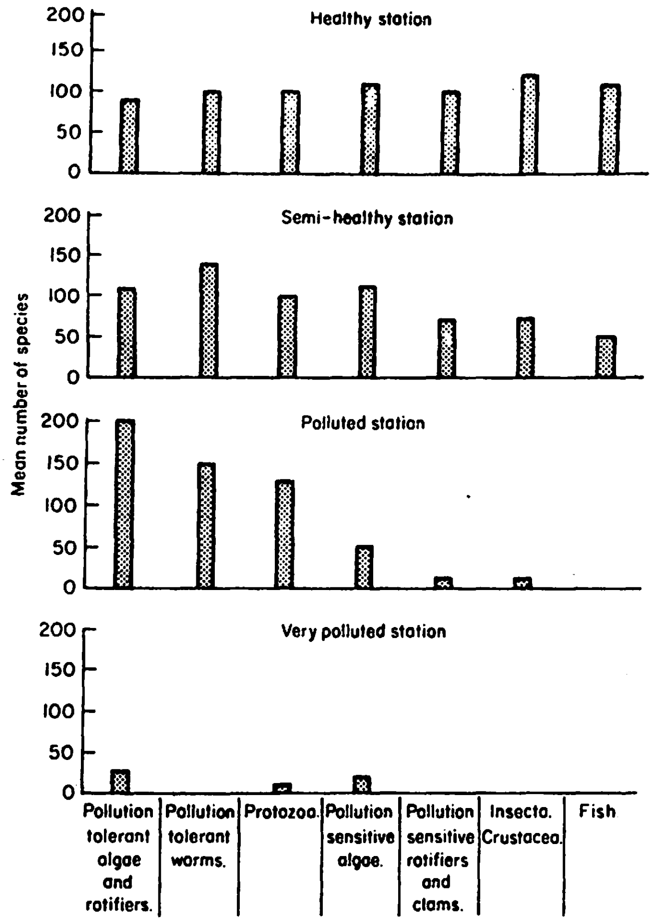

Fig. 10. Histograms illustrating the response of an aquatic community to pollution (from Patrick, 1949).

flagellates to ciliates or some other shift in species composition might occur. Additionally, there might be a shift in species function (e.g. from primarily autotrophic to primarily heterotrophic). Thus, even when using only a single taxonomic group, such as protozoan communities, the pollution investigator would do well to obtain sufficient data so that more than one analysis can be made. If time and financial circumstances permit additional evidence beyond that furnished by the microbial communities, such evidence should be obtained.

3.3 Sampling Natural Communities

The most important problem in sampling natural systems is defining the boundaries of the systems being sampled. For pollution studies in a river or a lake, sampling usually occurs along a gradient from high to low concentration of the offending material. Once the nature of the gradient has been established (and this is not always a simple task), the next consideration is to determine that the types of habitats sampled at each of the locations along the gradient are ecologically comparable to the reference (control) area. Very often a contaminant, such as heated waste water or a hazardous chemical, has thermal or concentration gradients that require sampling in areas where the habitats differ from those found in the reference station or from one part of the gradient to another. In such situations, the comparison may be facilitated by the use of artificial substrates (e.g. Cairns *et al.*, 1979). This ensures that the microhabitats are structurally comparable and that species from this type of habitat are comparable. Artificial substrates can be used in situations where normally only plankton are found because suitable invading species (i.e. those usually associated with substrates) are present in most freshwater situations. Whether this is true or not for marine environments outside the coastal zone has not been determined. Duplication of natural communities by artificial substates does not necessarily occur (although this is usually the result), but determining whether there is a biological stress due to pollution or some other effect differentially exerted from one sampling area to another is essential. If it is assumed that an equal colonization opportunity exists in each area, and that the primary variable is the presence or absence of the pollutant or stress, then comparisons in community structure and/or function that display differences should be considered valid. In pollution assessments, extrapolation from the response of a few organisms to many is usually the case and testing of all the species possibly at risk is rare. It is not possible to test all the organisms that could conceivably be affected. Consequently, testing an array of species through the use of community analysis assumes that the response range will be adequately displayed.

3.4 Sampling Natural Substrates

When analysing protozoan communities, particularly those that are difficult to preserve (most freshwater systems as already discussed), there has to be a choice of how to distribute the analytical time before the fresh sample is seriously altered through death, reproduction, or encystment. A sample from each of the major substrate habitats (i.e. surfaces of mud, rock, submerged vegetation) is examined separately. By coupling this information with the relative proportion of the various microhabitats within an aquatic ecosystem, a very detailed and realistic apportioning of the species distribution within the system and their abundance in the different habitat types can be obtained. The same physical structure (e.g. submerged vegetation or wood) in shady and sunlit situations as well as at different depths and in different current velocities should be analysed. Unfortunately, if one collected as many samples as should be necessary to evaluate properly all the differences just mentioned, the samples would be far too numerous to investigate before serious alteration in community composition began. The errors when the samples deteriorate are far greater than if only enough samples were collected to analyse before undergoing marked alteration. The recording of species occurring at different time intervals (the result of the rapid turnover that occurs in some microbial communities) introduces a serious distortion. Alternatively, composite results may be prepared from a series of samples. Compositing might reduce the apparent abundance of the extremely common species found in certain habitats but not found elsewhere, to the point where they might be below the threshold density that makes recognition possible. Normally it is generally preferable to compromise by taking a series of samples from the most common major habitats (i.e. surfaces of rocks, submerged vegetation or mud) and ignoring some of the finer distinctions previously mentioned. Since these are usually baseline studies carried out before, say, an industry begins discharging wastes, the type of community inhabiting the area in a general way is determined. By comparing the post treatment samples with the pretreatment samples, any major deleterious effects that occur may be determined.

The problem of how many species to take in characterizing the planktonic community is somewhat similar, although, of course, the life histories and ecological requirements of planktonic species are different. Planktonic species have an advantage in that many are comparatively easy to preserve and there is generally less detritus to interfere with sample examination.

4. THE USE OF PROTOZOANS IN THE ASSESSMENT OF WATER QUALITY

Protozoans provide a useful means of assessing water quality but they are not frequently used. Some of the advantages of using protozoans for this purpose are as follows:

(1) They require relatively small containers compared to those needed for fishes and macroinvertebrates.

(2) Some microbial species are no more difficult to handle than rainbow trout and other commonly used test species.

(3) They grow comparatively rapidly so that the effects of potential pollutants on reproduction, growth, metabolism and other properties may be readily tested over several generations without waiting months or years for results.

(4) They can be maintained in synthetic media under defined conditions so that assays are completely reproducible—a situation which is expensive for the large volumes of fluid required for higher organisms and difficult to achieve.

(5) Many species have both sexual and asexual reproduction stages—thus clonal uniformity is available as well as the unique characteristics associated with sexual reproduction.

(6) Unicellular organisms are in more intimate contact with their environment and often have shorter response times than higher organisms.

(7) Since most free-living microorganisms have a cosmopolitan distribution and are likely to be found wherever the natural conditions are appropriate, the same species may be used as an indicator organism on different continents and help to reduce the conflicts due to differing results with different species.

(8) Since many species can often be kept in stock culture at a slow rate of growth, it is possible to keep a collection more easily and in less space than is possible for higher organisms.

(9) The differences in tolerance to various waste materials among fishes, invertebrates, and microbial species is not as great as is generally supposed. Patrick *et al.* (1968) have shown that diatoms are sometimes more sensitive than fishes, sometimes less, and sometimes quite comparable, but rarely are there orders of magnitude differences in response. Thus, fish tolerance is no more or less representative of the entire aquatic community than a microbial or macroinvertebrate species.

(10) Protozoans, together with other microbial species, constitute the major portion of the biomass of many aquatic systems. Therefore, in terms of weight per unit area (or volume), they are frequently a dominant portion of aquatic ecosystems.

(11) A collecting permit usually is not required!

We are now entering an era where the hazard evaluation of toxic substances, biological monitoring, and environmental quality control systems are essential for the maintenance of the quality of life in industrial societies. In addition, such systems are necessary for the protection of human health. Even for situations where the sole intent is the protection of human health, the use of biological early-warning systems involving surrogate organisms will almost certainly become increasingly common in the future (Miller, 1977). As a consequence of widespread use, more careful attention will probably be given to the cost of generating the information. Cost will be partially dependent upon the space requirements of the organisms used in the various types of biological assessments. It is, therefore, highly probable that the use of protozoans for this purpose will increase (Cairns, 1979).

Protozoans may be used as indicators of water quality in three major ways:

(1) in surveys of rivers, streams, lakes, and other bodies of water which are carried out preferably by a team of specialists working with organisms ranging from bacteria to fishes;
(2) in laboratory bioassays designed to determine the effects of various changes in water quality, and
(3) in laboratory microecosystems, artificial streams, and the like which are designed to fill the void between the single species bioassay and the complex, highly variable natural systems (e.g. Cooke, 1977).

4.1 Use of Protozoans in Field Studies of Aquatic Ecosystems

The use of protozoan communities in the assessment of water quality of rivers, streams, lakes, estuaries and oceans has some serious drawbacks and some marked advantages which have been discussed at length by Cairns (1974).

Protozoan communities may be used in a variety of ways for assessments.

(1) They may be used as indicator species as exemplified by Bick (1971).

(2) They may be counted to give the number of species and the number of individuals per species, and this data can be analysed by cluster analyses, principal component analyses, ordination, and other procedures. Either natural substrates (Lackey, 1944) or artificial substrates (Spoon and Burbanck, 1967) may be used. Artificial substrates offer some advantages in this type of sampling. Firstly, the substrate may be positioned in the best locations relative to a waste discharge rather than depending on locating the natural habitat. Secondly, the species found are usually the same as those on natural substrates, which is expected since this is where the invaders of the artificial substrate

originate. Thirdly, the sampling process is simplified since composite results from a variety of substrates of dissimilar composition and location, in the proportion in which they occur in nature, do not have to be determined since the proportionality is to a certain extent taken care of by the invasion and subsequent colonization processes.

Examination of microbial communities is normally not done on artificial substrates during the early stages of the colonization process and often not until they are presumed to have reached a MacArthur-Wilson equilibrium condition. However, there is a strong possibility that the colonization process itself may be more informative than the equilibrium condition. This is demonstrated by Fig. 11 from a study of the protozoan communities on artificial substrates for six stations at Smith Mountain Lake, Virginia (Cairns *et al.*, 1979). The arm of the reservoir containing sampling stations 1 and 2 receives sewage and heavy metals from the city of Roanoke, Virginia, just above station 1 (Roanoke River Basin Comprehensive Water Resources Plan, 1975). This produces a well-defined eutrophic gradient with the arm of the reservoir containing station 1 representing the organically enriched situation with some water quality recovery at station 2 and a more marked recovery at the confluence of the two arms represented by stations 3 and 4. Stations 5 and 6 are on an unpolluted section of the reservoir. Although the equilibrium species number (day 21) is quite similar for all stations, the early colonization species number (days 1 and 3) is much higher for the polluted stations (1 and 2) than at the unpolluted stations. Smith Mountain Lake is not a badly polluted lake: if it were, this would be reflected in the species richness at equilibrium. The fact that it is in a threshold condition in certain areas and that the differences between stations under these circumstances are best determined during colonization occurring before equilibrium than after is worth further attention.

Ecological perturbations, such as organic pollution in an aquatic environment, generally produce certain predictable changes in community structure. Species with low tolerances are eliminated, while those species best suited for survival in enriched habitats become excessively dominant. Fig. 12a illustrates two hypothetical distributions of species into abundance classes. Stress distorts the normal distributi n U by eliminating many low to moderately abundant species and in reasing the number of high abundance species. The net result is that in a stressed situation (distribution S) a greater proportion of the total species are present in high abundance. Such changes in community structure must affect the colonization dynamics of initially barren islands drawing colonists from the perturbed system.

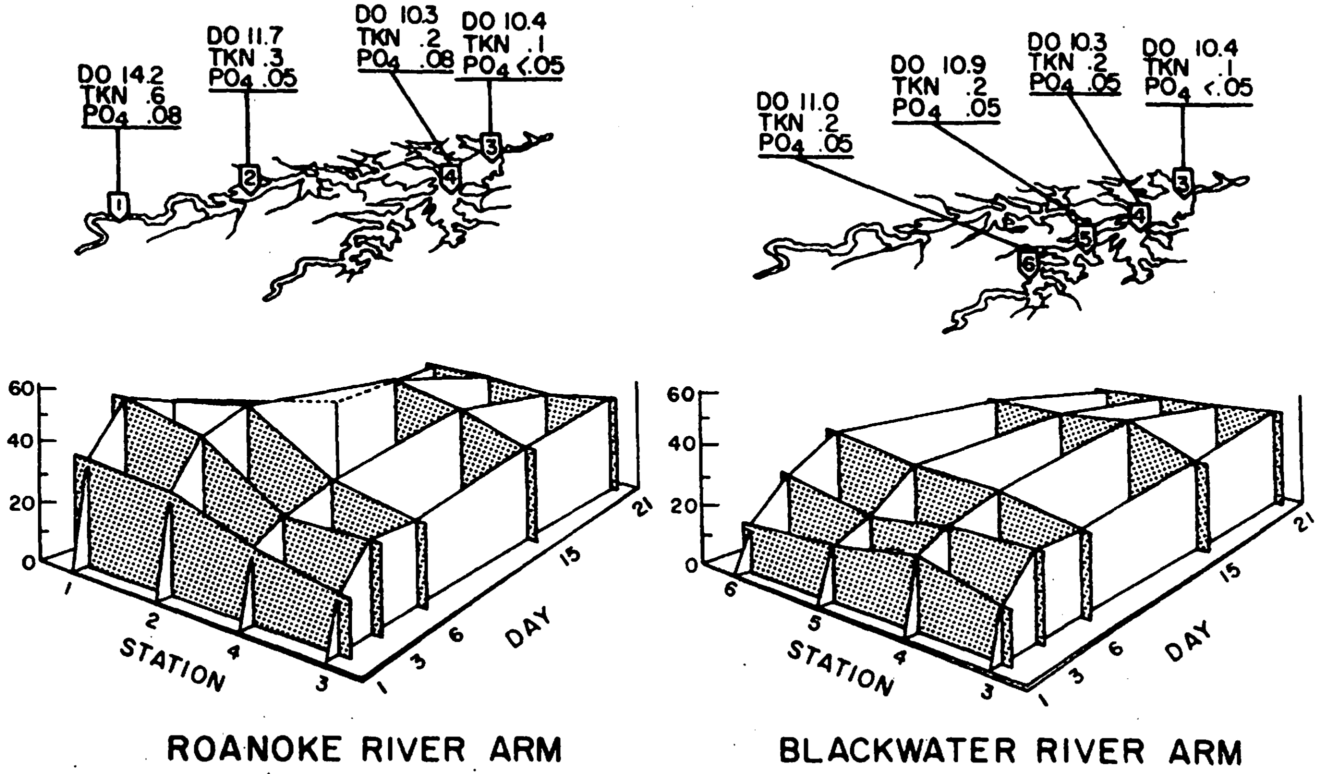

Fig. 11. Mean colonization curves showing the protozoan colonization of artificial substrates in Smith Mountain Lake (a reservoir) Virginia (after Cairns *et al.*, 1979). See text for details of stations 1 to 6. DO = dissolved oxygen concentration; TKN = the nitrogen concentration; PO_4 = the phosphate concentration.

There are four reasons to expect that the establishment of a stable equilibrium number would be more rapid for uncolonized islands (in this case polyurethane (PF) islands) drawing colonists from a stressed environment.

(1) In organically enriched areas, colonization is rapid because the PF islands supply immediately suitable habitats for immigrant heterotrophs. Under comparatively oligotrophic circumstances, however, colonization is slower because preparation of the substrate is first required before many protozoan species can invade successfully.

Gilroy (1975) noted that in defaunation experiments (Wilson and Simberloff, 1968) "non-ideal" islands (i.e. those which did not reach an equilibrium during the first year of the monitoring period and did not fit the theoretical model) had suffered considerable damage during the defaunation procedure. Recolonization was very slow because the suitability of these islands as habitats had been significantly diminished.

Foam islands at the cleaner Smith Mountain Lake sampling stations 3 and 4 were also comparatively slow to accumulate species. The initial phases of colonization at stations 3 and 4 were adequately described by the non-interactive model (Table 1, days 1 to 6), but the apparent acquisition of equilibrium by day 6 and the subsequent increase in species numbers suggested that habitat islands were not immediately suitable for certain components of the community. Because habitat islands at the cleaner stations are not subject to the large and immediate influx of organic nutrients which abound at more polluted stations, colonization and habitat preparation by early invaders was probably first required to modify these habitats for subsequent successful invasion by other heterotrophic protozoa.

(2) A stressed community is commonly dominated by r-selected opportunistic species. These species are particularly well adapted to the pioneer stages of colonization when resources are plentiful and competitive pressures are minimal (Luckinbill, 1979). Barren islands drawing propagules from this type of pool should accumulate species more readily than islands drawing from a more complex source.

Opler *et al.* (1975) have observed a situation analogous to this in the tropical lowland forests of Costa Rica. Recolonization of clear-cut plots drawing propagules from severely perturbed source areas was extremely rapid. This was also attributed to the relatively large numbers of pioneer species within this pool. Plots surrounded by more mature forest, however, exhibited much slower increases in species richness within an equivalent time frame.

(3) Since island community establishment is considered an essentially non-interactive process (Simberloff, 1969), it can be simply

Table 1. Nonlinear regression analysis of model $S = \hat{S}_{eq}(1 - e^{-GT})$ in 6 Smith Mountain Lake stations. Lack of Fit (L.O.F.) F and α level attained are presented ($\alpha[F] < 0.01$ is required for decision level). Estimates of the parameters are given where model was adequate ($t_{99\%}$ is time to reach 99% of $\hat{S}_{eq}$ the equilibrium species number).*

		Station 1		Station 2	Station 3	Station 4	Station 5	Station 6
			w/o 15†					
All data	F	6.97	0.622	1.403	27.34	45.78	4.98	1.078
	α(F)	0.0086	>0.50	>0.75	≤0.001	≤0.001	=0.023	>0.75
	$\hat{S}_{eq}$	52.04	55.12	58.72	LOF	LOF	48.26	48.47
	G	1.64	1.42	0.89			0.31	0.40
	$t_{99\%}$	2.81	3.24	5.17	(no equilibrium)		14.85	11.51
Days 1 to 15	F	—	—	—	12.92	86.36	—	—
	α(F)	—	—	—	0.004	≤0.001	—	—
	$\hat{S}_{eq}$	—	—	—	—	—	—	—
	G	—	—	—	LOF	LOF	—	—
	$t_{99\%}$	—	—	—	—	—	—	—
Days 1 to 6	F	—	—	—	7.43	11.84	—	—
	α(F)	—	—	—	0.037	0.017	—	—
	$\hat{S}_{eq}$	—	—	—	32.29	35.59	—	—
	G	—	—	—	1.99	1.14	—	—
	$t_{99\%}$	—	—	—	2.31	4.04	—	—

* Reprinted with permission from Cairns *et al.* (1979).

† Without Day 15.

viewed as a sampling phenomenon whose dynamics are a function of the source species distribution.

If the dispersal capacities of the organisms are similar which is likely to be the case for passively dispersed protozoa in a benthic system (Kuhn and Plafkin, 1977), then the likelihood of any particular species invading the substrate is directly related to its abundance: the greater a species' abundance, the greater its likelihood of colonizing. If samples are taken at an early time (T1, Fig. 12b), a greater proportion of the total species pool would be sampled from the stressed versus the unstressed distribution. As the sample time increases (T2, Fig. 12c), the likelihood of a substrate receiving new species from low abundance classes is increased. Eventually, as the sample time is expanded further, a point of diminishing return is reached where continued exposure draws in fewer and fewer new species. This point is reached sooner in the stressed situation than in the unstressed where there is a greater proportion of low abundance species (T3, Fig. 12d).

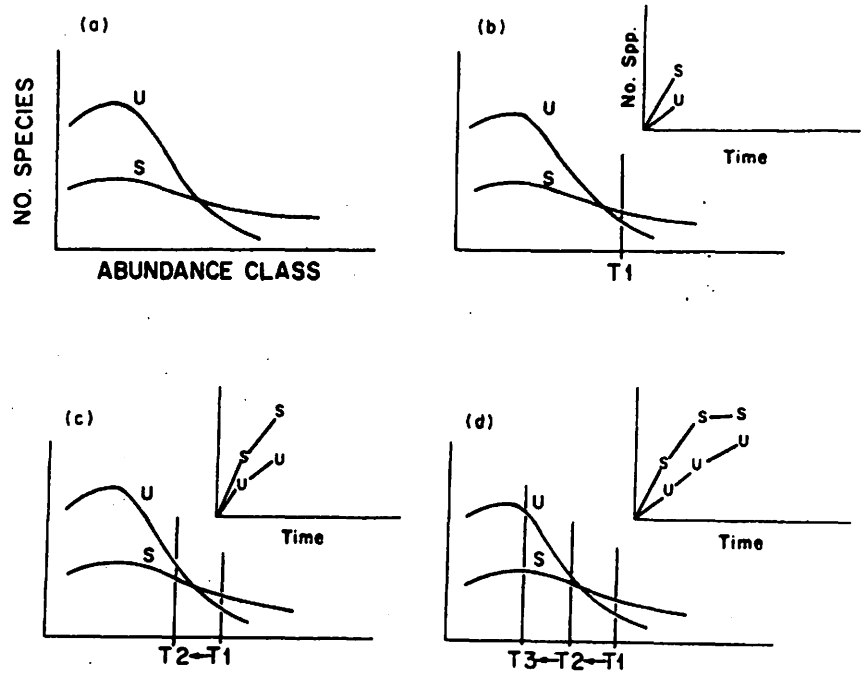

Fig. 12. Distortion of relative abundance structure by stress (from Cairns et al., 1979).

(4) Another direct result of organic pollution is to increase the carrying capacity of the habitat in question for heterotrophic organisms. Fertilization can be visualized as causing a displacement of the mode of the effected species pool distribution to the right (Fig. 13). This displacement effectively increases the number of high abundance species at enriched locations, potentially speeding substrate colonization even more than the distortions of relative abundance structure which have just been considered.

The protozoan colonization of barren habitat appears to be a direct function of the characteristics surrounding natural protozoan community. The process reflects characteristics of both the composition and productivity of the source pool.

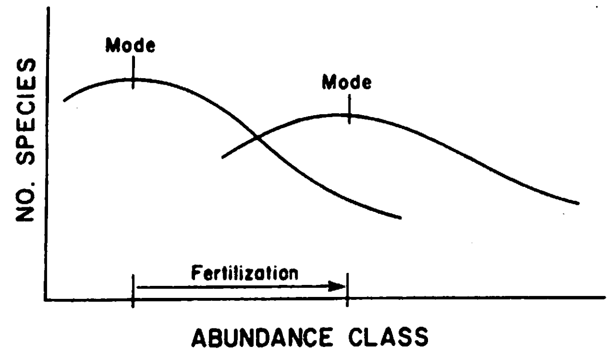

Fig. 13. Fertilization shifts the mode of the relative abundance distribution to the right, reflecting the increased carrying capacity of the enriched habitat. A hypothetical distortion of the curve's shape in response to pollutional stress is also illustrated (axes as in Fig. 9). (From Cairns *et al.*, 1979.)

4.2 Laboratory Bioassys

A variety of laboratory bioassay techniques using protozoans to determine the toxicity of chemical substances has been developed (Bovee, 1975; Bringmann and Kuhn, 1959; Butzel *et al.*, 1960; Gross, 1962; Gross and Jahn, 1962; Bick, 1971; Mitchell, 1972; Maloney and Palmer, 1956). It is worth noting that some Protista organisms may be referred to as either algae or protozoans. Some of the bioassays involved protozoan communities (Cairns and Plafkin, 1971); others involve single species (Yongue *et al.*, 1979); some involve inhibition (Kostyaev, 1973); others depend on direct lethal effects or the disappearance of members of a community (Cairns *et al.*, 1971b). Schultz and Dumont

(1977) exposed the protozoan *Tetrahymena pyriformis* to phenol and found that concentrations of less than 75 mg l^{-1} alter cell motility, shape, and contractile vacuole activity. After 3 min exposure to less than 10 mg phenol l^{-1} reduced the oxygen uptake with a concommitant increase in electron density of the mitochondrial matrics. Alterations in mucocysts, pellicle, and glycogen were also observed. Bringmann and Kuhn (1959) and Epstein *et al.* (1963) also devised tests for the protozoan *Microragma*.

A comparison of the protozoan response to phenol with some other commonly used aquatic test organisms demonstrates that the protozoan response is within the range of the response of other organisms. For example, Alekseev and Antipin (1976) found an LC50 value of 320 μg phenol ml^{-1} for *Physa fontinalis* and Bringmann and Kuhn (1977) report an LC50 for *Daphnia magna* of 31 μg phenol ml^{-1}. Additional evidence of the response relationship may be found in the literature (Buikema *et al.*, 1979) and a general discussion may be found in Hutner (1964) and Hutner *et al.* (1965).

The paucity of toxicity tests on protozoan communities forces estimation of the concentrations that will not impair community integrity. There are a number of drawbacks to this approach.

(1) Interactions between and among species are ignored.

(2) As the level of organization increases (i.e. species, community, ecosystem), properties emerge that were not apparent at lower levels (e.g. energy flow).

(3) Detoxification is more likely in a complex system as is disappearance into an environmental sink (e.g. sediments). Cairns *et al.* (1980) found that a sublethal dose of copper sulphate significantly decreased the colonization rate of uncolonized substrates in association with both mature (at MacArthur-Wilson equilibrium) and immature communities (not at MacArthur-Wilson equilibrium). The effects were more pronounced when immature communities were involved.

4.3 Microcosms

There is a vast chasm between single species laboratory tests in which variables are controlled and the highly variable complex natural ecosystems where they are not. Clearly, a system of intermediate complexity with more control over variables is quite useful for a variety of research endeavours, including the verification of predictive models developed either from single species laboratory or natural system data. Some of these are "species defined" gnotobiotic systems (e.g. Taub, 1969; Crow and Taub, 1979; Taub and Crow, 1981; Taub *et al.*, 1981), but many are not. Microcosm is defined as "a little world: a miniature universe." It is, in the context of this chapter, a patch of an ecosystem lacking many of its important characteristics but, if one has chosen the patch carefully, it exhibits the characteristics one wishes to study reasonably well (see Parkes, pp. 45–102). The recent literature on methods in this field has been summarized by Cooke (1977) and Salt (1971). Although investigations with microcosms are not yet widely established, it appears to be a very promising approach.

5. THE FUTURE OF RESEARCH ON PROTOZOAN COMMUNITIES

Although there is certainly no evidence of widespread interest in protozoan community structure, there are good reasons for believing that such interest will develop in the near future. A broad interest presently exists among biological scientists in the ways in which communities function and are structured. This interest seems to be growing quickly and some fundamental questions about community organization now seem to be resolvable given computer technology as an assistance in handling the masses of data necessary. The decline in federal and state support of university and private institute research, coupled with increased difficulties both in transportation and political unrest of getting to remote locations from the home base, make it likely that studies of the larger vertebrates and vascular plant communities may be partly replaced by studies of microbial communities where the hypothesis being tested is ammenable to such use. There is no intent to imply that hypotheses involving microbial communities for their own sake are not desirable, but rather that microbial community research may be a suitable surrogate for research on communities of larger organisms for which funding may not be as readily available as it has been in the recent past. Even if research with the larger systems is contemplated, it might well be advantageous to carry out some screening studies with smaller systems to define more precisely the parameters to be measured and the questions to be asked.

Three very important and essentially unanswered questions regarding the response of communities to stress illustrate this point.

(1) Are the single species toxicity tests useful for predicting responses of entire communities? Does an application factor derived from tests carried out on a single species actually protect an entire community?

(2) Do communities in different areas made up of different aggregations of species respond in a similar way to an identical concentration of a toxicant? In this instance one might have to allow for differences in water quality that affect toxicity.

(3) Do communities of different maturity respond similarly to identical concentrations of a toxicant?

The primary assumption stimulating question (1) is that the commonly used test species, such as the rainbow trout, the white rat, the guinea pig, the bluegill, and the rabbit, will furnish evidence from which extrapolations can be made that will protect all other organisms not tested. They are thought to furnish a reasonable representation of the range of biological response. Differences between the customary test species and those of other species not included in the testing procedure are thought to be adequately predicted through the intelligent and knowledgeable use of existing information. Questions (2) and (3) are fairly straightforward.

Protozoan communities have been useful in:

(1) Testing hypotheses in theoretical ecology.
(2) Biological monitoring of pollutional effects.
(3) Toxicity testing.
(4) Evaluating water quality.

It is curious that they are not used more frequently considering the many advantages mentioned in the text. There are only two major drawbacks:

(i) Skill in identification is not easily acquired, and
(ii) Preservation usually distorts the community structure and there is a loss of some taxonomic characters even for the more durable species.

These seem comparatively minor when one considers the many benefits.

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Management options for rehabilitation and enhancement of surface-mined ecosystems

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ABSTRACT

There are four basic management options for surface-mined land: (a) *restoration* to original condition; (b) *rehabilitation* of some desirable characteristics; (c) development of *alternative ecosystems* that may be quite unlike the original but may be desirable for a variety of reasons; (d) *neglect* or *natural reclamation* when evidence suggests that unaided natural processes will produce better results than human intervention. Checklists are provided so that essential information will be available in selecting the most suitable option.

INTRODUCTION

A few years ago,[1] I examined the pros and cons of various management options for reclaiming surface mined land. The options examined were:

1 Doing nothing and leaving the land as it was when mining was completed.
2 Restoring it to its original condition.
3 Reclaiming it to an ecologically improved (compared to its present state) and more socially acceptable condition of use or aesthetically pleasing.

In this paper,[1] I speculated that 'it seemed virtually certain that partial restoration to an ecologically improved and more socially acceptable condition is the option that will be most regularly exercised in many countries'. Included in this option were rehabilitation and alternative ecosystems that will be discussed at length in this article. Rather than repeat the analysis given in that paper, an ideal goal would have been to develop the beginnings of a decision-making protocol similar to a hazard protocol[2] and to explore at greater length some problems associated with rehabilitation or production of alternative ecosystems, both of which will be subsequently defined. A protocol used in this particular sense is designed to provide a systematic basis for generating information and identifying the kinds of information needed to make a management decision. Most protocols are arranged such that the evidence is gathered sequentially and checkpoints allow determination of a sufficient body of evidence to make a sound judgment in each particular instance. The underlying philosophy is that no two decisions require precisely the same kind of quality of information. The description of protocols commonly used in other management decisions, such as risk analysis and hazard evaluation, are readily available[2,3] so that detailed description in this article seems inappropriate. At the outset, the general strategy of the protocols developed for hazard evaluation (in which development of an evidence base alternates with a decision-making requirement[3]) seemed possible. However, after a number of unsuccessful attempts, this approach was abandoned in favour of a series of checklists. This does not mean that the protocol is impossible or undesirable for reclamation, but that more experience is necessary to produce a sound protocol. The field of hazard evaluation for environmental exposures has only recently been developed and was preceded by a long period of methodology development in toxicity testing, chemical transformation, partitioning and fate analysis, and the other components of the complex information base necessary for reliable hazard evaluation. Therefore, the field of ecosystem rehabilitation is not likely to become similarly systematised until a unifying theme is identified.

The words *reclamation, restoration* and *rehabilitation* are often used interchangeably in the literature. For example, Rorslett[4] cites Björk: 'Restoration of freshwater systems is a well-established practice in Scandinavia. By this we mean the rehabilitation of ecosystems which have been seriously impaired by human modification of the water system or its surroundings (*cf.* Björk, 1975)'. In this manuscript, an attempt will be made to use terminology consistently. However, in citing other publications (including some of my earlier works), the words must be those of the authors. It is worth noting that the words *species* and *religion* are not yet defined with a precision acceptable to everyone, but reasonably effective communication is still possible.

BASIC MANAGEMENT OPTIONS

Four basic management options exist: *(a)* restoration, *(b)* rehabilitation, *(c)* alternative ecosystems and *(d)* neglect or natural reclamation. These alternatives are shown in Figure 1.

Restoration as a policy returns the ecosystem in a direct route toward its initial state. Undesirable features of the initial natural state presumably would be accepted as part of the overall package.[5] Rehabilitation may be defined as a pragmatic mix of non-degradation, enhancement and restoration. The term *alternative ecosystems* has been substituted for *enhancement* as originally defined in the article just cited[5] because the term was applied solely to the Great Lakes, and *alternative ecosystems* seemed more appropriate when generalising ecosystems. In addition, some students have a hostile reaction to the word *enhancement* which they regard as a

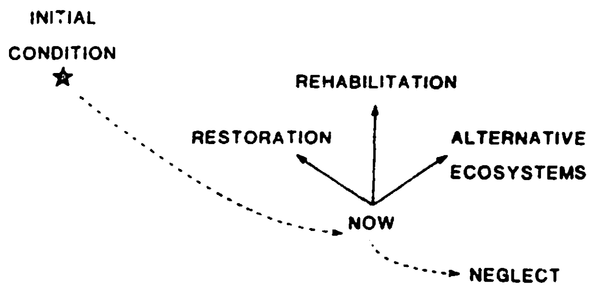

Figure 1 *Management options for surface-mined lands. Modified from Magnuson* et al.[5]

cover-up for industry's failure to act responsibly. Magnuson *et al.*[5] define *enhancement*: 'Enhancement or improving the current state of an ecosystem without reference to its initial state might lead an ecosystem further from its initial state, perhaps by adding desirable man-made features and suppressing undesirable natural features'. I am substituting *alternative ecosystems* to include all possible options that are now or might become available in the future. For example, one might replace a grassland ecosystem in Kansas, that had been surface-mined, with a lake because the area lacked standing surface waters. A well-managed lake might be regarded as more desirable than the original condition, and certainly more desirable than the present condition. Alternatively, one might have prairie grasslands in Southwest Virginia as an alternative to restoring the original slopes of surface-mined areas. Finally, one might create artificial wetlands for disposing of treated sewage and more rapid recharge of groundwater as an alternative to discharging directly into existing streams or lakes.

I am substituting the term *neglect* (which may result in *natural reclamation*) for *further degradation* in the original Magnuson *et al.* article[5] for several reasons. In their article focusing on the Great Lakes, they indicate that 'further degradation, more or less consistent with the degradative processes of the past two centuries, leads in the opposite direction from restoration'. However, abandonment or neglect for some ecosystems might result in a restoration to original condition, an alternative ecosystem, or further degradation. In those cases where a good information base exists, the outcome might be predicted with reasonable certainty even if no management intervention occurs.

PERTURBATION-DEPENDENT ECOSYSTEMS VERSUS PERTURBATION-INDEPENDENT ECOSYSTEMS - THE KEY ISSUE FOR SELECTING A MANAGEMENT OPTION

Although some question remains whether perturbation dependency is only a difference in degree, there is no question that some ecosystems require at least certain types of disturbance in order to maintain high species diversity, productivity and other presumably desirable characteristics.[6] Figure 2 indicates that such ecosystems decline when not perturbed by such naturally occurring events as fires, floods, hurricanes and so on. The degree to which this dependence enhances ability to recover from man-made disturbances is not well documented, but it seems reasonable that perturbation-dependent ecosystems should recover more rapidly from certain disturbances than perturbation-independent ecosystems.

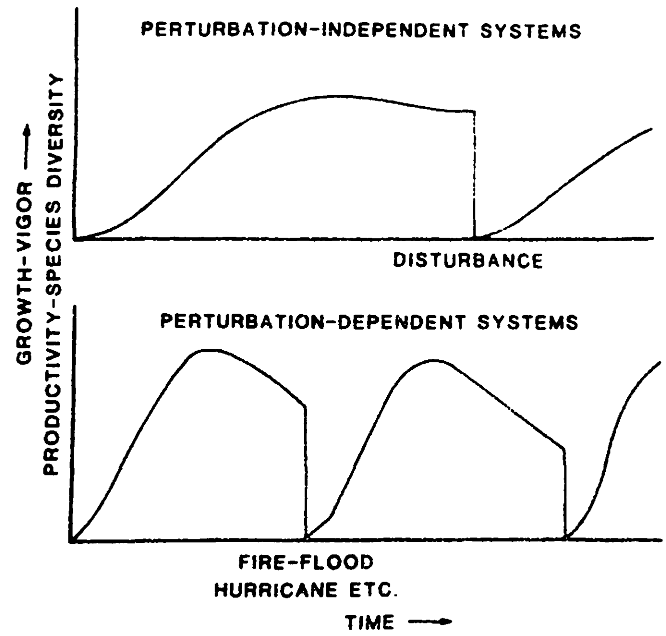

Figure 2 *Disturbances in general ecosystems create vegetational setbacks and complete recovery is slow, whereas disturbances in perturbation-dependent ecosystems usually stimulate pulses of growth which rapidly decline unless disturbed again*[6]

SELECTING MANAGEMENT OPTIONS

Probably the most crucial consideration in selecting an appropriate management option for land that has been surface-mined is whether the system is perturbation-dependent or perturbation-independent (Figure 3). Although some well known examples of both pertur-

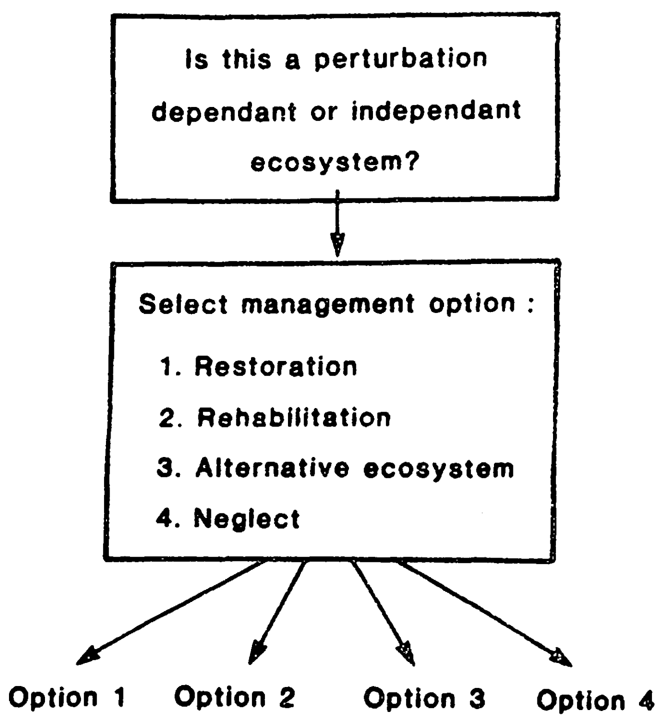

Figure 3 *Selecting management options*

bation-dependent and perturbation-independent ecosystems have been described in detail elsewhere,[6] an example of a perturbation-independent system would be the Amazon rainforest and a perturbation-dependent system would be a typical prairie grassland where fire keeps trees and other wooded vegetation out. Some pine forests are also perturbation-dependent, and some pine seeds require a heat stimulus for proper seeding and germination. If the ecosystem is perturbation-independent, neither the necessary information nor methodology for restoring it to its original condition may be available, particularly if the size of the disturbed area is substantial (since size increases the distance to living vegetation). Additionally, the time required to restore it to its original condition may be well in excess of a human life time, and society is often impatient with long-term responsibilities. Finally, there is no guarantee that the disturbance will merely push the system to an early successional stage and that the successional events that follow will replicate the successional events that led to the present condition. Although the body of literature on succession is quite large,[7] one cannot expect that the assumption just made would be the inevitable result of either the response to disturbance or the recovery process. As a consequence, long-term management responsibilities seem highly probable when the decision is to restore an ecosystem to its original condition.

The merits of creating major disturbance in natural parks, wilderness areas, unique habitats, etc., are not at issue here, but it is certainly possible the decision to utilise these resources may some day be made by the US government. If this is the case, attempts to restore to original condition would seem to be appropriate since presumably the systems were given their present designations because they were in some way unique. These unique properties should not be lost permanently to society. Therefore, despite the difficulties of time, lack of knowledge, methodology, and so on, some circumstances can be imagined where restoration to original condition might be mandatory.

Perturbation-dependent ecosystems might be restored to original condition quite rapidly but may present as many difficulties as perturbation-independent ecosystems. Knowledge of the degree of the perturbation-dependence or independence would also be essential for determining whether or not rehabilitation of certain desirable qualities is possible. Finally, some systems that are perturbation-dependent might be neglected because natural processes will enable them to recover just as quickly as any presently available management practices. Rickard and Cushing[8] have shown that merely excluding livestock from certain areas results in recovery that appears to be identical to original condition in a relatively short time (less than 20 years). Although not explicitly stated, no management practices, such as reintroduction of young plants, would probably have made a major difference in this natural recovery process.

At the present time, scientific, management, social and economic reasons exist for not always restoring to original condition. As the scientific basis for recovery and restoration to original condition becomes better understood, undoubtedly the frequency of selection of the other options will alter substantially. A time may come when restoration to original condition is the most probable option that will follow almost any disturbance. Probably the option of rehabilitation of the system (as defined by Magnuson *et al.*[5]) will also increase more quickly as an important option, but for the same reasons. As the present time, not enough is known about ecosystems to be able to restore desirable features easily except the production and/or establishment of certain species. Although some valuable conservation sites have arisen in abandoned, i.e. neglected, chalk quarries without any human intervention, this does not mean that scientists can determine when this will happen and when it will not. Also, except for small disturbances or ecosystems that recover with astonishing rapidity from displacement, neglect will probably not be a viable option. Therefore, our most likely short-term choice will be the development of alternative ecosystems.

As mentioned earlier, decision-making protocols were constructed for various management options that were similar to hazard-evaluation protocols. Dissatisfaction with all of these led to the production of checklists for the selection of management options. It is worth noting that checklists for toxicity testing and determination of the environmental concentration of chemicals preceded hazard-evaluation protocols. Presumably in every field where science and management decisions interact, science must progress to a certain degree before the production of useful protocols becomes possible. However, Bradshaw and Chadwick[9] do have an operational protocol in their superb book *The Restoration of Land* (Figure 4).

The decision-making protocols alternate data gathering and decision-making based on the assumption that various decisions (such as risk or hazard) do not all require the same amount or kind of information. The protocols are usually arrayed so that decisions to take action may be made at several points along the information gathering sequence. When not enough information is available, the kind and amount needed are explicity stated.

Since this approach has been effective for a variety of decisions, it will undoubtedly work for selecting and implementing options for management of surface mined lands. A series of checklists follow for each of the options. Readers will undoubtedly be able to add to or modify these checklists, both for specific sites and general use. The main purpose of the checklists is to bring order into the decision-making and data-gathering processes so that the methods and information presently available can be fully utilised and deficiencies in the science can be rectified. Decision-making protocols similar in strategy to those developed for evaluating hazard to ecosystems should ultimately be developed.

CHECKLIST FOR SELECTION OF MANAGEMENT OPTIONS

The purpose of this checklist is to ensure that adequate data are available before a decision is made:

1 Can any options be excluded because of low probability of success?
2 Is the cost of any option (including cost to society) prohibitive?
3 What time will be required to reach each of the goals?
4 Is further disturbance likely to occur on the site?
5 Can management responsibilities and/or costs be transferred to another group? If so, should this be

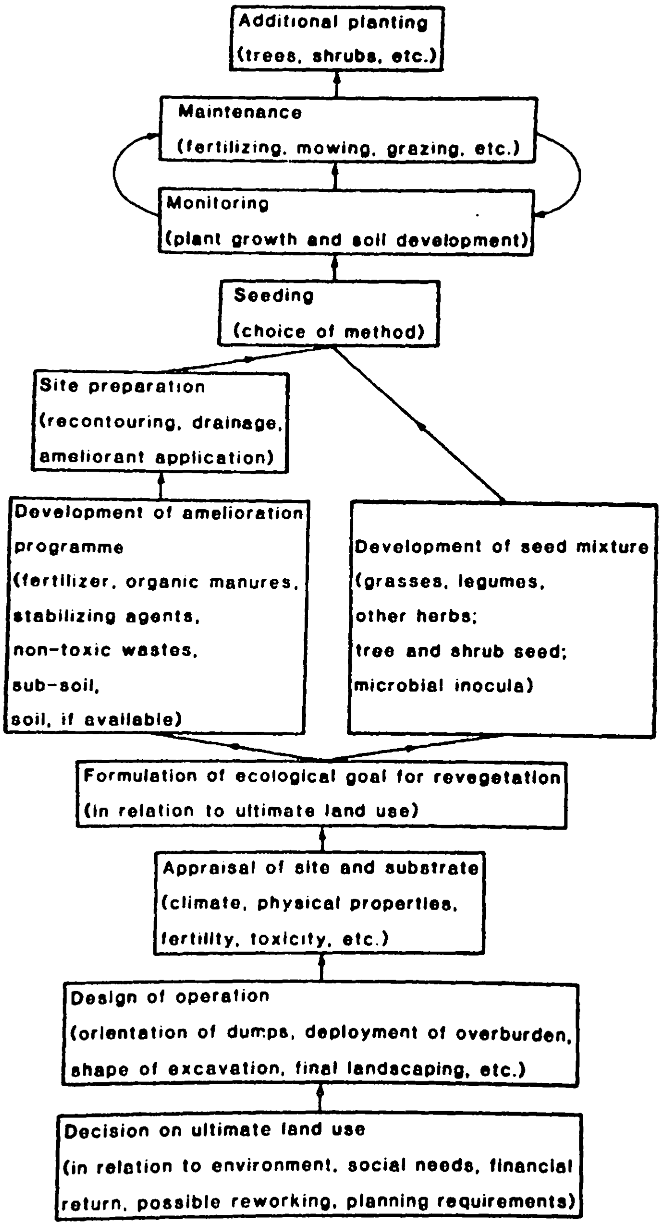

Figure 4 *The steps involved in the development of a successful restoration scheme: at each step careful observations and experiments must be made to ensure that the operations are planned correctly. Reproduced with permission of the authors and the University of California Press, Berkeley. Bradshaw (personal communication) credits Dr Jeffries in Trinity College, Dublin, with the original idea – this is also indicated in his acknowledgements*[9]

done? Often responsibility is transferred over the objection of management. Perhaps management would make more sound environmental decisions if it could retain control.

6 Which option conforms most closely to existing regulatory requirements?
7 What are the comparative costs for monitoring performance?
8 Is cooperation with third parties possible?

OPTION 1: RESTORATION CHECKLIST

1 Is restoration to original condition feasible?
2 Is adequate ecological information available about the original condition?
3 Is a reference ('control') site available?
4 Is the source of original species adequate?
5 Are management responsibilities (including performance monitoring), cost estimate and duration described?
6 Was the original ecosystem of local, regional, or national importance? Can societal response to this option be estimated accordingly?
7 Is there a superior ecosystem or land use other than the pre-mined condition?

This option should be selected: *(a)* for ecologically unique ecosystems, *(b)* for aesthetic reasons where the damaged area is part of a larger system of major recreational or ecological value, *(c)* as a precondition for surface mining in unique and irreplaceable ecosystems. In few cases will this be a viable option if substantive information about original condition is not available.

OPTION 2: REHABILITATION CHECKLIST

1 Have desirable parameters or ecological conditions to be rehabilitated been explicity stated?
2 Is evidence available that the above can be rehabilitated without restoring the entire ecosystem to original condition?
3 Will the mix of parameters and conditions be ecologically stable? If not, what additional qualities must be added to achieve stability?
4 Will the attempt to rehabilitate only some of the original ecological characteristics produce any undesirable side effects?
5 Is there a significant difference in cost between rehabilitating the ecosystem and restoring it to original condition?

In some instances, rehabilitation of certain desirable characteristics can be quite successful. For example, the River Thames in England has had obnoxious odours removed; the fishery in the tidal portion was rehabilitated from virtually nothing to over 100 species, and a number of other desirable qualities were rehabilitated. No one would claim that the river is in its original condition, but, in certain respects, it is more like the original now than it was a few years ago. This is one of the best cases of rehabilitation known and further details of this effort may be found in Gameson and Wheeler.[10] This situation has been reported by Edmondson[11] for Lake Washington in the United States. There appear to be no comparable situations in regard to surface mining; therefore, aquatic systems are used to illustrate this management option.

OPTION 3: ALTERNATIVE ECOSYSTEMS CHECKLIST

1 Have the specifications of the alternative ecosystem been explicitly stated?
2 If long-term management is required, who pays for it?
3 Is monitoring for potential adverse effects outside the site boundary required?
4 Is this alternative ecosystem partly or entirely experimental? If it is, do the appropriate regulatory and civic authorities know this?
5 If several alternative ecosystems might be established, have the options been discussed with appropriate decision-makers?

This is an area where ecologists and land managers could easily show much more creativity. Of course, they will need enabling legislation to do so, but, once the opportunity is clearly perceived, this should be no major problem. An example from Virginia will illustrate that this is not an attempt to give industry an 'easy out', but rather one which sometimes makes more sense both economically and ecologically.

During the past few years, an enormous battle has ensued in the state of Virginia, which has significant surface mining in the southwestern area in terrain very much like that of West Virginia that has received much more attention in the press. The argument centres over whether areas with a greater than 20° slope should be restored to original condition following surface mining. Structural stability of such slopes is not good, particularly if a torrential rain occurs before the area has revegetated. Given the length of time likely to be required for sufficient vegetation to stabilise such slopes, major erosion and destabilising of the contours is virtually certain. Additionally, revegetation under such unstable conditions is not easy, and methods for doing so have not been developed. Thus, it would be virtually impossible to restore the area to original condition, even if funds were available. Rehabilitation of some of the most desirable characteristics of the original system does not seem to be a viable option because many characteristics would depend on restoration to an approximation of original slope; that has already been discarded as a viable option. Since there are approximately 29,150 ha (72,000 acres) of abandoned surface mined lands of this type in Virginia, the consequences of neglect are apparent, and most citizens and environmentalists would discard abandonment as a viable option under these particular circumstances. This leaves alternative ecosystems as the only option, other than abandoning mining. Since the economy of western Virginia depends upon extraction of this resource and since our energy and economic needs also call for this removal, mining is quite likely to be supported by most citizens with the requirement that it be done with the least ecological disturbance. Therefore, conditions should be established for this situation to determine the nature of the alternative ecosystem. Desirable ecological conditions are listed here, not necessarily in order of importance:

1 Effects of erosion, leaching of heavy metals and other toxic materials, and so on, upon the surrounding areas, including streams and ground water, should be minimised.
2 The system should be physically stable so that heavy rains or rapid snow melt will not destabilise the system and cause mud slides, rock falls, etc., that block roads or endanger human habitations.
3 The alternative ecosystem should achieve and maintain ecological stability as quickly as possible.
4 The system should be, at best, aesthetically pleasing and, at least, not displeasing.
5 The alternative ecosystem should contribute to recreation and agriculture and to civic and other social values.
6 Management costs should not exceed a certain percentage of the value of the extracted material and the value added by the management practices. In short, the incentives to create alternative ecosystems should exceed incentives to evade legal responsibility or abandon the systems.

Whitmore[12] has noted that 43,225 ha (106,765 acres) of new grasslands produced from reclaimed mines in West Virginia represent an ecosystem unique to the state, but not too unlike the Great Plains in avifaunal composition. Although this was not planned as an alternative ecosystem, but rather to establish certain species, one can still cite it as an example of the alternative ecosystem option. The ecosystem lost, namely steep forested slopes, was replaced by an ecosystem similar in many ways to that of the Great Plains. (That this was only partially by design is irrelevent to this discussion). The ecosystem lost is still abundant following mining, but the type gained is unique to the state. The residents of West Virginia, especially ornithologists, now have the opportunity to view species that they would otherwise have had to travel great distances to see. The land mass involved, though substantial, is still a small percentage of the total land mass. This is important because alternative ecosystems should not replace a substantial percentage of existing ecosystems, but rather a small percentage of them. One assumes that some of the natural ecosystems will be protected. Thus, alternative ecosystems may have recreational value and may be aesthetically pleasing even though alien to the native ecosystems of that locale.

Considerable scientific benefits can be gained from exploring the possibilities of developing alternative ecosystems.[13,14] Most ecological studies have been observational, i.e. recording events in natural systems, rather than experimental. Properly designed and managed alternative ecosystems could furnish a wide variety of ecologically interesting information, precisely because they are experimental rather than observational.

Another possible benefit is the diversion of pressure on natural resources as a consequence of the creation of alternative ecosystems. Peltz and Maughan[15] have shown that fish populations have been established in five stripmine ponds. The ponds described by Peltz and Maughan were not designed as alternative ecosystems since the fish were thought to be introduced accidentally and because no comprehensive management plan or substantive quality control practices were put into place. However, Peltz and Maughan felt that: 'The ponds seem to have fishery potential with proper management'.

At least one site in the United States exists where citizens have become accustomed first to picnicking, then fishing and boating, and finally to human contact with a series of ponds consisting primarily of sewage effluent. If a recreational opportunity that is in short supply can be provided by developing an alternative ecosystem to that originally present, society may make full use of this ecosystem if it is properly planned and managed. This has occurred in this country and in Australia power plant cooling lakes that serve as recreational facilities and, therefore, qualify as alternative ecosystems.

Although the use of surface-mined lands to provide alternative ecosystems for recreation has attractive potential, the biggest potential involves the use of surface mined lands for waste disposal. This is done quite often in the UK, mainly in coal and gravel. This involves considerable risks, but no course of action for disposing of societal wastes is free of risk. Transportation costs are likely to be quite high because the surface-mined land may not be easily accessible to the sites where wastes are generated. In spite of the costs, some wastes have been barged considerable distances to sea, and, when they are treated in metropolitan or urban areas, treatment costs are often high. Consequently, if wastes can be concentrated and transported at a lower cost, such use becomes more feasible. At the very least, such options deserve more attention than they have received. Illustrations of some of the possibilities available in using surface mined areas for waste disposal follow.

Fly ash slag and bottom ash disposal

Carnes [16] estimated that at least 50 million tonnes of coal ash were collected from the burning of 385 million tonnes of coal in the USA in 1972. In 1975, more than 31 million tonnes of fly ash were projected[17] as electrostatic precipitators and mechanical collectors were installed by many power utilities in response to more stringent air quality standards. The ash content of coal varies over a wide range, averaging about 11%, and consists of bottom ash, slag and fly ash.[18]

Bradshaw and Chadwick[9] have an excellent, though short, section on the reintroduction of fly ash into the environment. As the authors prudently note, this sort of scheme has not justified itself in practice. Nevertheless, this is the creative approach needed for establishing alternative ecosystems.[19]

Fly ash produced by coal-fired steam electric generating plants presents a major solid-waste disposal problem. Although the ecological effects of fly ash have not received much attention, some evidence is available.[20] There is good evidence that some aquatic organisms may inhabit fly ash settling ponds; therefore, the material, if properly handled, will present a low environmental risk. Ideally, fly ash should be reincorporated into the natural environment from which it came so that it would have the least adverse ecological impact. At present, fly ash is likely to accumulate in piles and basins at sites near power plants where it serves no purpose and may be mismanaged and cause leaching problems, etc.

Accumulating societal waste is a strategy which pushes on future generations the responsibility for today's wastes. Returning wastes to the natural environment at minimal ecological or public health impact would be far better a scheme. This might be possible if alternative ecosystems were designed for this purpose (of course, highly radioactive and other 'special problem' wastes would still have to be stored). The paper by Chu *et al.*[21] is worth examining because it provides substantive evidence about these problems.

Human and animal wastes and vegetable byproducts – disposal on surface-mined ecosystems

Perhaps one of the greatest tactical errors of human society, particularly in North America and Europe, has been the mixture of human waste materials with water to carry the mixture away from dwellings to treatment systems or directly into the environment. In the last 10 or 20 years, we have further aggravated this practice by the widespread distribution and use of garbage disposal units that shred uneaten animal and vegetable products to be mixed with water and shipped to the sewage treatment plant. This widely accepted method of waste disposal, once regarded as enlightened, may soon be seriously questioned as our water and sewage transport systems in major urban areas fall into disrepair and require replacement. In the era when clean water was thought to be in virtually unlimited supply, energy was cheap, and the delivery systems were laid under our cities and towns at what now appears to be incredibly low costs with minor maintenance requirements, few questioned this practice. Now, entire systems may need replacement in the near future with water shortages almost everywhere. With one energy pricing shock in our past and another projected within the next 10 years and municipal, state and federal revenues in decline, perhaps we will re-examine these distribution systems with cost-effectiveness in mind. Since 2% of the human waste is mixed with 98% water, the transportation, treatment and disposal problems are exacerbated. The solids alone could easily be transported to alternative ecosystems for disposal. The Clivus Multrum, produced in Sweden, makes such segregation possible with odour-free units that can be installed in ordinary dwellings. Loehr's fine book[22] covers this topic of waste disposal quite well.

OPTION 4: NEGLECT (WHICH MAY RESULT IN NATURAL RECLAMATION) CHECKLIST

1 Is substantive evidence available that natural processes will be more effective than available management practices?
2 Will the resulting ecosystem closely resemble the original, or will it be of a fundamentally different character?
3 Are any adverse ecological effects expected from the site itself, e.g. from runoff? How long should they last? Can they be mitigated?
4 How should the public and appropriate regulatory agencies be informed of the rationale for this decision?
5 Are there any adverse health or safety considerations which threaten the general public?

Neglect is perhaps an unfortunate word choice to indicate a management option because it suggests indifference. However, there are a number of instances that indicate that doing nothing will achieve desirable results as rapidly as any presently known management techniques. Numerous examples exist of inappropriate management intervention that has done more harm than the impact that management was trying to mitigate. Probably the classic example of this is the attempt to clean up oil spills with detergents, dispersants, scrubbing, etc., which under some circumstances do more harm to the biota than the oil. There is no question that in some cases of environmental disturbance, doing nothing for positive reasons is the best course of action.

CONCLUSIONS

Requirements of our technological society will result in surface-mining and other disturbances for the foreseeable future. Fertiliser, metal and energy needs alone insure this. Legislation should be enacted that will provide more flexibility in managing surface-mined lands and other disturbed ecosystems. This flexibility can only be justified if more systematic management options for coping with the ecological problems caused by mining and other activities are developed, and if these are supported, in turn, by a broader scientific base. The interplay between science, management and social choices needs more structure and more thought. Creative use of research opportunities provided by the management options just described will be of considerable academic benefit and ultimately of social[23] and economic benefit. In addition, creative use of some disturbed ecosystems will help reduce some of society's other problems, such as waste disposal, and perhaps add to recreational facilities.

Many problems of our time require that groups who are not accustomed to working together, or who are even antagonistic to each other, must now work effectively and frequently together. This makes science more holistic by causing interactions among the disciplines that have for quite a number of decades been isolated and fragmented. Such an approach simultaneously causes interactions between academe and industry. While some dangers might surface in the academic integrity of universities and other institutions as a consequence of this latter relationship, numerous benefits will be realised, particularly for ecologists for whom experimental ecosystems will have been provided. It is already evident that such interactions have been occurring, but doubtless all will agree that they could be expanded and enhanced.

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REVIEW PAPER

BIOLOGICAL MONITORING
PART VI—FUTURE NEEDS

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INTRODUCTION

Biological assessment of water pollution has depended historically on observing effects directly in natural systems. This is unsatisfactory for two reasons: (a) best management practices require prevention rather than documentation of damage; and (b) direct observation of an event in one system does not necessarily permit one to make claims about events in other systems. The documentation of damage is a necessary part of biological monitoring, and methods for determining whether biological integrity has been impaired are still being developed although a wide variety of methods for different types of ecosystems is presently available for practical use. As a consequence, the most important of the future needs in biological assessment of pollution are: (a) development of a predictive capability; and (b) means of validating the accuracy of the predictions which will in turn enable corrections to be made when the predictions are in error.

Direct observation of an environmental impact on a particular system cannot be the sole basis for the development of a predictive capability. Transfer of direct observations in other similar systems and development of a theoretical understanding of the interactions which occur under stress situations that permits predictions to be made is essential. The study of factors causing the effect is a necessary basis on which to develop predictive capabilities. Direct observation of pollutional effects has been popular with biologists because it involves minimal dependence on theoretical constructs which, in a newly developing field, may often be wrong. Its major weakness is that this sort of evidence has not proven adequate for extrapolation to ecosystems other than the one in which the event occurred except in the most general way. Since carrying out experiments, particularly with toxic chemicals on natural ecosystems, is not going to be substantively more useful than sound observations of natural systems already receiving wastes and because if such experiments were carried out on any significant scale we would soon run out of natural systems to use as controls, a prime need is microcosms or experimental systems which can be used for these purposes. This must be accompanied by the determination of the correspondence in response to pollutants between the microcosms and/or experimental systems and the natural systems.

Predictive models permit extrapolation because they provide explanations for effects in terms of comparatively readily measured processes and because such models tell us which components of the systems determine the nature of the response. Unfortunately, sound predictive models require an understanding of the dynamics of natural systems far beyond that presently available. Since neither approach (i.e. observations or predictions) is free from error, a sound biological monitoring strategy should use both lines of evidence so that the sources of errors in predictive models can be gradually eliminated and the observations on natural systems which provide the most useful information can be determined. In the initial stages of development of both predictive and observational strategies, much replication will be necessary. But as our understanding of cause–effect relationships improves, the need for such replication almost certainly will be reduced substantially.

It is a *sine qua non* that without the ability to predict environmental concentration of chemicals and their subsequent transformation products through the use of fugacity equations (e.g. Mackay, 1979) and a study of transformation kinetics prediction of a hazard will be seriously flawed. The same sort of statement applies to physical factors such as thermal loading, suspended solids transport and distribution, velocity changes, and the like. The coupling of biological evidence with chemical/physical evidence is long overdue.

SINGLE SPECIES TESTS

The single species toxicity test has been the workhorse of the hazard evaluation process practically since the field began. Compelling reasons exist for abandoning the practice of placing sole reliance on single species testing. This should not be taken as a reflection upon the single species toxicity test or a denigration of the data so produced, but rather that a scientifically sound hazard evaluation must be based on an array of information. Single species toxicity tests are essential for measuring such things as lethality, alterations of growth rate, fecundity, behavior, and the like. This essential information can only be obtained with single species tests. Therefore, such tests should continue to be an important and integral part of hazard assessment strategy. However, most single species tests use organisms of uniform age, size, and so on, and, therefore, do not display the heterogeneity of a typical population. In instances where the rate of development of population tolerance to a particular chemical is an important factor as it would be for persistent chemicals, even the single species tests might well include some population dynamics not now included. Such tests would be particularly important for situations where there is a particular target organism, such as the lamprey infesting the Great Lakes, which might make adjustments in sex ratio in response to continued application of a chemical.

Virtually all of the toxicity tests carried out are single species tests. Data generated from these single species tests have been relied upon to estimate those concentrations of chemicals in the environment that are thought to be incompatible with the maintenance of ecosystem integrity. The reasoning behind this decision appears to be that if one tests an array of species, selecting for intensive study the most sensitive species, and then determines the response of the most sensitive life history stage that this will inevitably protect all other species. One rarely finds such an explicit statement of this belief, but it seems to be the underlying assumption for regulatory agencies that place so much reliance on single species tests in the determination of chemical concentrations that will not prove harmful to ecosystems. It is worth noting that an information base as comprehensive (i.e. a substantial array of species and life history stages) as the one just described is uncommon. Most decisions on acceptable concentrations of a compound are based on evidence from a few species, and only occasionally are varied life history stages a part of the evidence base, although the strategy of using an array of species and more than one life history stage now seems favored. Still, even if the number of species tested is increased from a few to 7 or 10 and a number of life history stages of the most sensitive species are tested as well, this will represent only a small portion of the variability found in the thousands of species in natural systems. It is well documented that single species tests can provide much information on doses and times of exposures that result in changes in the survival, growth, physiology, reproduction, behavior, and other characteristics of individuals within a particular species. Looking beyond single species testing, multispecies toxicity tests are needed for such things as estimating transfer rates of a chemical through biological processes such as predation. These multispecies tests are not so greatly different from the single species tests that a major renovation of existing equipment would be necessary to carry out such assessments. Most of them could be done in already existing facilities (e.g. Coutant *et al.*, 1974). It is less well documented that testing a limited array of species for these characteristics in a graded series of concentrations of a particular chemical will provide adequate evidence to extrapolate, with some degree of precision, from the limited range of response variability to the range of variability of a large array of species. It has been widely assumed that this is the case because major environmental catastrophies have not occurred, or at least have not been detected, following discharge of wastes at concentrations determined in this way. Rarely is the validation of the accuracy of predictions as systematically and objectively carried out as one would wish. More subtle effects upon ecosystems such as displacement of species, changes in energy flow, or nutrient spiraling almost certainly would not have been detected in the absence of a comprehensive monitoring program. Rarely are such programs in place to validate predictions of hazard. Therefore, one of the key assumptions of toxicity testing, namely that single species tests can be used to protect ecosystems, is neither proven nor disproven by scientific evidence. The reasons for failure to validate this hypothesis are not at all clear, possibly it is because it is seldom explicitly stated. Test methods rarely include a preamble asserting that ecosystems can be protected by single species tests, yet this is the generally established dogma in the field. Short-term toxicity tests, particularly those limited to determining mortality, do not contribute significantly to an understanding of even single species responses to chemical exposures of considerable duration. Unfortunately, long-term tests, using more subtle parameters such as growth, reproductive success, physiological condition, and the like, are comparatively expensive, require more sophisticated facilities for longer periods of time, and also require a higher degree of professional competence both for obtaining results and interpreting them. Limitations in appropriately skilled personnel and adequate resources ensure that short-term or acute single species tests using lethality as an endpoint continue to be the major basis for estimating hazard of toxic chemicals and other environmental pollutants. Unfortunately, the uncritical use of short-term toxicity tests using lethality as an endpoint is likely to produce erroneous conclusions regarding the probable impact of a chemical on a complex ecosystem. These conclusions could err on the side of overprotecting the ecosystem and result in expenditure of funds that will produce no demonstrable biological benefits or result in underprotection and consequent damage to the ecosystem.

If one had a laboratory with all of the equipment, staff, and financial support that one wished for and could carry out as many tests on as many species as one wished, the evidence obtained from such tests would only indicate in a scientifically justifiable manner how the species tested would respond in isolation from other species. The evidence from these tests could not provide a reliable estimate of responses at any higher level of organization which includes such things as predator/prey relationships, energy exchange, inter- and intraspecies competition for resources such as nutrients or habitat, or the resistance to parasites. It should be emphasized to non-ecologists that there are many even higher levels of organization in complex ecosystems which include a complicated series of regulating processes and feedback loops. Although these may be only poorly understood by ecologists, their existence is overwhelmingly accepted.

The example which follows illustrates how laboratory results may inadequately predict events in natural systems. A laboratory toxicity test might show increased mortality of the test species which results in markedly reduced population size. However, if one introduced the chemical into a natural system inhabited by the test species, one might find the population size unchanged compared to its size before the chemical was introduced. Upon examination in the natural system, one might find that a predator which preyed upon the test species also had inhibition of reproduction or had its population size reduced in some other way. As a result of this relief from predator pressure which compensated for the mortality loss caused by the chemical, the population of the test species remained unchanged in nature. Note that the single species laboratory evidence would have predicted a marked reduction in population size. Roberts *et al.* (1978) noted that although results of lethal dose (LD 50) and lethal concentration (LC 50) tests for aquatic invertebrate species indicated low toxicity for polychlorinated biphenyls, subsequent field work and multispecies tests revealed a decrease in the diversity of invertebrate population. In contrast, Eisele (1974) carried out subchronic tests using several invertebrate aquatic species which were exposed to methoxychlor. Some of the exposed species were adversely affected at a concentration of $0.2\ \mathrm{mg\,l^{-1}}$. However, in a 1-year exposure in streams involving the original concentration of $0.2\ \mathrm{mg\,l^{-1}}$, only very subtle changes were detected in individual species, and multispecies interactions such as predator/prey relationships appeared unaffected (Eisele & Hartung, 1976).

THE CASE FOR ENVIRONMENTAL REALISM

The concepts of ecological and pollutant realism were introduced by Blanck *et al.* (1978). Test conditions that account for important characteristics of the natural environment, including both individual species and ecosystems, are considered to be ecologically realistic. Pollutant realism is attained when the characteristics of the compound in the natural environment are incorporated into the laboratory test system. These things, of course, are easier said than done, but unquestionably a dialogue is needed between those involved with natural systems and those carrying out laboratory tests so that such tests are as realistic as possible. Such groups have had little or no interaction in the past, and, as a consequence, toxicity testing procedures have not incorporated features that would improve environmental realism. At the same time, ecologists, unaware of the nature and limitations of laboratory toxicity tests, have not developed the methodology that would be helpful in validating predictions made with laboratory toxicity tests. Heath *et al.* (1969) pointed out that it was not possible to characterize the response of any system to general or specific perturbations solely from the knowledge of the response of a few component parts. Unfortunately, not much attention was given to this warning.

Biologists have given most of their attention to the impact of chemicals on the biota. But the biota may also act on the chemical by changing its nature, concentration, and partitioning (e.g. Klein & Scheunert, 1978; Maki *et al.*, 1980). Chemical and physical transformation processes will also have major effects which deserve attention if environmental realism is to be attained (e.g. Korte, 1978). Present inadequacies in predicting various transformation processes should not be used as a basis for ignoring them altogether since even a crude approximation of probable events is better than total ignorance.

MICROCOSMS AND MESOCOSMS

If one believes that single species tests, however skillfully carried out and however broad the range of species tested, cannot provide adequate information for effectively assessing chemical and other stress effects on populations or ecosystems, some qualitatively different types of information are necessary. Extrapolation from results obtained with one species to approximate the response of another species when their response to a wide variety of chemical and other stresses are known may be possible. While such extrapolation will probably not be precise, a reasonable approximation can be expected *if the data base is adequate*. Even if such efforts proved satisfactory, the cost of generating an adequate data base might be so great that one might wish to place limited resources toward other types of data generation. Extrapolation of results obtained with one form of a chemical to approximate probable results with another quite similar form may be possible, although, again, precision should not be expected. However, such basic ecosystem properties as nutrient spiraling, mineralization rates, energy flow, and successional processes must be studied by toxicity tests that are specific for these phenomena. As noted earlier, such tests carried out in natural systems are not in society's best interests because preventing such displacements before they occur is our intent. The question is whether or not some of these phenomena generally requiring the presence of a number of species can successfully be studied in laboratory systems. Fortunately, the answer appears to be a qualified *yes*. The now classical work of Metcalf and his associates (Metcalf *et al.*, 1971), the long continuing work of Taub (1974), and the variety of microcosms discussed in Cooke's summary article (Cooke, 1977) show that such studies are indeed possible although they have difficulties and drawbacks. In practice, a microcosm is not a miniature ecosystem with all its components, but rather a considerably smaller system than the natural with sufficient complexity to enable one to study certain characteristics of natural systems with environmental realism. In short, microcosms are reduced scale models of natural ecosystems or portions of natural systems housed in artificial containers kept in a controlled or semi-controlled environment.

Although microcosms have not had a major role in toxicity testing thus far (for example, they did not provide a major source of the evidence used for the 65 criterion documents prepared by the U.S. Environmental Protection Agency in response to the consent decree), the advantages are considerable. Because microcosms are small, replication and a degree of standardization are both possible. Lack of replication and uniformity are major weaknesses of many system level studies. Control of both chemical and species composition is substantially greater than in natural systems. Comparisons of different concentrations of single chemicals or an array of different chemicals are facilitated under these conditions. This absence of environmental variability may facilitate inference of causal relationships in some instances and impair it in others. From a safety standpoint, potentially dangerous substances can be tested in contained systems without contaminating the natural environment. Of course, disposal will still be a problem, but a more manageable one. The absence of complicated spatial heterogeneity permits a more complete definition of physical, chemical, and biological characteristics as it does in some natural systems (e.g. certain Antarctic lakes). Properly used, this simplification can be advantageous, but used improperly could produce serious error. By using higher concentrations than one would expect in nature, the type of impact may be determined in less time. The most important advantage, however, is that effects beyond those at the single species level can be studied.

There are, of course, difficulties whenever problems of scale are involved. Simplication and miniaturization may produce error (Whittaker, 1961; May, 1974; Jassby *et al.*, 1977a, b; Dudzik *et al.*, 1979; Harte *et al.*, 1979, 1980). The chemical-physical environment in microcosms may be quite different in many aspects from that of natural systems. Important biological phenomena, such as succession, are difficult to replicate in microcosms. Increasing the complexity of microcosms may reduce these problems, but increases cost and makes standardization more difficult. This in turn reduces the opportunity for replication. The rather shallow depths of most aquatic microcosms may produce unrealistically large nutrient fluxes and decomposition rates in benthic compartments, as well as distorting vertical migration patterns of zooplankton. Inclusion of larger organisms, such as fish, can distort biomass relationships and nutrient cycles. Various distortions of surface–volume ratios can have a variety of chemical, physical, and biological effects.

Sometimes these deviations from environmental realism can be offset. For example, Perez *et al.* (1977) have designed benthic chambers that provide a reduced surface area of the sediment in contact with the water. Harte *et al.* (1979) coped with surface-volume problems by using single-operating procedures. However, in most instances, a technological solution will be not feasible, and an awareness of the problem will be the chief protection against error.

Since the experimental ecosystem is selected or designed to display a particular quality that might be altered by the stress being studied, the results obtained will depend upon the skill of the experimenter in selecting significant parameters most likely to be affected by the stress and the degree to which these characteristics in the experimental ecosystems resemble those in natural systems. The source of the material to assemble experimental ecosystems deserves careful attention. Ideally, the material should be from an ecosystem for which a substantial amount of information is available. The source ecosystem should be as free as possible from contaminating materials, and evidence should be provided that this is the case. The most likely source for materials fitting these specifications would be ecological preserves. They should be selected at a national level to represent a spectrum of ecosystems throughout the country that would have ongoing surveillance of biological/chemical/physical conditions with the information stored in a readily accessible form. Naturally, samples for assemblage of microcosms or allocation of a portion of the system as an experimental ecosystem would require careful management to avoid destroying the very properties that make the preserve valuable.

Despite the difficulties and problems just discussed, microcosms offer an opportunity to obtain information that cannot be obtained from single species tests and which should not be obtained from natural systems which we wish to protect.

FIELD TESTS IN NATURAL ECOSYSTEMS

Circumstances undoubtedly exist where a combination of single species and microcosm toxicity tests do not adequately define the hazards of using a particular chemical. If the benefits from using a chemical are particularly attractive and no alternative formulations appear to work as well, field testing may be necessary to determine fully the environmental risk involved. Field enclosures that contain the test material should restrict the damage to the area allocated for the test. Even so, such tests should be used only when satisfactory evidence cannot be obtained in laboratory systems. Field enclosures of a representative portion of a natural system have already been

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shown to be useful (e.g. Odum & Jordan, 1970). Although field studies provide a degree of environmental realism difficult to obtain with microcosms, replication is a major problem, as is skilled operation in a test system.

Tests carried out in unenclosed natural systems are unquestionably the most realistic since few extrapolations are needed. The subtle effects and interactions not present or easily detected in simplified systems can often be detected in field experiments (Nelson *et al.*, 1975), although they may be masked by high variability. Some serious disadvantages exist. Since our purpose is to protect natural systems, one does not wish to damage even a portion of one in the process since rehabilitation may take years and be quite costly. Also, the number of natural systems with a sufficient data base both in breadth and duration to ensure such tests will serve this purpose well are pitifully few. Until more systems are included in this category, the few that now exist represent a resource that must be cherished. Even when this desirable state has been achieved, such systems must never be squandered. In short, proposed tests must meet the most rigorous professional standards before authorization for testing is given.

A number of authors have discussed the disadvantage of field tests (Cooke, 1971; Nelson *et al.*, 1975; Draggan, 1976; Lighthart & Bond, 1976; Heath, 1979). Difficulty of replication due to environmental variability seems to be the most significant problem.

ESTABLISHMENT OF ECOLOGICAL RESOURCE AREAS

In addition to the need for field sites with an adequate data base for testing purposes, compelling reasons exist for establishing a number of "ecological resource areas" (ERA). For some years, I have espoused the establishment of such areas to furnish "seed" organisms for recolonizing damaged areas (Cairns, 1976). These ERA's would be selected to provide an array of land and water ecosystems representative of those most common in each country. Preference should be given to areas under surveillance for some time with substantive data bases available. A variety of biological, meteorological, and chemical-physical data would be routinely collected and stored in a readily retrievable form. A few such sites already exist (e.g. the Savannah River Environmental Research Park near Aiken, SC). These would be academically useful in studying long-range ecosystem changes, determining natural variability in structure and function, and the like. However, with careful planning they could serve some other purposes as well.

Having "pedigreed" sources of biological material for microcosms and other experimental ecosystems would be helpful. That is, material from an area where the living conditions and performance of the organisms is reasonably well understood and documented would be useful. The ability to replicate microcosm studies will depend heavily on the background information available on ERA's.

Some types of pollution affect vast areas (e.g. acid rain). These must be studied in isolation from other pollutional effects. In such studies, field enclosures might be used to simulate "normal" or pre-acid rain conditions.

Certain areas of these ERA's might be set aside to validate predictions made on the basis of single species toxicity tests, microcosm tests, and the like. In instances where the predictions were in error and damage resulted, the site could then be used to study recovery from perturbation.

MATHEMATICAL MODELS

Although mathematical models are not commonly used in pollution assessment, they do provide a link between observations and predictions. For example, if one wishes to use observations of steam electric power plant effects on one river system to predict effects on another, a model might be developed based on the characteristics common to both systems with the expectation that the results might be somewhat similar. To a degree, success will depend on the number of inferences required because these will increase the probability of error. Acquiring skill in the development and use of mathematical models would enhance the extrapolation of data from one site to a number of other places. The advantages are so numerous that it is clear that development of mathematical models well beyond our present capability deserves serious attention. However, mathematicians attempting to do this would do well to read the article by Slobodkin (1975).

MONITORING WASTE DISCHARGES AND OTHER INTRODUCTIONS OF CHEMICALS INTO THE ENVIRONMENT

A prudent industrial plant manager will see that all point source discharges under his control have laboratory toxicity tests run on them at the earliest possible moment. Presumably for new plants, this would be done during the pilot scale operation when simulated wastes should be produced. In many regions of the United States, such toxicity testing is now required as part of the permitting process. In addition, a prudent industrial plant manager would also have some evidence on the ecological condition of the system into which the wastes are discharged, preferably before the discharges begin. Under these circumstances, determination of whether the estimates of risk were correct by biological monitoring of the receiving system both before and after the waste discharges enter is possible. The use of natural systems receiving waste discharges for which permits have been obtained probably offers the single most inexpensive means of checking the accuracy of the predictions and determining what additional evidence is needed if the predications are in error. If several similar waste discharges are being introduced into ecologically different receiving systems, a situation not uncommon when the same product is produced in different parts of the country or world, the factors which cause the similarity or differences in response can be determined. Mathematical models can then be developed that will enable utilization of information generated in one system for other not identical ecological systems. If each major discharger would carry out laboratory toxicity testing in conjunction with biological monitoring of the receiving system, this would substantially reduce the need for field testing in natural systems, development of complex microcosms, and the like.

Another situation in which such studies would be extremely valuable is the improvement of waste treatment facilities to provide additional protection to the receiving system. In the United States, such studies have been carried out on the Wabash River near Lafayette, IN (Spacie & Hamelink, 1979), on the lower southfork of the Shenandoah River near Front Royal, VA (Seagle *et al.*, 1980), and on the South River near Waynesboro, VA (Cairns, 1981). In each of these instances, improvements made in the waste treatment system were accompanied by biological benefits. Of all the means of determining the correlation between single species and system level responses, this simple, straightforward, readily available and comparatively inexpensive source of data appears to have been largely overlooked.

REHABILITATION OF DAMAGED ECOSYSTEMS

Despite some very notable success stories (e.g. Holdgate & Woodman, 1978) of rehabilitation of damaged ecosystems, there is no generally accepted plan for: (a) systematically identifying ecosystems where rehabilitation is desirable; (b) establishing priorities for attention; (c) determining the degree of rehabilitation that would provide amenities attractive to the public; (d) estimating the degree of ecological improvement that would result from implementing improvements in waste discharges and nonpoint source discharges into the system; and finally (e) providing a professionally endorsed set of parameters that would enable us to track the improvements as they occur and a monitoring system that would ensure quality control measures to protect the rehabilitated state. A schematic depicting several policy options for management of natural ecosystems is given in Fig. 1. In this diagram, *restoration* takes the ecosystem back in a rather direct route toward its initial state. Presumably, undesirable features of the initial state would be accepted as part of the overall package. *Enhancement*, or improving the current state of an ecosystem without reference to its initial state, might lead an ecosystem farther from its initial state, perhaps by adding desirable man-made features and suppressing natural features. *Rehabilitation* may be defined as a pragmatic mix of nondegradation, enhancement, and restoration. To the extent that natural ecosystem recovery can be fostered, restoration of some desirable features can be expected to be a cost/effective tactic within such a mix. Further degradation, of course, is always an option but one which society seems to have rejected in recent years.

The remarkable rehabilitation of the Thames (e.g. Gameson & Wheeler, 1977) and Clyde Rivers in England provides compelling evidence that present methodologies and waste treatment technologies, however imperfect conceptually, can produce remarkable results in a relatively few years. Furthermore, this rehabilitation was not financially ruinous to the industries using the water and can, at least in the case of the Thames, simultaneously turn an ecological eyesore into a multi-use recreational facility. Perhaps the best confirmation of the success of the overall program on the Thames is the problem of meeting a variety of demands which may in part conflict with each other (e.g. sailing and fishing).

In an era when energy costs are skyrocketing, recreation close to one's residence will likely become increasingly common. As a consequence, rehabilitation of aquatic ecosystems near large concentrations of population seems prudent.

From an academic standpoint, some urgent needs are:

1. The degree of ecological improvement resulting from a specific reduction in point and nonpoint pollutional loading must be determined with greater precision.
2. The estimation of recovery time once particular pollution stresses are removed must be substantially improved.
3. Means of communicating various ecological options in terms of rehabilitation must be developed.
4. Quality control monitoring during both the rehabilitation and/or recovery period, as well as for the maintenance of desirable quality once achieved, must be improved markedly.

Although rehabilitation of damaged ecosystems has not had a high priority in the U.S. Environmental

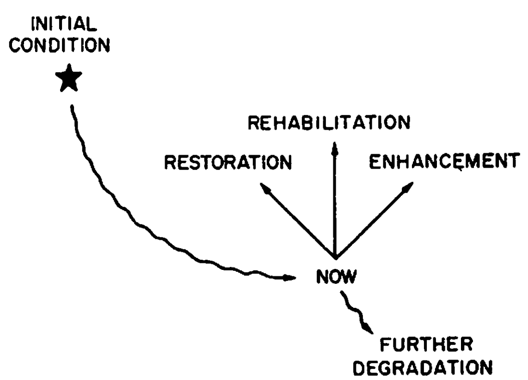

Fig. 1. Diagram to illustrate the meanings of several policy options for management of natural ecosystems.

Protection Agency in past years, it is one of the items selected for serious consideration in the research and priority recommendation prepared for the USEPA by a group of consultants coordinated by Pennsylvania State University (1979).

DEVELOPMENT OF PREDICTIVE CAPABILITY

The Toxic Substances Control Act (TSCA; U.S. Public Law 94–469) and some other federal legislation, such as the National Environmental Protection Act (Public Law 91-190), require the development of a predictive capability. TSCA requires determination of the hazard to human health *and the environment* in the extraction, transportation, manufacture, use, and disposal of all new chemicals and all old chemicals used for a new purpose. Environmental impact statements are required for the construction of new power plants, pipe lines, and a variety of other activities. Unfortunately, "impact statements" have generally been a list of species and other data without any substantive attempt to make a probabilistic determination of hazard based on this evidence (e.g. Kates, 1978). TSCA has not yet been substantively implemented due in part to the lack of predictive capability in this area. This lack is striking in the 65 criterion documents on toxic chemicals prepared by the USEPA as a consequence of the consent decree resulting from a suit brought against the administrator by the Friends of the Environment, the Natural Resources Defense Council, and other environmental groups (*Federal Register*, 1978a,b, 1979a,b). The criterion documents (some of these are mentioned in the report of the Water Quality Criteria Subcommittee, 1980) suffered from the following deficiencies:

1. The information was not gathered in the systematic, orderly way required for a scientifically justifiable estimate of risk.

2. A large variety of methods, species, and time limits were used which made analysis of conflicting results quite difficult.

3. In some criterion documents, no information was included about environmental partitioning, transformation processes, and other phenomena important to sound hazard evaluation. Rarely was the information about the environmental fate of the chemical linked in any substantive way to the selection of organisms at risk, the length of exposure, the probability of toxicity from secondary transformation products, and the like. In short, not only was the biological information fragmented but the essential chemical/physical information also was not properly coupled to the biological information.

4. Statistical analyses were absent or less than desirable.

5. Almost all of the information used to estimate the no adverse biological effects concentration was based on single species tests. This approach has several weaknesses: (a) The species used were usually those easily culture in the laboratory and often did not inhabit the areas in which the chemical substance was likely to intrude. (b) There were virtually no system level tests (i.e. energy transfer, nutrient cycling, predator-prey interactions, and the like). (c) The tests were usually at a constant concentration with all environmental variables held constant. In the "real world", both the environmental variables and the potential contaminant are likely to vary rather widely, although probably not in concert. (d) There was no description of the types of environmental measurements necessary to check the accuracy of the predictions made—in short, no form of error control that would enable one to adjust the predictions based on experience.

DRAINAGE BASIN MANAGEMENT

It is unfortunate that there are so few drainage basin management groups for a variety of reasons: (a) long-term records of water quality and changes in the organisms which inhabit the system are rarely available. As a consequence, it is difficult to know how much change is normal, and often one cannot distinguish natural cyclic changes from degradation. (b) It has been very well established by the Hubbard Brook Studies (Bormann and Likens and their associates) as well as Coweeta Studies (Patten and Webster and their colleagues) that the terrestrial system surrounding a drainage basin is a regulator of water quality as well as the primary source of energy in many headwater areas. Unfortunately, large scale events (building developments, destruction of woodland, etc.) which influence water quality are rarely studied in a systematic way and, even when they are, it is unusual to find sufficient careful documentation to enable one to develop correlations with changes in water quality. (c) The absence of an organization interested in the entire system almost ensures that system level questions regarding a particular course of action will not be asked. As a consequence, water management problems are dealt with in a fragmented way and rarely is more than superficial attention given to the whole. (d) Remedial quality control measures which might be taken immediately if a monitoring system run by a management group were in place are frequently postponed until a major perturbation occurs. (e) There is no overall strategy for restoring damaged aquatic ecosystems and, even on the few occasions when the outline of a plan is developed, it is usually not implemented in any systematic way because of the lack of regional authority.

The Ohio River Valley Water Sanitation Commission (ORSANCO) is an interstate agency representing Illinois, Indiana, Kentucky, New York, Ohio, Pennsylvania, Virginia, and West Virginia which assesses water quality in the Ohio River and some of its tributaries. Water quality analyses of the dissolved oxygen concentration, temperature, pH, conductivity, and flow are reported monthly, and a comparison with the desired values is included in the report. Biological data are rarely included, and the data base lacks a frequent assessment of levels of toxicants or other materials in the river that might impair biological integrity. Nevertheless, even these limited data enable one to make some modest predictions about the condition of the aquatic biota. Unfortunately, few river basin commissions in the United States do as much as ORSANCO. Even in such important, but relatively straightforward matters as water allocation, the Potomac River situation (see reports of the Water Supply Review Committee, National Research Council, Washington, DC) provides clear evidence of the difficulties when regional management is not in place. In contrast, the management of the Thames by the Thames Water Authority has superb water allocation and maintenance of water quality accompanied by a very marked improvement in the condition of the aquatic biota as well as recreation amenities such as sailing, swimming, and so on. For example, since the early 1950s when there were few permanent fish residing in the tidal Thames, the situation has been improved so that there are now over 90 species to be found. The restoration of the salmon run is a very real possibility despite the fact that this is one of the most heavily used rivers in the world running through one of the largest concentrations of humans on the globe. The river Clyde has had significant, though less well publicized, recovery.

This clearly illustrates that present technologies and methods are adequate for not only management of river water quality (including preservation of aquatic biota) but for rehabilitation of the river in a cost/effective manner without severe dislocations to municipalities and industries.

DETERMINATION OF ASSIMILATIVE CAPACITY

The assumption that ecosystems have assimilative capacity is based on the lack of evidence for deleterious effects at all concentrations of a chemical below a certain threshold. EPA's maximum allowable toxic concentration is also based on the assumption that a demonstrable threshold exists. Prominent ecologists (e.g. Woodwell, 1975) believe there are no thresholds in ecosystems and that the presumed thresholds are an artifact of testing procedures and limitations in the number of parameters assessed. In contrast, another prominent ecologist (May, 1977) provides theoretical support for the existence of thresholds and breakpoints in ecosystems. It is worth emphasizing, as Odum *et al.* (1979) have done, that not all change is deleterious and that some inputs into a system producing a measurable response may be regarded as subsidies, particularly if one uses anthropocentric criteria. For example, an increase in phosphorus loading of the system producing an increase in desirable biomass (sport or commercially valuable fish), and, therefore, a desirable subsidy to fishermen, might still be regarded as a form of degradation by an ecologist since certain species of microorganisms sensitive to high nutrient loading might disappear or be severely reduced in population size. Additionally, some persons make a distinction between a naturally occurring chemical and those produced by an industrial society. However, some commonly used industrially produced products may also be found in nature. For example, the mushroom *Gyromitra* (Helvella) *esculenta* releases monomethylhydrazine as a decomposition product (Lincoff & Mitchel, 1977). Since monomethylhydrazine has been commonly produced as a rocket fuel, one might assume that it is not likely to have ever occurred in natural systems before the industrial revolution. However, there is evidence that it can be released from mushrooms under circumstances likely to have occurred, at least occasionally. Thus, what appears to be a space age chemical has been around, albeit in smaller quantities, for quite some time.

While the continuum concept (i.e. no thresholds) may have theoretical validity, it seems to have little operational value. Many years ago, the poet Francis Thompson wrote "...Thou canst not stir a flower Without troubling of a star." Newton's Law states "Every body attracts every other body with a force that..." However, although the validity of this hypothesis is recognized, it is frequently ignored because it is of practically no importance. The theoretical effects are quantitatively beyond our ability to measure, and, therefore, of no operational utility. The problem is very similar to that faced by taxonomists who name species. Species tend to follow all sorts of gradients through time and space, and one could make the case that species are a consequence of the taxonomists desire to classify. At any rate, however, the practice of placing organisms into categories called species, which are frequently not very discrete (thresholds are not sharp) in the real world, has proven to be of considerable operational value and utility. The practice has enhanced communication among scientists in different geographic regions and speaking different languages and has also permitted communication through time by way of museum records, published documents, etc. The people who have pointed out that there are no thresholds in ecology have done all of us a great service by reminding us that the "thresholds" we see may be artifacts of our inability to measure responses, just as we cannot measure the displacement of a star when someone lifts a flower. Operationally, we have a much better chance of measuring the biological effects than we do the movement of the planets, and, therefore, we should be prepared to adjust as new and more discriminating measurements become available.

Where the question of assimilative capacity is concerned, however, differences from one site to another which we can measure fairly effectively should be a matter of considerable interest and concern. Present legislation in the United States, such as Section 316A of Public Law 92-500 (*Federal Register*, 1974) permits biological information to be used to obtain a variance

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from existing standards. Exceptions are made if evidence can be provided to show that no harm to the indigenous biota is occurring or will occur under the proposed conditions of heated wastewater discharge. This merely recognizes that where heated wastewater discharges are concerned some ecosystems may not be displaced in either structure and/or function by discharges that would do so in another location. There seems to be no rational justification for not extending this concept to include chemical substances for which the same statement is also true. This would permit dischargers who are prepared to gather a substantive body of biological and ecological evidence to use the nondegrading assimilative capacity of the system more fully than they otherwise would. Additional waste treatment requires energy both in the operation of the system and in the construction of the facility, and the production of this energy itself will probably cause an ecological displacement somewhere (e.g. strip mining). It seems prudent to compare the ecological damage done by producing additional energy for waste treatment and the damage done if no additional waste treatment were put in place. By looking at the entire picture of ecological displacement, one might often come to quite different conclusions than those presently formed. The use of the assimilative capacity concept fits in with the risk/benefit debate now going on between business and regulatory agencies in the United States (Large, 1980). Representative Ritter (Pennsylvania) wants to require regulators to make objective comparisons between the risks of various actions. Before banning fluorocarbons which deplete ozone in the upper atmosphere, the regulators would have to compare the dangers of a general increase in ultraviolet radiation with a person's deliberate exposure to ultraviolet radiation through sunbathing. In a case of assimilative capacity, the benefits of additional treatment at a particular site would have to be weighed against the ecological damage caused elsewhere by the steps required for the additional waste treatment.

TIERED VS SIMULTANEOUS TESTING STRATEGIES

Most of the hazard evaluation or toxicity testing protocols proposed in the recent past have been sequential or tiered (e.g. Cairns & Dickson, 1978; Cairns *et al.*, 1978; Dickson *et al.*, 1979). The reasoning behind the tiered or sequential arrangement follows:

1. It is necessary to do the inexpensive, single species, relatively crude range finding tests using lethality as an endpoint in order to select the concentrations that would furnish the most information in the more expensive, longer range, and more subtle tests involving sublethal responses.

2. The amount of information necessary for estimate of hazard would depend upon the relationship between the no adverse biological effects concentration of the chemical and the environmental concentration of the chemical. If early tests showed that these were far apart and the environmental concentration were well below the no adverse effects biological concentration, one might reasonably not require as much testing as one would if the two were close together. In the latter case, it would be more difficult to determine their relationship or even which one is definitely below the other due to the large uncertainty about the location of both concentrations in the early stages of tiered testing. In other words, for some chemicals one would proceed only part way through the tiered system, and in others might go half way or all the way through. If one carried out all these tests simultaneously for all compounds, there would be an information "overkill". As a consequence, proceeding sequentially would be more cost effective.

3. If tests are carried out sequentially, the information generated in earlier tests could be used in the design of subsequent tests.

However, limited experience with these protocols has indicated that simultaneous testing might be more cost effective for the following reasons: (a) The longer term tests invariably are placed at or near the end of the tiered sequential testing series and, therefore, are started long after the first tests. If time is a major factor in determining costs (e.g. Schramm, 1979), less might be spent if all tests were started simultaneously. (b) It is becoming increasingly evident that the single species range finding tests and other tests carried out at the beginning of a series are not likely to furnish useful information about ecosystem responses or even about the specific range in which the sublethal responses are likely to occur for a single species. If there were an inevitable relationship between the concentrations obtained in the earlier part of the tiered system and those in the latter part, one could easily apply a "K" factor to the early ones and obtain the latter ones without doing the work. Since this consistent relationship does not appear to exist, the amount of information obtained from the earlier tests appears not to be worth the delay in starting the more subtle tests. (c) The scientists and technicians who do the more sophisticated tests will probably not be the same ones who did the range finding tests. Those who do the range finding tests will almost certainly be less skilled and less well educated than those who are charged with the responsibility of carrying out the longer term tests. As a consequence, it is highly unlikely that significant amounts of information will be transferred from those who carry out the range finding tests to those who carry out the more comprehensive and complicated tests. (d) The historical sequence in which tests were developed (i.e., from the simpler to the more complicated) cannot be assumed to be the best sequence for a testing strategy. An array of related but qualitatively dissimilar data is likely to enable one to make more accurate predictions of environmental effects than a limited array of data.

Sequential testing might seriously delay the acquisition of the critical data mass needed to make a sound environmental decision.

CONCLUDING REMARKS

A strong belief exists among some of the people who carry out single species toxicity tests that no system level tests are necessary to protect ecosystems. This goes beyond the argument against ecosystem tests because: (a) they are thought to be too expensive, and (b) there is not a strong professional concensus supporting particular system level toxicity tests as there is for single species toxicity tests. Those who believe that single species tests are sufficient say that when such tests have been done and properly utilized there has been no evidence of ecosystem damage. There is no substantive evidence either supporting or refuting this statement. In fact, only extremely rarely has the value of single species toxicity tests as a means of protecting ecosystem properties been validated in a scientifically justifiable way. The fact that no evidence has appeared of ecosystem damage under the circumstances described may be merely because there was no program designed to test this hypothesis and that the ecosystem effects were usually too subtle to be identified by casual observers.

It could also be that ecosystems are more resistant than single species tests indicate (e.g. Eisele & Hartung, 1976). If this is true, money may be spent on waste treatment that results in no biological benefits.

Another objection to system level tests has been "who cares whether an ecosystem rate or process such as colonization rate is impaired?" This is surely not a valid criticism since a hazard evaluation process requires an estimate of the probability of harm to human health and the environment. In short, identifying the types of harm likely to occur under certain conditions is an essential component of the hazard evaluation process. The determination of whether it is an acceptable risk in terms of the benefits to be derived is a social-political decision. Those carrying out the hazard evaluation should not prejudge the social decisions likely to be made once the evidence is complete. Finally when a decision is made to do something about environmental problems, the number and kinds of personnel needed to do the work properly should receive more attention than it has in the past (e.g. Gloyna *et al.*, 1977).

This paper goes so far beyond the present practices and legal requirements that it will undoubtedly be labeled as visionary. The charge will be made that all of these tests cannot be done for all chemicals, despite the fact that nowhere in this series of articles has this been recommended. We are passing through a transitional period not dissimilar to the "agricultural revolution". During that period, society found that harvesting and gathering food from the unmanaged environment did not deliver this resource in either sufficient quantity or quality to meet society's expectations. As a consequence, environmental management was undertaken to ensure that the delivery more closely approximated society's needs. Although this has not been fully accomplished, the unmanaged environment could not possibly feed even a fraction of those humans now alive. Similarly, the unmanaged environment is no longer capable of assimilating society's waste without careful management. Elsewhere (Cairns, 1979) I have noted that biological assessment and monitoring of pollutional effects will probably go through 4 developmental phases: (1) awareness, (2) observational, (3) predictive, and (4) managerial. Naturally, these phases will not be clearly demarcated, but it is reasonably clear that we are now entering the predictive phase by attempting to estimate the hazard to human health and the environment of the use of various chemicals before the use is authorized. This is also true of other types of industrial society development. Present practices have proven inadequate for both the prediction of effects and the validation of these predictions. As a consequence, one must ask what additional evidence is needed to improve our predictive capability, including the validation of predictions where correct and the determination of error when not. The management phase cannot be effectively initiated until appropriate management tools are available, including the capabilities just discussed. At the risk of redundancy, I reiterate that this article only attempts to outline the types of information for which there are future needs if estimates of hazard are to be scientifically justifiable. It would be ridiculous to use every test we are capable of carrying out on each and every chemical, and I have stated this repeatedly (most recently Cairns, 1980). The appropriate mix of evidence will have to be determined on a case-by-case basis using protocols established for this purpose. In no case do I believe it possible to do anything with zero risk.

It is quite likely that these suggestions may be considered scientifically sound but economically impractical. The determination of hazard or risk is a probabilistic determination requiring scientific evidence of the kinds discussed in this and other articles in the series. When the cost of proceeding without an adequate estimate of risk exceeds the cost of getting the information by a substantial margin, as it clearly has in many cases in recent years, one might ask whether the acquisition of information leading to a more reliable estimate of risk is impractical.

These statements may be regarded as platitudinous by some readers but, since these issues keep reappearing with monotonous regularity, perhaps they are platitudinous to only a small minority of the human race.

Clearly, we are not using presently available practical biological monitoring methods on the scale that is needed for adequate environmental quality control. Implementation of use can be accompanied by development of new methods. The highest priority should be given to multispecies and system level tests.

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John Cairns, Jr.
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Estimating Hazard

John Cairns, Jr.

In 1976, the Natural Resources Defense Council and other environmental groups won a class action suit against the administrator of the U.S. Environmental Protection Agency (EPA) for noncompliance with Section 304 of Public Law 92-500. This section charged the administrator with developing and publishing water quality criteria that accurately reflected the latest scientific knowledge on the kinds and effects of compounds that may be deleterious to biological communities and their component species. As a consequence of this suit, a court order directed EPA to publish criteria in 1979 on 65 chemicals and classes of chemicals identified as hazardous to aquatic life. The second drafts of these water quality criteria for a number of hazardous compounds were published in the *Federal Register* on 15 March and 25 July 1979.

Although the words *criteria* and *standards* are often used interchangeably, there is, in fact, an important difference. Because *criteria* are quantities and qualities based on scientific determinations, they must be scientifically justifiable. *Standards* derived from criteria may be influenced by local or national political, economic, and social factors; they include plans for implementation and questions of water use and management. In short, although the numbers recommended in a criterion document may be converted directly by the states without further thought and reflection, the intention was that this should not be the case. It should be a *sine qua non* that, without adequate information, criteria cannot be produced.

Several simplistic assumptions have clouded the processes of criterion development and standard setting, including the following:

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- Some chemicals are inherently safe and some are inherently dangerous.

- The assumption that waste-treatment technology is capable of removing all alien (not there originally) materials from industrial process water, and that the discharge pipe could then be hooked up to the water-intake pipe to produce a totally self-contained system, has hindered criterion development. Even if such removal were technologically possible—which it is not in most cases—the energy requirements alone, not to mention the economic cost, would make it prohibitive.

- There are no acceptable discharge rates or concentrations for most industrial chemicals. This conviction has not been altered by laboratory and field evidence of no-adverse-biological effects because the advocates of this position maintain either that the tests were not carried out for a sufficient length of time or that the wrong parameters were measured. Industries that might be willing to carry out a prescribed number of tests are unwilling to begin testing if they cannot see the terms under which clearance will be granted.

Nothing we do has zero risk; intelligent choices are best made when one can estimate the hazard of a particular course of action with reasonable reliability. This does not mean that harm will not be done; rather, the amount of harm likely to occur from a particular course of action will be defined and decision-makers will be able to balance the benefits and the risks reliably.

The assumption underlying the cost/benefit approach is that for most chemicals there is a site-specific, time-dependent, nondegrading loading capacity, which I have elsewhere termed *assimilative capacity* (Cairns 1977). Not all change is deleterious. Odum et al. (1979) have articulated the difference between *stress*, *perturbation*, and *subsidy* very concisely. In ecological usage, *perturbation* is any deviation, or displacement, from the "nominal stage" in structure or function at any level of organization. Then *stress* describes unfavorable deflections from the nominal state, and *subsidy* describes favorable deflections from the nominal state. In short, chemical/physical alterations can be made without having deleterious effects.

The recognition of these—and the following—distinctions is essential to a clear understanding of hazard estimation. *Risk* is the probability of harm from an actual or predicted concentration of a chemical in the environment. *Safe concentrations* are those for which the risk is acceptable to society. As a consequence, the assessment of hazard requires both a scientific judgment based on evidence and a value judgment of society and/or its representatives. Evidence for a scientific judgment must cover (a) *toxicity*—the inherent property of the chemical that will produce harmful effects to an organism (or community) after exposure of a particular duration at a specific concentration, and (b) *environmental concentration*—those actual or predicted concentrations resulting from all point and nonpoint sources as modified by the biological, chemical, and physical processes acting on the chemical or its byproducts in the environment.

THE TOXIC SUBSTANCES CONTROL ACT

The Toxic Substances Control Act (TSCA) provides that no person may manufacture a new chemical substance or process a chemical substance for a new use without obtaining clearance from EPA. One of the main purposes of TSCA is to establish a procedure for estimating the hazard to human health and the environment before widespread use of a new chemical occurs. After examining the data produced to implement the evaluation, the EPA administrator must judge the degree of risk associated with the extraction, manufacture, distribution in commerce, processing, use, and disposal of the chemical substance. If the evidence indicates that the chemical substance under described conditions of use presents an unreasonable risk of injury to human health or the environment, the administrator may restrict or ban manufacture and use of the substance.

The purpose of this paper is to discuss the activities necessary to estimate biological hazards of chemical pollutants. These activities include analyses of empirical facts (data) followed by appropriate scientific judgments (conclusions). This important, but badly neglected exercise, and the lack of attention given it by the academic community have resulted in confusion, disagreement, and inaction on some very pressing problems.

MAGNITUDE OF THE PROBLEM

In an article entitled "Chemicals: How Many Are There?" Thomas H. Maugh II (1978) gave a one-page summary of the overall problem. The American Chemical Society's computer registry contains 4,039,907 distinct entities, and the number has been increasing at a rate of about 6,000 per week. Of these, the ACS has given EPA a preliminary list of approximately 33,000 chemicals that are thought to be in common use. EPA believes there may be as many as 50,000 chemicals in daily use not including pesticides, pharmaceuticals, or food additives. And the Food and Drug Administration estimates approximately 4000 active ingredients in drugs and 2000 more used as excipients (inert ingredients), as well as 2500 additives for nutritive purposes and 3000 more to promote product life. Taking all of these together, Maugh estimated about 63,000 chemicals in common use.

The number of people competent to carry out toxicity tests and environmental fate-and-effects determinations, however, is exceedingly small (Committee on Human Resources 1977). Although people can be quickly trained (i.e., a year or two) for the crude short-term tests using lethality as an endpoint, it is extremely time-consuming to educate people to conduct the long-term tests or interpret the data. Moreover, facilities suitable for carrying out such tests are not abundant, and funds for conducting hazard evaluations are limited even when facilities and personnel are available. One cannot carry out every test developed by the academic community on every chemical. Recognition of this simple fact makes mandatory the development of a process for determining testing priority and a means for determining when sufficient evidence is available to proceed with the manufacture and use of a chemical at a risk level acceptable to society.

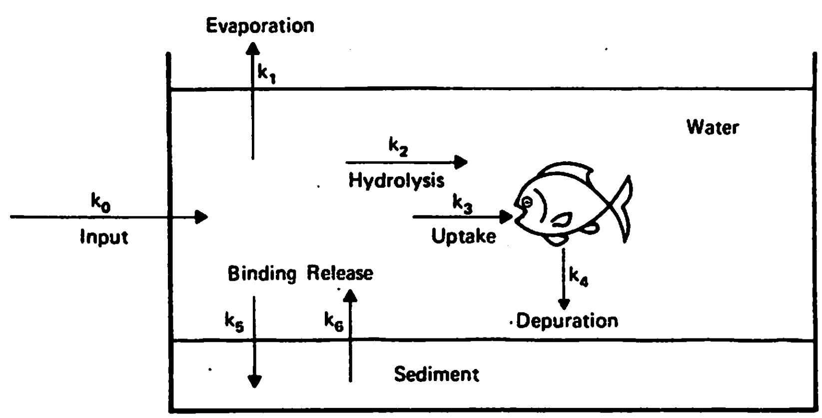
Biologists must be concerned not only with the effect of a chemical upon the environment, but also with the effect of the environment on the chemical. Various types of transformations, including biotransformation, are common for most chemicals. Frequently the transformation results in less toxic products. As a consequence of transformation, dilution, and other factors, the diminution of toxicological properties is often rather substantial. Even for persistent chemicals, we must know the environmental pathways: how the material is partitioned between air, water, and soil; what types of environmental sinks are operative; and under what conditions releases might be expected from these sinks.

Determining Environmental Pathways

The determination of the fate of a potentially hazardous substance introduced into the environment is a necessary precursor of any meaningful attempt to establish effective standards for regulatory purposes. For example, a chemical associated primarily with lake sediments requires a different assessment strategy from that for a chemical associated with the water column: Toxicity tests for the former should involve benthic organisms, whereas planktonic organisms would be most appropriate for the latter. A rapidly degrading substance would require fewer chronic tests than a persistent one.

Stern and Walker (1978) have developed an approach to identify the principal medium into which a chemical may be distributed after release into the environment. The following series of tests was used: water solubility, partition coefficient (octanol/water), adsorption by natural solids, desorption or leaching, and volatility. For example, if a chemical is soluble in water, does not transfer to octanol, does not readily adsorb to soils, readily leaches from areas in which it is deposited, and has a low degree of volatility, testing of persistence and ecological effects could be limited to these conditions and biological targets associated with the liquid phase of bodies of water and, to a lesser degree, their sediments. The integration of data developed in tests of environmental mobility with the information required by TSCA Section 8 will provide a useful indication of possible "target" organisms.

The new environmental rates approach (Branson 1978) requires that properties be measured as *time-concentration rates*. These are properties of the compound that have predictive value and extrapolating results to the "real world." This exercise involves predicting (as opposed to measuring) the concentration of



MATERIAL BALANCE EQUATION

$$V\frac{dCw}{dt} = k_0 - k_1ACw - k_2VCw - k_3FCw + k_4FC_f + k_5SCw + k_6SC_S$$

Input | Evaporation | Degradation (Hydrolysis) | Fish Uptake | Fish Depuration | Sediment Binding | Sediment Release

Fig. 1. Pond model. (From Branson 1978, p. 58. Reprint, with permission, from ASTM STP 657, *Estimating the Hazard of Chemical Substances to Aquatic Life.* Copyright ASTM, 1916 Race St., Philadelphia, PA 19103.) Equation key: V = volume of water (milliliters), A = surface area (cm^2), F = fish mass (g), S = sediment mass (g), Cw = concentration of chemical in water, k = rate constant, C_f = concentrations of chemical in fish, and C_s = concentrations of chemical in sediment.

a chemical in a natural environment through time. These rate constants are based on known biological mechanisms (including kinetics of enzyme reactions).

The time-concentration rates are then incorporated into a suitable model for predicting environmental concentrations. Fig. 1 depicts a pond model, the key properties, and the materials-balance equation for predicting the fate of chlorpyrifos in the pond. The predicted and experimentally found concentrations in the fish and water (Table 1) were in close agreement.

The environmental concentration of a chemical is governed by the properties of the chemical, the rate of its introduction into the environment, and the characteristics of that specific environment. Many of the properties—e.g., molecular structure, water solubility, vapor pressure, absorption spectra (ultraviolet and visible), and particle size (if the substance is particulate)—may be readily available from data banks. Others that may be readily attainable before biological program design begins could include some rate constants—i.e., for photodegradation (ultraviolet and visible), biological degradation, chemical degradation, evaporation, sediment binding, uptake by organisms, depuration by organisms—and some partition coefficients—i.e., octanol/water, air/water, and sediment/water (Johnson et al. 1978, p. 73). The characteristics or properties of the environment—e.g., surface area, depth, pH, flow/turbulence, carbon in sediment, temperature, salinity, suspended sediment concentration, trophic status, and absorption spectra (ultraviolet and visible)—are equally fundamental.[1]

The determination or estimation of persistence is important in designing biological testing. Fig. 2 depicts a simple schematic for persistence testing. The screening sequence was designed for testing only photochemical and chemical degradation; biodegradation should be included in the sequence where it is important.

The figure illustrates three levels of screening: Level 1 was designed to differentiate between persistent and degradable chemicals. Those that prove to be persistent would then be subjected to

[1]Methods for deriving these properties are being prepared by the task groups of ASTM Subcommittee E35.21 on Safety to Man and Environment. (Reprint, with permission, from ASTM STP 657, *Estimating the Hazard of Chemical Substances to Aquatic Life*. Copyright ASTM, 1916 Race St., Philadelphia, PA 19103.)

TABLE 1. Predicting the fate of chlorpyrifos in a pond.* (From Branson 1978, p. 59. Reprint, with permission, from ASTM STP 657, *Estimating the Hazard of Chemical Substances to Aquatic Life*. Copyright ASTM, 1916 Race St., Philadelphia, PA 19103.)

	% of total	
Compartment	**2 days**	**25 days**
Water	48.0	0.8
Soil	25.0	0.5
Fish	0.8	<0.1
Air	3.8	11.4
Metabolized	2.9	11.0
Hydrolyzed	25.0	76.0

*Seven-day (similar agreement at days 2, 4, and 28) concentrations of chlorpyrifos: (a) water (5.75 μg/liter at $t = 0$)—predicted 1.0 μg/liter (Neely and Blau 1977), found 1.0 μg/liter (Macek et al. 1972); (b) fish—predicted 0.8 μg/gram (Neely and Blau 1977), found 1.0 μg/gram (Macek et al. 1972).

detailed appropriate tests for ecological effects. Degradable chemicals would be shunted to Level 2 testing, which consists of an evaluation of substrate disappearance usually involving chemical half-life determinations. If results show that degradation is insignificant, the compound will receive the same ecological testing as the persistent chemicals identified at Level 1. Level 3 testing (identification and quantification of degradation products and their rates/conditions of formation) is so expensive that it should be carried out only if toxicity testing reveals that these products have significant adverse effects on human health or other organisms.

Environmental chemistry-fate covers the transport and transformation of a chemical for all modes of input from the point of entry into the environment to its final disposition. The physical processes of advection-transport and dilution-dispersion must be defined for the terrestrial, atmospheric, and aquatic components of the environment. Environmental chemistry-fate also covers the chemical processes that influence the form(s) or chemical species of the contaminant as well as its transformation products in each of the major areas of the environment (Lee and Jones 1980). A meaningful hazard-assessment program must consider not only the parent compound but also the potential environmental significance of transformation products. If there appears to be no concrete evidence that the toxicological properties persist or are substantively increased, then the precise nature of the transformation products is of little significance in the hazard-evaluation process.

Since environmental fate and concentration of the new chemicals cannot be directly measured before the chemical is manufactured and used, they must be estimated. Baughman and Lassiter (1978) and Branson (1978) have developed mathematical models to describe the expected concentrations in various components of the environment based on the properties of the chemical, modes and rates of input to the environment, and reactions that can occur in the environmental compartments with which it is associated. Verification of these models can be accomplished in laboratory microcosms such as those developed by Metcalf (1974), which evaluated compounds producing biological magnification, or in the field mesocosms men-

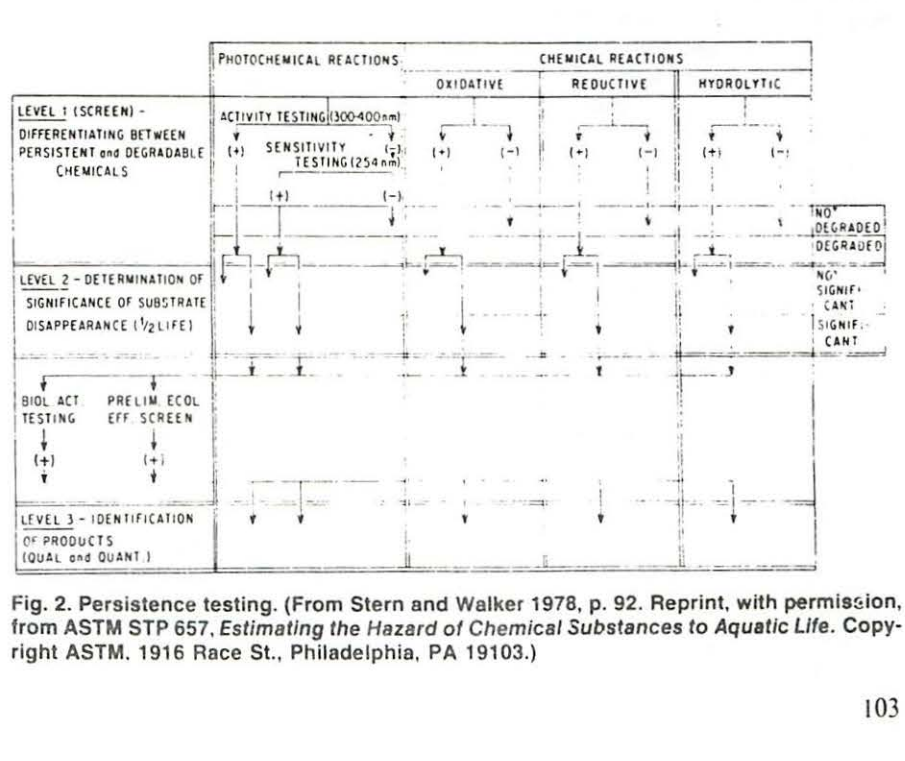


Fig. 2. Persistence testing. (From Stern and Walker 1978, p. 92. Reprint, with permission, from ASTM STP 657, *Estimating the Hazard of Chemical Substances to Aquatic Life*. Copyright ASTM, 1916 Race St., Philadelphia, PA 19103.)

tioned by Odum et al. (1979), for linking the observational-theoretical modeling approach with the straightforward experimental approach.

Relationship between Toxicity and Environmental Concentration

A sequential toxicity series starts with a determination of short-term lethality and proceeds through increasingly sophisticated tests (often of greater duration) to define the more subtle effects. Eventually, a reasonable estimate of risk can be made, based on an estimation of the environmental concentration of the chemical—including whatever by-products or transformation products have toxicological properties—and the no-adverse-biological-effects concentration (Fig. 3). The sequential testing procedures provide increasingly accurate estimates of the concentration of the chemical substance that does not cause adverse biological effects and the environmental concentration that results from intended use (see Fig. 4).

At the beginning of a test series, one would have to estimate the properties of the chemical on the basis of its affinity to other chemicals. Since closely related chemical species may have substantially different toxicological properties, the confidence interval must be very broad. Even after testing begins, if one has tested only three or four species representing three or four trophic levels and only one life-history stage of most of the species tested, one would still have relatively low confidence that the boundary conditions for biological response have been properly defined. As one increased the variety of tests, one would have increasing confidence (but not certainty) that the no-adverse-biological-response threshold had been identified. Furthermore, since environmental conditions strongly mediate the toxic response, one would also need to see how much variability in toxicity was produced by differences in water hardness, pH, dissolved oxygen concentration, and temperature.

We are not yet able to culture, or even maintain, most species in the laboratory. So we must extrapolate from a small number of test species to a large number of species likely to be exposed in the "real world," and there are errors inherent in this process. Moreover, few toxicity-test procedures are available for microcosms, and these are comparatively expensive and infrequently used. Therefore, single-species toxicity tests often

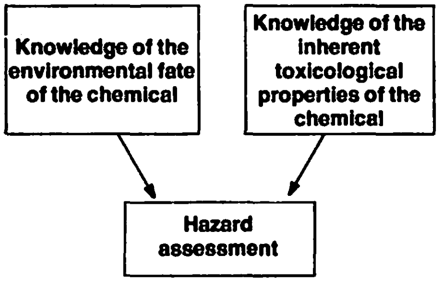


Fig. 3. Components of a hazard assessment.

must be used to make estimates of response thresholds of communities.

The same general problems of inability to examine all chemical species in all situations plague those attempting to determine environmental concentration. Substantial analytical costs for environmental fate-and-effects studies further restrict the size of the data base.

Two recurring views have been voiced on this subject. The first is that our present state of knowledge and relatively primitive methodology are insufficient to make any decision whatsoever, and, therefore, it is arrogant to do so until more precise and refined methods have been developed. But even present methodology properly used would have prevented or markedly reduced the tragedies involving kepone, PCB, PBB, and mercury contamination. Second, some academicians would rather see an exhaustive study made of a few chemicals than a process that requires at least some information on all. One might identify these using a decision matrix, such as the one in *Principles for Evaluating Chemicals in the Environment* (NAS 1975) shown in Table 2.

The decision on when enough information has been gathered can be based on the proximity of the expected environmental concentration to the highest test concentration producing no adverse biological effects. If the former is well below the latter, a decision can be made at that

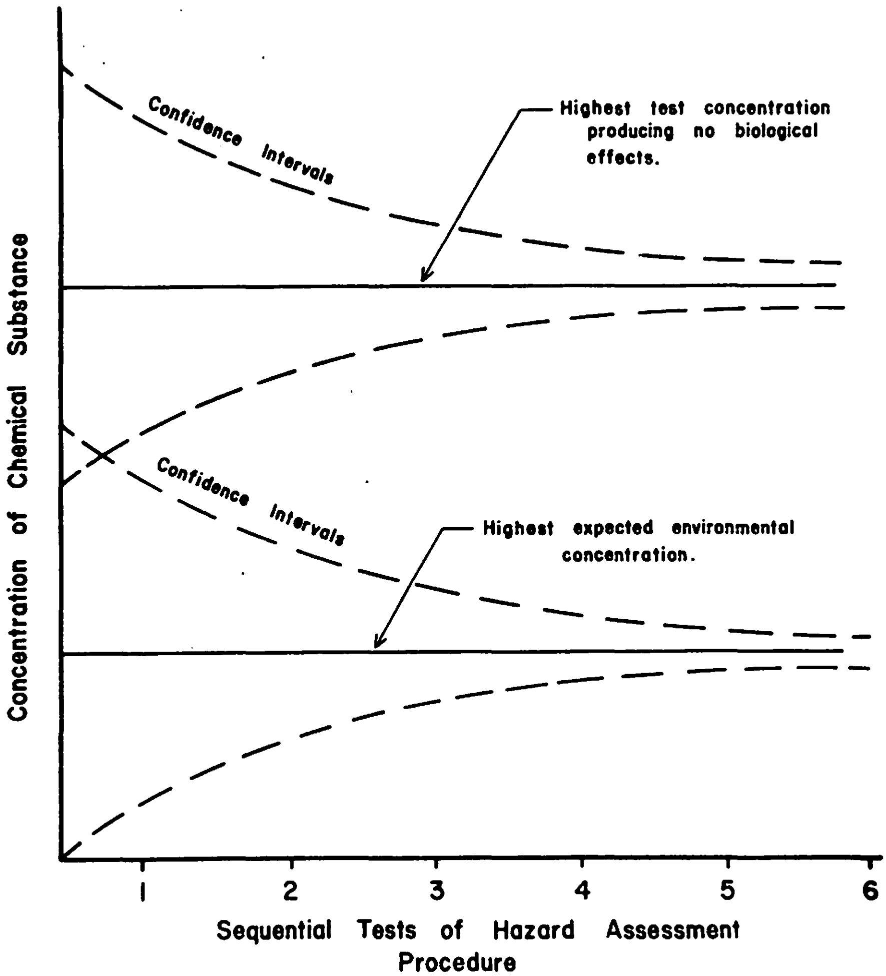


Fig. 4. Diagrammatic representation of a sequential hazard-assessment procedure demonstrating increasingly narrow confidence limits for estimates of no-biological-effect concentration and actual-expected-environmental concentration. (From Cairns et al. 1978b, p. 195. Reprint, with permission, from ASTM STP 657, *Estimating the Hazard of Chemical Substances to Aquatic Life*. Copyright ASTM, 1916 Race St., Philadelphia, PA 19103.)

point in the testing sequence when the zones of uncertainty (dotted lines, Fig. 4) no longer overlap. As the environmental concentration approaches the no-adverse-biological-effects threshold, a greater number of tests will be necessary to ensure that the environmental concentration is indeed lower than the no-adverse-biological-effects concentration. Of course, should it be higher, the use of the chemical would be either restricted or banned.

An example of the tiered or phased system of sequential testing to reduce uncertainty about the response threshold is given in Fig. 5 from Stern and Walker (1978). Other sequential testing protocols to estimate the no-adverse-biological-effects concentration for aquatic organisms may be found in Dickson et al. (1979), which contains the protocols developed in various countries including the United States, Germany, England, Japan, and France. Other methods, including the one developed for the American Institute of Biological Sciences, may be found in the related documents section of Cairns et al. (1978a).

TABLE 2. Scheme for classification of chemicals according to biological impact and dispersal.

Chemical dispersal	Biological impact* High (1)	Medium (2)	Low (3)
Widespread			
High release (1)	1	2	3
Low release (2)	2	4	6
Localized			
High release (3)	3	6	9
Low release (4)	4	8	12

*Low number indicates high testing priority.

PREDICTIVE AND REACTIVE ERROR CONTROL

Viewed as an environmental quality-control problem, the process of hazard evaluation might be considered a predictive form of quality control designed to prevent errors. Despite the advantages of estimating the hazard of a chemical before it is manufactured and used, there are two severe disadvantages: (a) an extrapolation must be made from a few species to many and from a limited array of environmental conditions to an enormous array, and (2) the system lacks a feedback loop from the environment to indicate the validity of the predictions made.

Establishment of a feedback loop is a form of error control to alert the system managers to the impending or actual onset of conditions deleterious to the biota. I discussed one early warning system in a recent *BioScience* article (Cairns and Gruber 1979). Other types of biological monitoring methods are used in the receiving system itself (i.e., any natural system receiving societal wastes in any form) (Hellawell 1978). *Biological monitoring* is a term often used in the United States merely to indicate the gathering of data, frequently not even in a systematic way. I prefer the more rigorous definitions of the terms *survey*, *surveillance*, and *monitoring* by Hellawell (1978):

- *Survey:* an exercise in which a set of standardised observations (or replicate samples) is taken from a station (or stations) within a short period of time to furnish qualitative or quantitative descriptive data.
- *Surveillance:* a continued programme of surveys systematically undertaken to provide a series of observations in time.
- *Monitoring:* surveillance undertaken to ensure that previously formulated standards are being met.

One monitors a patient in an intensive care ward or the instrumentation in the cockpit of an aircraft with the intention of taking corrective action when desirable conditions are not being met. This restricted definition of the word *monitoring* should be mandatory in the United States: otherwise a regulatory agency can say that it is monitoring a situation—implying that corrective action will be taken when conditions become hazardous—but without actually doing so. Furthermore, corrective quality-control action should not have to be taken

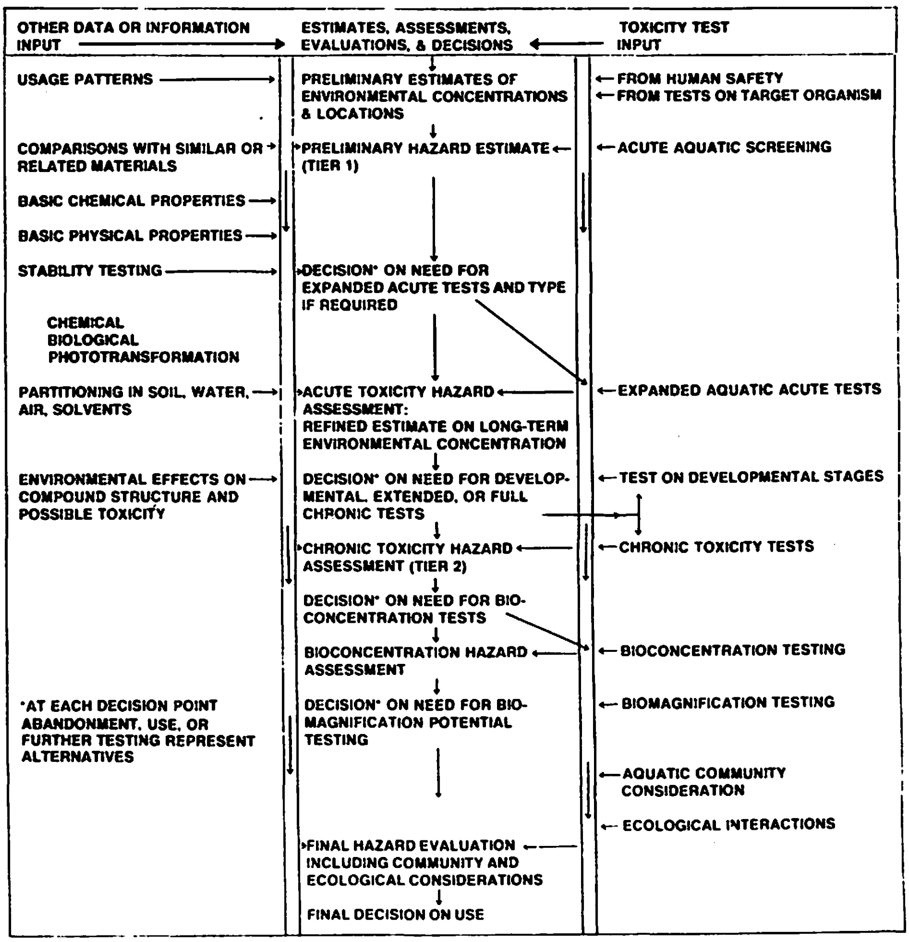


Fig. 5. Overall diagram showing data inputs and sequential assessments and decisions. (From Cairns et al. 1978a. Reprint, with permission, from ASTM STP 657, *Estimating the Hazard of Chemical Substances to Aquatic Life*. Copyright ASTM, 1916 Race St., Philadelphia, PA 19103.)

through the courts because effective quality control requires an immediate response to remove the stress and return conditions to normal (Odum et al. 1979). Predictive and error control should be used in concert and not individually, although this is not the practice today. A simplified diagram of the appropriate relationship of these for a point source discharger is given in Fig. 6.

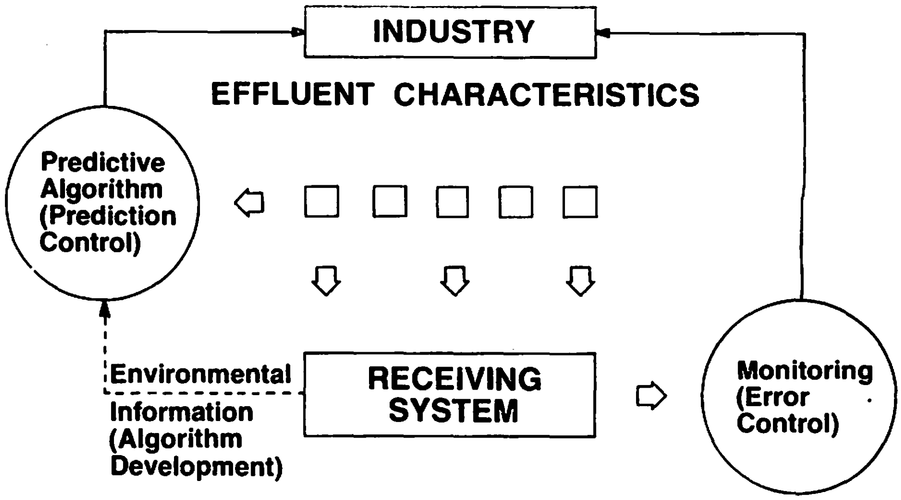


Fig. 6. Information flow in environmental control processes. (From Herricks and Cairns 1979)

THE REGULATORY DILEMMA

This article began with a discussion of the class action suit against the EPA administrator for noncompliance with Section 304 of Public Law 92-500. Subsequently, 65 chemicals and classes of chemicals were identified as hazardous to aquatic life. EPA has made a major effort to cope with a legal requirement to resolve a complex problem with an inadequate data base and insufficient time to generate new data. For some substances (e.g., ethylbenzene and naphthalene), there were insufficient data to justify derivation of criteria, and very little of the available information was generated systematically.

Thus, even when tests for a particular chemical are carried out with an array of organisms, or when several tests with a single species are carried out by different investigators, the methodologies are often different, and comparisons are exceedingly difficult. Biologists have only just begun to produce standard methods similar to those used in carrying out tests to protect humans. Only recently has the certification of those carrying out the tests received any attention, although both standard methods and certification are common to many other professions.

The organisms likely to be exposed to a compound are often not those for which toxicity-test information (for that compound) is available. When there is insufficient time or funds to generate a large body of new information, one alternative is to apply various "correction factors" to the data available in an attempt to extend the usable data base. This option has been given considerable attention by EPA (*Federal Register*, 18 May and 5 July 1978 and 15 March and 25 July 1979), although the tactic may soon be abandoned.

Correction factors may be used to convert one type of data to another. For example, there is a correction factor to estimate the "measured" concentration when the toxicant concentration was not determined. There are correction factors for converting 24-, 48-, and 72-hour LC50s to 96-hour LC50s and for estimating flow-through LC50 values from static and renewal data; these are sensitivity factors, which are supposed to produce a value that will protect all aquatic life. Substantial errors are possible (Buikema and Cairns 1979) if one uses a limited series of correction factors in sequence. The only alternative is to generate an adequate data base and evaluate hazard in a scientifically justifiable way.

WHERE DO WE GO FROM HERE?

We are now in a major transitional stage comparable to the agricultural revolution, which resulted from the fact that the unmanaged environment would not produce either the quantity or quality of food that society desired. As a consequence, the human race moved from the hunting and harvesting stage to the agricultural production stage.

The unmanaged environment cannot receive society's unregulated wastes indefinitely without serious harm. We must, therefore, exert control qualitatively and quantitatively in order to minimize or eliminate harm. That is, we must develop better means of reducing the environmental impact of a highly technological urban society without causing a major social displacement.

So, we must manage the present system far better than we have before. For biologists this means (a) determining, as a profession, the best biological parameters to estimate the probability of harm to the biota and ecosystems, (b) standardizing the best methods for measuring these parameters, and (c) determining who is qualified to make the measurements and indicate this publicly by some form of certification. I hope the AIBS and its member societies will take a strong leadership role in these areas and encourage the development of curricula that do more than make students aware of existing problems. I believe biological assessment and monitoring of pollutional effects will go through four developmental phases, each painful and aggravating, but eventually something in which scientists and society as a whole will have confidence (Cairns 1979):

1. *Awareness Phase:* This begins for many people with a single well-known ecosystem (or species) and may expand to a global perception. For most people in the USA, this expansion began with Earth Day.
2. *Observational Phase:* Documentation of the damage in a qualitative, and eventually a quantitative, way. (We are well into this phase.)
3. *Predictive Phase:* Development of the capability to estimate reliably the consequences of a particular course of action with reasonable precision. (We are reluctantly being forced into this phase.)
4. *Managerial Phase:* Development of a capacity to orchestrate environmental and health effects to optimize costs and benefits. (It will be a long time before we enter this phase.)

There are, however, grounds for cautious optimism. We now have sufficient assessment, monitoring, and predictive capabilities that, if properly used, will substantially reduce risks to human health and the environment. Unfortunately, our skill in, and zeal for, generating data far exceeds our ability to interpret them, particularly in terms of system integrity and response. The preoccupation with minutiae, which is the foundation for the generation of sound data, is the most formidable obstacle to its enlightened use. If we are to evolve from phase 2 observation to phase 3 prediction, we must learn to combine precision with vision.

Finally, if we are to become effective managers of the environment (phase 4), the various disciplines must learn to work together much more effectively than they now do. Today, we consider ourselves enlightened if we merely listen to those in other disciplines. We must go beyond this. The only question is whether we will do it skillfully and gracefully by anticipating needs and enlarging our perspective beyond that of a single discipline, or whether we will do so grudgingly in response to major catastrophes (Cairns 1979).

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REVIEW PAPER

BIOLOGICAL MONITORING PART I—EARLY WARNING SYSTEMS

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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

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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 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 parameters, 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 the luciferin-luciferase materials extracted from fireflys.) Total coliform and fecal coliform levels are estimated by measuring hydrogen gas evolved by these organisms in the presence of lactose at certain incubation temperatures. The whole system is automated by two computers and housed in a mobile trailer. Although expensive and complicated, this system does indicate the technological potential for development of biological monitoring systems.

Respiration (oxygen consumption) is probably the most widely tested bacterial monitoring technique. An experimental chamber is supplied continuously with a bacterial nutrient material as well as the water to be tested. Oxygen electrodes measure the difference in dissolved oxygen between the influent and effluent liquids. Normally, bacterial respiration results in a certain decrease in oxygen concentration in the effluent depending in part on the retention time of the chamber and temperature. Toxic materials, by inhibiting bacterial respiration, will reduce the difference between the two oxygen levels. A monitoring system of this type described in Morgan (1976) was developed by Axt (1972, 1973a,b). Responses were obtained to several heavy metals at concentrations of 0.1 mg l^{-1}, but organic pesticides tested (e.g. endosulphan) did not cause a response at levels less than 10 mg l^{-1}.

Oxygen consumption in a biological filtration system has been used to monitor the toxicity of the effluents from several industrial sources in Sweden since 1974 (Solyom *et al.*, 1976; Solyom, 1977). Synthetic sewage is added to the filter along with the industrial waste or toxicant being tested; the oxygen level in the water leaving the filter then is monitored by an oxygen electrode. Toxicity is indicated by an increase in oxygen concentration, which is shown on a recording device. This apparatus known as "Toxiguard", sounds an alarm when the oxygen level reaches a pre-set value. Tests with toxicants indicate that this system responds to copper, cyanide, and low, as well as high, pH (Table 1). Toxic changes in the effluents from chemical and pharmaceutical companies have been detected in a number of cases.

Reeves (1976) evaluates an influent toxicity monitoring system which used a commercially available respirometer to record oxygen uptake continuously in a small activated sludge unit. Tests were done with both simulated raw wastewater from several sources and with actual influent at a municipal plant. The respirometer showed rapid and significant changes in oxygen consumption to a wide range of toxicants, but only at relatively high toxicant levels. Reeves describes a similar device called the Biomonitor (Brubaker & Moss, 1976) in which toxicity is determined by the degree of oxygen uptake by two samples of aeration basin liquor exposed to raw wastewater.

An influent monitor ("BioMonitor") to protect municipal waste treatment systems against shock loadings from industrial waste sources was developed by Clarke *et al.* (1977). Early warning of shock loadings was provided by measuring changes in dissolved oxygen concentrations in an activated sludge unit receiving a continuous feed of the materials being tested. Tests with ten different industrial wastes indicated a rapid response to shock loads which was proportional to the magnitude of the load.

Nitrification is another functional parameter which has been used to indicate a toxic environment. This method uses "a continuously operating percolating filter in which nitrifying organisms were selectively cultured" (Holland & Green, 1975). Ammonia was introduced continuously into the filtration column along with the water being tested. A specific ion electrode measured the reduction in ammonia concentration after its passage through the column. The presence of a toxicant inhibiting nitrification made the inlet and outlet ammonia levels nearly equal. Tests with a number of toxicants showed that nearly all produced a measurable response within three hours after their introduction. Toxicity threshold concentrations (the level of toxicant resulting in a 50% decrease in nitrification efficiency) were determined. With three exceptions (hydrochloric acid, paraquat, and diquat), recovery of the column's function was quite rapid after toxicant addition was stopped.

Field tests of the nitrification column were conducted to determine its usefulness in monitoring pollution levels in several rivers (Stroud & Jones, 1975). The rivers tested had varying pollution loads, but in no case was the toxicity threshold of the column reached. An addition of cyanide to the water of one river did cause a response by the system at a level of the same order as in earlier laboratory tests (see Table 1). The operation of the column was quite reliable. With periodic maintenance, it was capable of running for several months without deteriorating.

A number of devices has been developed for continuous measurement of activity and respiration (oxygen consumption) of macroinvertebrates. Arnold & Keith (1976) developed a continuous flow respirometer suitable for recording changes in respiration of larger macroinvertebrates. Oxygen concentrations are measured with a dissolved oxygen meter; most of these have an analogue or digital voltage output that could readily be adapted to an automatic recording system. A continuous respirometry device used by Livingston (1968, 1970) could be automated only with some difficulty due to the degree of manual operation required by the manometric technique used to determine oxygen levels. Another disadvantage is that water flow must be stopped while oxygen consumption measurements are made. Maki *et al.* (1973) designed a continuous-flow respirometer which requires that water be recirculated through the unit while oxygen measurements are being made. Sublethal levels of the organophosphate insecticide Dibrom caused significant changes in oxygen consumption in both the stonefly *Hydroperla crosbyi* and the hellgrammite *Corydalus cornutus*.

Several types of instruments have been used to record activity levels of smaller aquatic organisms. One, developed by Kapoor (1971), continuously monitored the respiratory body movements of aquatic insects. The animal was placed on a small beam connected to strain gauges in such a manner that any movement of the animal produced a proportional electrical signal which then could be recorded. A different approach was taken by Heusner & Enright (1966). Activity was measured indirectly by sensing heat loss from a thermistor caused by water currents produced by the animal's movements. An electronic circuit then emitted continuous electrical pulses, the frequency of which was related to the rate of water movement past the thermistor. It is necessary to keep the water temperature fairly constant in order to ensure proper operation of the device. Either of these two techniques could be used in a computer-based system.

Another kind of activity monitor using crayfish has been automated by Maciorowski *et al.* (1977). Movements of the crayfish in individual chambers created small electrical signals (apparently due to muscle contraction) which were received by wire electrodes affixed to the ends of the chamber. This same method was used previously by Camougis (1960) and in seawater by Idoniboye-Obu (1977). After amplification, signals from a total of eight crayfish were monitored by a microcomputer; the number of peaks in the electrical signal per unit time, corresponding to the total amount of movement, was counted and totaled every 15 min. At that time, the counts for each crayfish were recorded in digital form on paper tape for later analysis. Several days of data taken prior to toxicant exposure were used by a previously developed statistical program to set 95% confidence limits on the movement counts for each crayfish and hour of the day (Hall, 1972; Hall *et al.*, 1975). Data taken during toxicant exposure were compared to the appropriate confidence limits to determine if abnormal movement was present. This approach is similar to that for two other systems using ventilatory and movement patterns (Cairns *et al.*, 1973a,b). In tests with cadmium, the crayfish showed a definite response within 1 h to concentrations of 2.5 and 0.5 mg l^{-1}. Responses to 0.10 mg l^{-1} were registered after 40–45 h (Table 1).

Simonet *et al.* (1978) used the inhibition of negative phototaxis in first instar larvae of the mosquito *Aedes aegypti* as a measure of toxic effects. The inability of the larvae to move 30 cm in 1 min after exposure to strong light was tested following 8 h of exposure to toxicants. The 8-h LC50 values were lower than comparable 24-h LC50 values based on mortality.

Many techniques became available which may be used to monitor toxicity with fish. Some of the parameters which may be monitored include the cough reflex, oxygen consumption, movement patterns, avoidance, and rheotaxis (Morgan, 1977). To this list changes in blood components, heart rate, and swimming endurance might also be added. These possibilities are discussed below along with some already developed toxicant detection systems.

Both heart rate and blood composition may be altered by toxicants. Changes in heart rate in response to environmental hypoxia have been described (Randall & Shelton, 1963; Marvin & Heath, 1968; Marvin & Burton, 1973; and others). Swimming activity also affects heart rate (Stevens & Randall, 1967). Lunn *et al.* (1976) found that DDT altered the heart rate of rainbow trout at sublethal levels, but that dieldrin did not have an effect until lethal concentrations were reached. Carbamyl did not affect heart rate at the levels tested. Acutely toxic levels of zinc caused decreased heart rates while sublethal levels of copper caused bradycardia in the carp (*Cypruinus carpio*, Labat *et al.*, 1976) in bluegills within 2 h (Cairns *et al.*, 1970a,b). Elevated levels of blood glucose have been found in several species of fish exposed to sublethal and lethal levels of various toxicants, but a significant response may take several hours to several days to develop (McLeay *et al.*, 1972; Silbergeld, 1974; McLeay, 1977). Elevated levels of cortisol were found in coho salmon (*Oncorhynchus kisutch*) within several hours after their exposure to acutely toxic levels of copper (Donaldson & Dye, 1975; Schreck & Lorz, 1978), but no response to cadmium was found (Schreck & Lorz, 1978). Sellers *et al.* (1975) found that the blood pH and arterial pO_2 of rainbow trout were decreased by a 48-h exposure to a concentration of zinc near the 96-h LC50.

Whatever the sensitivity of blood components to toxicants, there are technical problems associated with sampling blood that would complicate development of a monitoring system. Cannulae would have to be implanted and the fish restrained from movement; this would cause a good deal of stress on the fish. Furthermore, the blood analysis equipment required would likely be complex and difficult to use on a continuous basis. Although the measurement of heart rate generally has required the use of implanted electrodes which can also be troublesome (Cairns *et al.*, 1972). Venables & Smith (1972) developed a method which allows the fish to be unrestrained, while Ibaragi (1970) used a small FM transmitter attached to the fish to provide remote sensing of heart rate. In some cases, heart rates may be monitored with remote electrodes in a manner similar to opercular movements (Rommel, 1973; Drummond & Carlson, 1977).

Measurement of total fish activity can be done with less stress on the fish. One method used by Spoor (1946) related the amount of activity to the degree of deflection of an aluminum paddle. These deflections, caused by water currents produced by the fish, activated a relay that generated impulses which then could be recorded. Another method was to provide a chamber in which the fish was free to swim, the amount of activity was recorded as the number of breaks per unit time in a light beam from a photocell(s) which crosses the tank. With some variations, this technique has been used by several investigators (Shirer *et al.*, 1968; Hudson & Bussell, 1972; and Hafeez & Barber, 1976). Cripe *et al.* (1975) recommended using an infrared light-emitting diode as the light source to avoid possible disturbance of the animal being monitored. One difficulty with using light beams is that the beam may be blocked in turbid water or in water having large particulate matter. Alignment of the light source and light detector also can be a problem.

An automatic monitoring system based on changes in total activity of bluegills has been under development by Cairns and others at Virginia Polytechnic Institute and State University. An early version (Waller & Cairns, 1972) recorded the breaks by a single fish in three red light photocell beams (650 nm) at three different levels in the test tank. Data were stored in counters and automatically photographed and reset each hour. Normal and abnormal movements were determined by changes in the variance of light beam interruptions. In a 96-h exposure, the lowest detectable levels of zinc were between 2.94 and 3.64 mg l^{-1}. In a 14 day test, Ellgaard *et al.* (1978) found that locomotor activity in bluegills was affected by zinc at concentrations of 0.1 and 5.0 mg l^{-1}. Activity was also altered by sublethal concentrations of DDT (Ellgaard *et al.*, 1977), cadmium, and chromium.

Further automation of this system was achieved when data from electronic counters of photocell beam interruptions were fed every half-hour directly into a minicomputer which stored the information for later statistical analysis (Westlake *et al.*, unpublished; Cairns *et al.*, 1975). A statistical program (used also by Maciorowski, discussed previously) used several days of pretoxicant exposure data to set upper and lower 95% confidence limits for each fish and half-hour of the day. Subsequent corresponding values recorded outside of these limits were said to indicate abnormal movement. This system was able to detect sublethal levels of complex effluents from a U.S. Army munitions plant, including TNT and nitroglycerin wastes (Westlake *et al.*, 1974a,b). No significant response was found to changes in pH between pH 5 and pH 10 (Westlake *et al.*, 1974c).

Up to six fish activity monitors developed at the Stevenage (England) Laboratory of the Water Research Centre are now being used on rivers in England. Alarms are triggered when the activity of the fish exceeds a pre-set level. Work is continuing to produce a monitor which will take factors such as feeding and the time of day into account in determining a proper alarm level (Miller, personal communication).

Another parameter related to activity which has been used as an indicator of toxicity is the loss of rheotaxis, which is the ability of a fish to maintain its position in a current. Rheotaxis is to some extent a function of swimming ability, and laboratory studies have shown the swimming performance of fish to be affected by a number of toxic materials, including zinc and ammonia (Herbert & Shurben, 1963), hydrogen sulfide (Oseid & Smith, 1972), pulpwood fiber (MacLeod & Smith, 1966), pulp mill effluent (Howard & Walden, 1974), detergents (Cairns & Scheier, 1963), and the insecticide fenitrothion (Peterson, 1974). Poels (1975, 1977) designed an automatic system which employs photocells to determine when loss of rheotaxis occurs in a chamber through which test water is flowing. When any one of the three fish in the chamber falls back to the downstream end of the chamber, photocell beams are broken and a mild electric shock is applied to force the fish back into the upstream area. If two of the three fish spend more than 5 min of a 15 min period in the downstream end or pass into it more often than normal in 15 min, an alarm switch is actuated. This system has been tested for 18 months using Rhine River water. No toxicant alarms have occurred during this time, and there have been no operational difficulties despite the turbid nature of the river water. Experiments with toxic substances (Table 1) do show that acutely toxic levels can be detected well before death occurs. A similar arrangement was used by Besch *et al.* (1977). Loss of rheotaxis is measured by the interruption of vertical light beams, and surfacing of the fish is measured by horizontal light beams. Also, a kinetic screen has been added at the downstream end of the chamber. This device produces electrical pulses when touched by the fish and provides a measure of the amount of time spent in the downstream area. Besch employed alternate periods of rest (generally 10 min of slow vertical current) and stress (5 min of strong longitudinal current) instead of a continuous downstream flow of test water. This automatic system detected phenol at sublethal concentrations and DDT at acutely toxic levels.

Two similar systems have been described, but data on their toxicant detection capabilities were not given. A monitoring system based on fish rheotactic responses was used at a water treatment plant on the Oise River near Paris (Vivier, 1972). When these fish were unable to swim against water flow after electrical stimulation, an audible alarm was generated. Another unit, which was used at a wastewater treatment plant in Sweden to monitor effluent quality, relied on strong light instead of electrical shock to promote the rheotactic response (Hasselrot, 1975).

Another way to test the ability of a fish to maintain its position in moving water is to place it in a narrow tube through which water is flowing and begin to rotate the tube. At a certain speed of rotation, the fish will be unable to compensate and will also start rotating (Lindahl & Schwanbom, 1971a,b; Lindahl *et al.*, 1976, 1977). Fish exposed to sublethal concentrations of methyl-mercuric hydroxide were less able to compensate than nonexposed fish. A similar effect on the minnow (*Phoxinus phoxinus*) occurred at levels of zinc less than those causing long-term effects on mortality, growth, reproduction, activity, or histology (Bengtsson, 1974). The potential of this technique as an early warning system is limited by the time needed for recovery of the fish between trials and by difficulties likely to be encountered in automating the testing procedure.

The locomotor behavior of fish has been found to be sensitive to a number of toxicants, and automatic monitoring techniques have been developed. Avoidance of toxic materials has been demonstrated repeatedly (for example, see Jones, 1947, 1948, 1951, 1952; Ishio, 1964; Sprague, 1964, 1968a,b; Hansen, 1969; and Fava & Tsai, 1976), but the response of a given species may vary widely for different materials. In one study, the green sunfish (*Lepomis cyanellus*) avoided only 8 of 40 toxic substrances tested (Summerfelt & Lewis, 1967).

Among the techniques available for automated monitoring of fish movement patterns is one in which a square matrix of photocells is positioned with respect to a test tank such that a fish in the tank continually interrupts photocell light beams corresponding to its location. The photocells are monitored by a minicomputer which provides rapid, detailed analysis of fish movements in response to environmental variables, including various toxicants (Kleerekoper *et al.*, 1970, 1972; Kleerekoper, 1977). Preference–avoidance behavior can also be monitored continuously by a small computer; the computer analyzes the signal from a video camera that scans a preference–avoidance chamber into which a single fish has been placed (Westlake & Lubinski, 1976; Lubinski *et al.*, 1977). The design of the chamber is such that water flowing across it remains in two halves with negligible intermixing although there is no physical barrier present. A toxicant can be introduced into either side and the movement of the fish automatically recorded. The usefulness of these methods as part of an early warning system may be limited by their mechanical complexity. A somewhat less complicated automated approach was developed by Cripe (1979). The positions of several fish in a toxicant gradient trough were detected by infrared light-emitting diode–phototransistor pairs and recorded by a microprocessor. In a 9 day test, pinfish (*Lagodon rhomboides*) avoided chlorine-produced oxidants at levels of 0.02–0.04 mg l^{-1} Should any of these systems be used as real-time toxicity monitoring devices, the complex behavioral patterns displayed by the fish may make it difficult to detect toxic materials rapidly.

A simpler approach monitoring toxic effects has been to measure oxygen consumption rates. Materials found to affect oxygen consumption include copper (O'Hara, 1971b), acid-mine drainage (Pegg & Jenkins, 1976) bleached Kraft pulp mill effluent (Davis, 1973), pulpwood fiber (MacLeod & Smith, 1966), benzene (Brocksen & Bailey, 1973), ammonia (Callahan, 1974), and methoxychlor (Waiwood & Johansen, 1974). Two methods for measuring oxygen consumption are well suited for automation. Oxygen electrodes may be used to measure oxygen uptake in a flow-through system (for example, O'Hara, 1971b, Rosseland, 1976; and Marais *et al.*, 1976) Determination of oxygen levels in the water before and after passage through the test chamber combined with a known flow rate of water permits computation of oxygen consumption. Voltage signals from the oxygen electrodes can be handled on a continuous basis by a computer or other data analysis device. An alternative technique involves the use of an electrolysis cell which can be used automatically to replenish oxygen depleted in the atmosphere inside a sealed respiration chamber (Callahan, 1974; Tackett *et al.*, 1974). The amount of electricity used by the cell is directly related to the amount of oxygen used by the fish in the chamber and can also be recorded automatically. Oxygen levels in the test chamber can be maintained at any desired level. One disadvantage is the need to stop water flow through the chamber while oxygen consumption is being determined. In any case, consideration must be given to the large number of variaoles affecting oxygen consumption. Among these are temperature (Beamish & Mookherjii, 1964), oxygen level (Beamish, 1964b), day length and reproductive development (Burns, 1975), and activity level (swimming speed, Brett, 1964). Standardization of test conditions would be necessary to prevent false warnings caused by responses to these variables.

Two systems have already been developed to indicate toxicity based on changes in the oxygen consumption of fish. In one arrangement, the difference in dissolved oxygen between water entering and leaving a chamber containing several fish was monitored. Too small a difference was taken as indication that the fish had died and an alarm was sounded (Kitsutaka, 1974). In another system, fish were confined individually in electrolytic-type respirometers through which water flowed continuously. The flow was stopped automatically during the measurement of oxygen consumption and then restarted. A data acquisition system stored the data from each fish on paper tape or magnetic tape. In laboratory tests, sublethal levels of ammonia and two aircraft firefighting foams caused significant changes in oxygen consumption (Callahan, 1974).

The ventilatory movements of fish offer two other means of assessing toxic effects: first, because oxygen consumption is related to the movement of water over the gills produced by the ventilatory movements; and second, because the gill tissues are delicate and thus susceptible to toxic materials in the water. Ventilatory parameters commonly observed include opercular movement rates and coughing rate A cough is a rapid movement of the ventilatory apparatus which usually results in a reversal of water flow over the gills and is a response to irritation of the gill surface (Hughes, 1975). Many aspects of monitoring coughs were reviewed by Drummond & Carlson (1977)

Several techniques developed to monitor fish ventilatory movements were reviewed by Heath (1972) The method found to be simplest and causing the least stress on the fish was to monitor the electrical signals generated during ventilatory movements by means of dual external electrodes affixed to opposite ends of a test chamber (Kleerekoper & Sibakin, 1956; Spoor *et al.*, 1971). With sufficient amplification, this electrical signal may be recorded by, for example, a physiograph. Lonsdale & Marshall (1973) used an electrode arrangement in conjunction with an FM transmitter to record ventilatory signals from trout.

It has long been known that ventilatory patterns are affected by toxic substances (Belding, 1929). A large number of materials have been shown to affect ventilatory and or coughing rates at sublethal levels, including heavy metals (Sparks *et al.*, 1972; Sellers *et al.*, 1975; McIntosh & Bishop, 1967; Morgan, 1977), pesticides (Schaumburg *et al.*, 1967; Lunn *et al.*, 1976; Morgan 1977), and complex waste effluents (Schaumburg *et al.*, 1967; Walden *et al.*, 1970; Davis, 1973; Howard & Walden, 1974; Thomas & Rice, 1975; and Carlson & Drummond, 1978). Other materials include coal dust, wood pulp and kaolin (Hughes, 1975) and acetone, ethanol, and propylene glycol (Majewski *et al.*, 1978). There is some indication that coughing rates may be useful as a short term indicator of long term toxic effects; cough responses in brook trout (*Salvelinus fontinalis*) occur at concentrations of copper and mercury close to chronically toxic levels (Drummond *et al.*, 1973, 1974). Maki (1980) found that the ventilatory rates of bluegills were altered by levels of surfactants that caused chronic toxicity (reproductive effects) in the fathead minnow (*Pimephales promelas*). As with oxygen consumption, various environmental variables can alter ventilatory patterns. Among these are dissolved oxygen (Hughes & Saunders, 1970), temperature (Hughes & Roberts, 1970), activity level (Heath, 1973; Sutterlin, 1969), pH (Hargis, 1976), turbidity (Horkel & Pearson, 1976), season of the year (Beamish, 1964a), and even seismic shock (Sparks & Cairns, 1972).

Among the several continuous monitoring systems that utilize fish ventilatory signals, the simplest have timing devices which periodically activate a physiograph on which ventilatory signals are recorded (Sparks *et al.*, 1972; Drummond *et al.*, 1974). The recordings must still be analyzed manually, a procedure which requires a great deal of time and may introduce subjectivity into the counting of coughs, especially if several persons are involved (Maki, 1980).

These problems have been alleviated partially in a recently developed automated electronic system (Morgan & Kuhn, 1974; Morgan, 1977). Twelve fish in individual chambers are monitored by an apparatus that produces a d.c. voltage proportional to the ventilatory rate every minute (coughing rates are not determined). Data gathered from each fish over a five day period are used to set a 99% upper confidence limit on the ventilatory rate for that fish. A voltage level proportional to this limit then is set into a device which activates an alarm light should any fish exceed its confidence limit. With a criterion for toxicant detection set as the response of at least 60% of the fish, a wide variety of materials were detected within 24 h at levels of 5 10% of their 48-h LC50s (see Table 1).

Relatively few of the laboratory toxicity monitoring systems described thus far have been tested under field conditions. One which has is a computer based system which automatically monitored changes in the ventilatory patterns of fish exposed to a dilution of an industrial waste effluent as it flowed into a river (Westlake *et al.*, 1976; Westlake & van der Schalie, 1977; and van der Schalie *et al.*, 1979). No known toxic spills occurred in the effluent during the operation of this system, but acetone added to the effluent waste caused responses from the fish within an hour at concentrations which peaked near the 96-h LC50. The major problem with this system was a large number of false responses from the fish. These apparently were caused by the effects of the harsh industrial environment on the monitoring equipment and to shortcomings in the design of the system. Gruber *et al.* (1977) describes a second generation fish monitoring unit which overcomes many of these deficiencies. This system is currently being tested at a new industrial site (Gruber *et al.*, 1978).

Work also is proceeding on the evaluation of devices which use fish to assess the quality of public water supplies. The Anglian Water Authority in England is currently establishing fish monitoring systems on three rivers. Each system will monitor a different parameter (initially the activity, behavior, or avoidance of rainbow trout), and each will be used in conjunction with existing automatic physical–chemical monitoring facilities (Price, 1978). A similar automated river monitoring system planned for England was described by Wallwork *et al.* (1977). Finally, an automated unit on the Rhine River in Germany (Kalweit, 1977) will monitor up to 15 water quality parameters including a fish testing device and a bacterial monitor similar to the one developed by Axt (1972, 1973a,b). These field tests will ultimately establish the usefulness of biological early warning systems.

The sensitivity to toxic substances of many aquatic organisms and the availability of many types of monitoring equipment indicate the real possibility for developing automatic biological monitoring systems. If the basic requirements given here can be met, the resulting systems could become useful tools in water pollution control.

SUMMARY

The summary (Cairns, 1976) written for *Biological Monitoring of Water and Effluent Quality* (Cairns *et al.*, 1976) still seems sufficiently appropriate as a summary of this article and is included here with permission of the American Society for Testing and Materials.

Biological monitoring implies regular or continuous assessment of one or more parameters and may be used to detect harmful conditions. One might credit the development of biological monitoring to the laymen who used people to test the food and

drink of kings prior to their consumption of it and to the miners who placed canaries in mines to detect the presence of obnoxious gases. Since the assassin with a vial of poison has been replaced by an assassin with a revolver or bomb and the canary has been replaced by a more sophisticated chemical monitoring device, these early forms of biological monitoring are no longer in vogue. However, biological monitoring has attacted increasing attention in recent years in the field of water pollution for two primary reasons: (1) the response of organisms to various toxicants is often mediated by the chemical-physical qualities of the receiving system water and these may change rather frequently, thus changing the toxicity of the material; and (2) there is a possibility of interactions with other waste discharges which will markedly alter the nature of the toxic response. Thus, both environmental interactions and interactions with other potential toxicants may substantially alter the response of an organism from that obtained with a pure chemical in a laboratory situation. However, most of the standards have been derived from laboratory data and, therefore, when applied nationally may be either too high or too low for the circumstances existing at a particular site of discharge. Only recently has the scientific community become seriously interested in biological monitoring and, even now, only a small fraction of the total global scientific effort is involved in such studies. However, the representation at this symposium was an indication of widespread geographic interest even though biological monitoring does not involve many scientists now. The growing interest in the field evidently far exceeds present expenditures of money, time, or effort.

Although there appears to be fairly widespread dissatisfaction with the present systems of regulating waste discharges, there is no consensus support for alternative approaches. Three alternatives seem to be available: (1) improvement of current practices using all available scientific information and disseminating this information through such books as *Water Quality Criteria of 1972* [1]* and *Principles for Evaluating Chemicals in the Environment* 1975 [2]; (2) use of maximum feasible treatment with available technology on each waste discharge with the assumption that technology will improve substantially and therefore environmental conditions will improve; and (3) use of biomonitoring techniques to fully permit nondegrading utilization of the receiving or assimilative capacity of ecosystems for waste discharges and at the same time protect them from deleterious effects. Alternative (2) is espoused by a variety of groups, but it is unsatisfactory to others for a number of reasons. The two most prominent reasons are: (1) reconstruction of the treatment system might be necessary with each major technological advance, and there is no way of predicting when all this would end or how frequently changes would be necessary; and (2) there might be no measurable or demonstrable environmental benefits resulting from the improved waste treatment even though it might be considerably more expensive than the system replaced. Objections to biological monitoring are that there is no strong body of evidence indicating that it will work as is supposed to nor is there a sizable body of evidence on cost. If the cost is high, small industries might be priced out, and, in any case, the monitoring must still be accompanied by a well functioning waste treatment system. There will also be difficulties in formulating appropriate legislation.

Neither of the systems alternative to the one presently used for regulating waste discharges is likely to replace it without massive evidence of superior effectiveness at acceptable cost. This evidence of superiority should be strong in three principal areas: (1) scientific justifiability, (2) operational reliability, and (3) cost effectiveness. Before addressing Areas 2 and 3, Area 1 must be supported to the satisfaction of a substantial segment of the academic community. It is our hope that this publication will provide sufficient evidence for the academic community to make a tentative judgment on the efficacy of biological monitoring and will identify questions which must be answered before substantial confidence can be placed in biological monitoring. In addition, papers in this publication will provide a means for preliminary assessment of the operational reliability of biological monitoring systems. More examples should be forthcoming so a thorough evaluation may be made of both the conceptual soundness and the operational capabilities of various biological monitoring methods. Although some indirect evidence will be available for the cost effectiveness of these methods, it would be presumptuous to attempt a detailed evaluation at such an early developmental stage for the field. However, it appears likely that the use of minicomputers and other technological innovations will result in a substantial saving as they have in other areas of applied science.

Scientific merit

There are many questions regarding the conceptual soundness of biological monitoring methods that must be answered more definitively before these methods will be accepted by the academic community, potential industrial users, and regulatory agencies. For an "in-plant" monitoring system, the most important questions are probably the ones listed here.

1. Will the system detect spills of lethal materials before they reach the receiving waters?
2. If only one organism is used as a sensor (for example, the bluegill sunfish), will this organism be so much more tolerant to the particular toxicant in question that it will pass undetected and harm other members of the aquatic community in the receiving system (for example, algae and invertebrates)?

* The numbers in square brackets refer to the list of references appended to this summary

3. Is it possible to monitor chemical–physical parameters and achieve the same results at lower cost and greater efficiency?

4. Since the biological response alone will not identify the particular toxicant causing the response but only indicate that some deleterious material is present, is it possible to couple a chemical physical monitoring system with a biological monitoring system that will expedite the identification of the particular deleterious component causing the warning response?

5. Will a false signal cause an expensive shutdown of the plant or an undue expenditure of time and effort by the waste control personnel?

6. Should an organism indigenous to each receiving system be used, which would require a long site-specific developmental period for each new drainage basin, or can some "all-purpose" organism, such as the bluegill, be used for all types of systems (or perhaps one organism for a warm-water and one for a cold-water system)?

7. Is it possible to use in-plant biological monitoring systems to detect the presence of spills of materials having either acute lethality or long-term effects or only the former?

8. Are the in-plant monitoring systems only for very large industries with sizable waste control staffs, or is it possible to develop compact miniaturized reliable in-plant monitoring systems that can be used by persons inexperienced in monitoring without undue expenditure of time, etc.?

Questions such as these related to the utility and scientific justifiability of monitoring will undoubtedly be asked by persons representing a regulatory agency and the industrial point of view elsewhere in this book. Doubtless, many additional questions on efficacy will be raised by other authors. The eight questions just presented were raised primarily because they are important in the acceptance of biological monitoring, and they indicate how far we must yet go in developing the methods. For example, it took nine years before the apparatus and methods for in-plant biological monitoring were ready for use on a trial basis in an industry (discussed elsewhere in this book by Westlake and van der Schalie). During this developmental period, the apparatus has gradually evolved from strip charts and other visually examined data recording systems to the present computer-interfaced, automated data recording system. In addition, a variety of responses were examined (for example, the coughing response [3], which is presently being used by the Environmental Protection Agency (EPA) [4]). In the course of this investigation, it became apparent that any characteristic of an organism's normal life which was evident continually (for example, respiratory signals of fish) could be used effectively in a biological monitoring system. A key factor in successful use was the ability to quickly and reliably detect deviations from the normal condition. Although some attention has been given to the development of statistical methods for this purpose (for example, see Ref. [5]), much more attention must be given to this critical area.

Industrial utility

Although it is essential to demonstrate the scientific merit of biological monitoring systems and although this is a formidable task, it will be neither as time consuming nor as expensive as demonstrating the industrial utility of biological monitoring systems. That is, proving with reasonable certainty that the hypotheses upon which the systems are founded are valid will probably be substantially less difficult than demonstrating their utility for a variety of industrial operations encompassing a diverse array of ecosystems and receiving water conditions. This will require an extended developmental period during which industrial and academic communities must collaborate to provide demonstration situations. The companies that do this will acquire a staff which is knowledgeable in both the theory and the working requirements of biological monitoring systems as well as their faults and strengths. A company installing a unit in the early stages of development of biological monitoring systems will certainly have much more aggravation than the companies which follow. However, any pioneering effort has its advantages and its disadvantages, and biological monitoring is no exception. One of the most difficult aspects of the demonstration projects, at least if they are to provide information in the public domain, is that various industrial spills must be identified and documented. In short, if a company has spills of deleterious materials, it must be prepared to acknowledge this if there is to be general conviction that biological monitoring systems can detect the spills. In addition, all of the difficulties encountered in installing a biological monitoring system must be clearly identified and disclosed so that mistakes are not repeated over and over. Public disclosures of this sort are painful but are necessary in order to establish creditability. This is, of course, much easier for the academic community, which is accustomed to talking about both experimental failures and successes, than for the industrial community, which may rightly feel that regulatory agencies will use information about spills to an industry's disadvantage.

Regulatory agencies

Undoubtedly, the most dramatic change produced by the general adoption of biological monitoring systems as environmental quality control units would result if monitoring requirements were to replace existing legislation directed toward either the regulation of specific toxicants individually at their point source or maximum feasible treatment. This is only one of three possible scenarios. A brief outline of each of these three scenarios follows.

Scenario 1

Standards for water quality continue to be directed primarily toward regulation of amounts of toxicants and physical changes (for example, thermal loading), and the biological monitoring systems are used as a supplemental safeguard to prevent "fish kills" and other highly visible catastrophes resulting from accidental spills and other emergency situations. Even if waste dischargers conform to all the limitations imposed by present standards for discharges of toxic materials, heated wastewaters, and the like, there is a high probability that a certain number of deleterious biological effects will result in aquatic systems from both synergistic interactions and enhancement of toxic effects due to natural environmental changes. There is abundant evidence in the literature to document both the existence of synergistic interactions and the effects that changes in natural environmental parameters such as hardness, pH, temperature, and dissolved oxygen concentration can have upon the response of organisms to toxicants. Despite the probability of occasional kills of aquatic organisms occurring even though discharges conform to existing standards, most waste dischargers will probably rely solely upon conformance with existing standards to protect them against legal action and will not use either in-plant or in-receiving system biological monitoring systems as an additional safeguard unless required to do so by law or regulation. Waste dischargers most likely to use in-plant or in-receiving system biological monitoring units even though not legally required to do so are: (1) those with wastes impossible to analyze either because appropriate analytical methodology does not exist or because the standards are set below the level of detectability with current methodology; (2) those located in areas of extreme biological sensitivity such as spawning grounds, commercially valuable fisheries resources consumed by humans, and the like; (3) large industries with many plant locations for which adverse publicity is likely to have a national impact; (4) certain government installations desirous of providing the best possible protection to the environment for both public relations and safety reasons; and (5) commercial organizations wishing to have a good stance vis-a-vis the environment.

Although this is a somewhat lengthy list, it is unlikely that at the present time the total percentage of dischargers falling into one or another of the aforementioned categories will be a very substantial percentage of the total waste dischargers. Whatever may be the capital investment requirements and operating costs of biological monitoring systems, it is likely that little money will be expended by most waste dischargers to install such systems unless their installation is required by law. Even then, it is likely that substantial delays in installation would result because of lack of trained personnel and management's inexperience with these new methods. However, the number of inquiries we have had from industry and evidence received from other groups both in this country and in Europe suggest very strongly that enough waste dischargers fit into one of the aforementioned categories to provide a thorough testing of the efficacy of biological monitoring in a variety of situations. Thus, the most probable course of development for biological monitoring is that a few waste dischargers will then furnish a data base which will enable the methodology to be evaluated more fully than is possible in a strictly academic situation. This is already beginning to happen as papers in this publication show.

Scenario 2

Standards and legislation continue to follow the pattern of the recent past as mentioned under Scenario 1, but provision is made for exceptions to these standards if positive evidence of no biological harm resulting from discharges in excess of the standards can be furnished on a continuing basis. Further legislation might be enacted requiring that industries wishing to develop sites in recreationally or aesthetically desirable areas, or those with particularly valuable commercial or sport fisheres, carry out biological monitoring in addition to complying with all of the other requirements because of the uniqueness or high value of the particular ecosystems. An industry contemplating waste discharges in such an area would then have to determine whether the additional cost of biological monitoring would so decrease the attactiveness of the particular site in question as to render alternative sites more desirable. In view of the enormous costs involved in plant construction, land acquisition, planning and so on, it seems unlikely that the relatively small costs of biological monitoring would be often a decisive factor, but sometimes this could be the case. In the latter event, the cost of improved waste treatment could probably not be readily afforded either and, therefore, it would be in the best interests of all concerned if the marginally profitable industry were to locate in a less vulnerable or less ecologically desirable site. Although not quite so specific, there are some portions of the Federal Water Pollutional Control Acts Amendments of 1972 (Section 316) which indicate that appropriate evidence may result in exceptions to or modification of existing requirements. Thus, once the methodology for biological monitoring has become more widely accepted by the academic community, regulatory agencies, waste dischargers, and the general public, it is quite likely that modifications of legislation of this sort will become more frequent.

Scenario 2 might also be considered a stage in the evolution of the acceptance of biological monitoring which might naturally follow the stage represented by Scenario 1. It would also represent an introduction into the regulatory process of the types of evidence generated by biological monitoring systems, and this would enable regulatory agencies to develop a capability for dealing with such information on a trial basis. Thus, personnel capabilities as well as other capabilities for information transfer, storage, and routine data recording could be established on a small scale.

Scenario 3

The present standards based primarily on chemical–physical parameters are shifted so that the biological parameters become the principal parameters with chemical physical parameters still being measured but no longer the prime determinants regulating waste discharges. In short, the response and condition of the biological system will be the prime regulator of discharges, and these must be kept within bounds that will not damage the biological integrity of the receiving system. Some indications of this view have appeared elsewhere [6]. Among the definitions of biological integrity was the maintenance of the structure and function characteristics of a particular, relatively undisturbed ecosystem or the return to a level of structure and function considered for a disturbed ecosystem.

In Scenario 3, the environmental quality control would be implemented by a series of biological and ecological standards designed to directly measure the integrity of each ecosystem into which the wastes are being discharged. These standards would involve both the structure and function of the ecosystem. This would require regional management, regional specifications for integrity, an extensive biological–chemical–physical monitoring network in each region, a central authority capable of taking immediate corrective action without waiting for a court decision, and possibly a user tax to pay for the operation of the quality control system based on the degree of use made of the ecosystem (this would take environmental use out of the economic "free goods" category).

It is difficult to imagine this very profound change occurring without some intermediate steps between the type of environmental quality control just proposed and those now in use. At the very least, Scenarios 1 and 2, or some version of them, would have to take place before Scenario 3 could become a reality. An entirely different set of attitudes and procedures would be necessary, and to a large extent, different types of training. A major consequence of following this form of environmental quality control would be to associate the standards on a continuing and frequent basis with the condition of natural ecosystems. Ideally, it would also develop a cooperative rather than an adversary relationship between users and protectors, but this may be too idealistic for the real world.

CONCLUDING REMARKS

The purpose of this symposium is to rigorously examine biological monitoring to determine: (a) its scientific justification, (b) its reliability, (c) the general acceptance of the methodology by the academic community, industry and regulatory agencies, (d) its cost, and (e) if it is found to be a sound methodology, the reasons why it is not being more widely accepted by regulatory agencies and industry.

It is to be hoped that this symposium will focus more attention on biological monitoring than such monitoring has received in the past and that a scientifically justifiable body of information will be developed for either accepting or rejecting the concept. If the concept is found acceptable, it is to be hoped that regulatory agencies and industry will be equally objective in their appraisal and subsequent adoption of the sound aspects of biological monitoring even if this means abandoning well entrenched bureaucractic positions.

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and Edwin E. Herricks

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Almost anything placed in water will be colonized by aquatic organisms, whether the substrate be glass, a ship hull, a dock piling, or even an industrial cooling system through which water is passed. Theoretical ecology can benefit from studies of colonization of artificial substrates and other processes associated with species accrual. Better management practices to keep ship hulls and cooling pipes free of encrusting organisms can be developed if the colonization is better understood. This is but one of the many immediate practical benefits.

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THE RECOVERY PROCESS IN DAMAGED ECOSYSTEMS

Edited by **John Cairns, Jr.,** Distinguished Professor and Director, Biology Department and Center for Environmental Studies, Virginia Polytechnic Institute and State University

This new book focuses attention on the ecological processes involved in the recovery of damaged ecosystems. Offers encouragement in an era when environmental damage is severe because it shows that damaged ecosystems can be restored to a more ecologically acceptable condition, although it is highly unlikely in most cases that restoration to original condition will be possible. Lays the foundation for thinking about the components of the recovery process. Relates the recovery process to well established ecological phenomena such as succession; speculates about the nature of the recovery process and whether it will be possible to quantify it.

Poses these fundamental questions: 1) How is recovery defined? 2) What criteria are important in measuring recovery? 3) Do societal perturbations (e.g., strip mining) have a different effect upon natural communities than natural perturbations (e.g., floods)? 4) How much restoration is possible, acceptable, and economically feasible? 5) How much disturbance should be permitted at any one time?

Useful to regulatory agencies, industrialists, developers and all persons required to cope with the aftermath of a catastrophic spill of oil, toxic chemicals, or other hazardous materials. Gives an appreciation of the magnitude of restoring a damaged ecosystem both in costs and time.

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Citizens Against Toxic Waste Foundation
9 North Third Street, Warrenton, VA 22186
(202) 347-2936

Over 26,000 toxic waste sites are poisoning us and the groundwater we drink. You ask "What can I do about it?" Let me tell you what we're doing and how you can help.
John V. Albertella

Dear Friend,

I don't want to alarm you, but our research tells us that <u>at least one EPA-listed toxic waste site is located right near your home</u>!

To find out the toxic waste site name, location and other vital information about the site, use the FREEDOM OF INFORMATION ACT Request Form I've enclosed.

As an American taxpayer, you have the <u>right</u> to know about health risks that may affect you and your family because of contaminated drinking water and toxic waste sites near your home.

When we receive your FREEDOM OF INFORMATION ACT Request to get the <u>facts</u> about the very real threat to your family, Citizens Against Toxic Waste Foundation will immediately transmit your Request to the Environmental Protection Agency (EPA). By law, the EPA <u>must</u> respond to your request.

So please sign and mail us your specially prepared form today. <u>Your family's health and the fate of this whole toxic waste crisis could depend on it.</u>

This is incredible: here we are in 1987 -- approximately 10 years since the discovery of the "Love Canal" toxic waste site disaster; 7 years and $1.6 BILLION after the "Superfund" was created by the EPA to supposedly clean up waste sites like the one near your home.

Practically nothing has been done about it, and I'll bet that before reading my letter, <u>you never even knew there was an environmental time-bomb ticking near your home</u>.

Well, don't feel like you're all alone. The sad fact is that there are millions of Americans, like you, who don't know (more importantly, <u>who were never told</u>) they live near official EPA-listed toxic waste sites.

And the horror is that the list of sites is growing larger every day. At last count there were more than 26,000 toxic waste sites in the country on the official EPA list!

Experts now estimate that drinking water for **half** the American population is contaminated by toxic waste dumping. Every state, nearly every county in the nation is in danger!

(Over, please)

That's why Citizens Against Toxic Waste Foundation has mounted this unprecedented FREEDOM OF INFORMATION campaign -- for your protection and for that of your family.

So please -- in order to file your FREEDOM OF INFORMATION ACT Request at the EPA offices in Washington, D.C., send your signed Request to Citizens Against Toxic Waste Foundation right away.

Citizens Against Toxic Waste Foundation is a non-profit research and education organization. We sponsor research and other information programs like this one to alert you and others to the urgent dangers of toxic chemicals and nuclear wastes.

And in the past several months, our attention has focused more and more on the relationships between our drinking water, toxic waste contamination and deadly disease rates among men, women and children right here in the United States.

Report after report indicates a strong link between the water you drink and your chances of being exposed to harmful contamination.

In fact, on September 22, 1986 the families of eight leukemia victims in Woburn, Massachusetts were awarded $1 million each in an out-of-court settlement with the W.R. Grace Company who was accused of polluting their water supply with potentially cancer-causing chemicals.

There is now no doubt that toxic wastes present a potentially irreversible threat to our groundwater (drinking water) supplies and our personal health.

Toxic waste contamination is shutting down thousands of wells and local water supplies throughout our country and pollution is spreading like a stain:

- ** In its 1982 survey of large public water systems served by groundwater, the EPA found 45% of them were contaminated with organic chemicals.
- ** In New Jersey, for example, every major groundwater supply is affected by chemical contaminants.
- ** A 1980 study of 350 hazardous waste sites found they had caused 168 cases of groundwater contamination in 32 states, forcing the closing of nearly 500 wells.

So what is the EPA doing about it? Unfortunately, simply not enough.

There are more than 26,000 toxic waste sites. But so far, the EPA has put only 951 of them on their "priority list". And of these 951, they have only cleaned up 13! After 7 years and billions of dollars, that's all that's been done!

But the worst news is that after the EPA used $34 MILLION to clean up the Stringfellow Acid Pits near Los Angeles (the nation's most serious site), the clean-up effort so far has failed, and a giant plume of deadly poison still moves toward the Los Angeles underground water supply at an **unstoppable** rate of 3 feet per day!

Pretty scary, isn't it?

You bet it is. This is why all of us here at Citizens Against Toxic Waste Foundation are doing everything humanly possible to get the EPA to speed the clean-up.

Now I'm not going to tell you that we are going to solve this catastrophe over-

(More)

night. To really change things, we're going to need the participation of a lot of Americans, including you.

It's very important that everyone I'm writing to participates in our FREEDOM OF INFORMATION campaign. Not nearly enough is being done to safeguard our health -- federal, state, and local agencies have proven by their snail's pace that not until we can rally tens of thousands, even hundreds of thousands of Americans around our efforts, will we finally succeed in keeping society's wastes out of our drinking water and our backyards.

By filing your FREEDOM OF INFORMATION ACT Request with us, you are taking an important first step in tackling a problem that could well be affecting you personally.

I also urge you to take a second and equally vital step.

Citizens Against Toxic Waste Foundation needs your help to continue this and other projects. A tax-deductible gift of $12, $15 or $25 or more would be a big help to us.

There is no other group like us working on a national scale exclusively on this issue. And it's easily going to take as much money as friends like you can afford to give.

So -- please -- send a contribution today.

We do not accept government subsidy or support. So it's very important that you help.

Because, quite frankly, the alternatives are terrifying.

You and I are facing a problem that is well on its way to becoming the next great crisis in America. Unless all of us get involved now, I'm concerned that as bad as it is today, it could get a whole lot worse.

The EPA, other government agencies, and private industry have already shown that without a massive public outcry, they will do little or nothing at all.

Our most pressing need right now is to build this massive groundswell of citizen support from people like yourself who live near a toxic waste site, and who obviously have a very personal stake in the outcome of our efforts.

If you won't help support us, I frankly don't know who will. So can we count on your help now? Please be as generous as you can.

Sincerley,

John Albertella

John V. Albertella
Executive Director

P.S. whatever you do, please send your FREEDOM OF INFORMATION ACT Request back to us today. The EPA is required by Federal law to answer your request. Should you not hear from them within 30 days, please drop me a note. It's important you get this information.

TOXIC WASTE FACT SHEET

- Chemical contamination of our drinking water and ground water supplies has become the most serious crisis of our time. Industry is producing *270 million tons every year of toxic waste* containing chemicals such as dioxin, vinyl chloride, PBB's, PCB's, lead, mercury and arsenic—and 80% of them are dumped into the ground and ultimately endanger our water.

- *The EPA has already inventoried 26,457 toxic waste sites*—dumps and sources. The Congressional Office of Technology Assessment estimates that *the number will reach 36,000* before it's over.

- *87% of the 24,000* pits and ponds used to store toxic industrial chemicals lie directly atop vulnerable underground drinking water supplies.

- In a 1982 survey of large public water systems served by groundwater, the EPA found *45% of them were contaminated with chemicals.* Forty-nine percent of Americans drink groundwater.

- According to the Council on Economic Priorities *8 out of 10 Americans live near a toxic waste dump or source.* These toxic sites can and do contaminate ground water.

- Cleanup has been slow and ineffective. After 6 years and $1.6 Billion the *EPA has completed work on only 13 sites.* The $9.6 Billion passed in 1986 is for 5 years. Meanwhile, the Office of Technology Assessment estimates the cleanup may eventually cost more than $100 Billion.

TOXIC WASTES AND YOUR HEALTH

Although it is extremely difficult to predict the harm of long-term exposure to low levels of toxic chemicals in drinking water, some effects have been observed in humans and animals. Some people are more susceptible than others. *Children for example, are often more vulnerable* because of their lower body weight, growing body organs, and faster respiratory rate. Genetic factors, general health, and lifestyle (including smoking and diet) can also affect susceptibility.

Arsenic: liver, kidney, blood and nervous system damage and cancer.

Benzene: chromosomal damage in both humans and laboratory animals. Benzene affects blood and immune sysems to cause anemia, blood disorders, and leukemia.

Cadmium: kidney damage, anemia, pulmonary problems, high blood pressure, possible fetal damage and cancer.

Carbon tetrachloride: liver, kidney, lung, and nervous system damage and cancer.

Chloroform: liver and kidney damage and suspected cancer.

Chromium: suspected cancer from some forms.

Dioxin: skin disorders, cancer, and mutations. Dioxin is extremely toxic.

Ethylene dibromide (EDB): male sterility and cancer.

Lead: headaches, anemia, nerve problems, mental retardation and learning disabilities in children, birth defects, and possibly cancer.

Mercury: nervous system and kidney damage.

Polychlorinated biphenyls (PCBs): liver damage, skin disorders, gastrointestinal problems, and suspected cancer and mutations.

Trichloroethylene (TCE): in high concentrations, liver and kidney damage, skin problems, depression of the contractibility of the heart, and suspected cancer mutations.

Vinyl chloride: lung, liver, and kidney damage; pulmonary and cardiovascular effects; gastro-intestinal problems; cancer.

FREEDOM OF INFORMATION ACT REQUEST

Issued by Citizens Against Toxic Waste Foundation
(Pursuant to 5 U.S.C. 552)

TO: Freedom of Information Office
United States Environmental Protection Agency
Washington, D.C. 20460

RE: OFFICIAL EPA LISTED TOXIC WASTE SITE OR SITES LOCATED WITHIN THE ZIP CODE AREA OF THIS ADDRESS:

FROM:

Name ______________________________

Address ______________________________

City/State/Zip ______________________________

Please PRINT clearly.

DESCRIPTION OF RECORDS REQUESTED

In accordance with federal law, the Freedom of Information Act of 1966, I, the undersigned, wish to know any and all information you presently have regarding the toxic waste site (and all other sites) located near my home. Please include information regarding the possible health risks to myself and my family, what precisely is contained in this waste site, and how and when I can expect it to be cleaned up.

DO NOT COMPLETE
Request No:
Received at CATW/Date:
Transmitted to EPA/Date:
/By:

Respectfully,

____
Signed

____
Date

IMPORTANT DOCUMENT • IMMEDIATE RESPONSE REQUESTED

—Please Do Not Detach—

Citizens Against Toxic Waste Foundation
9 North Third Street, Warrenton, Virginia 22186
202-347-2936

CONTRIBUTION REPLY

TO: John V. Albertella
Citizens Against Toxic Waste Foundation
9 North Third Street
Warrenton, VA 22186

(Please make your tax-deductible check payable to CITIZENS AGAINST TOXIC WASTE FOUNDATION.)

Dear John,

☐ **YES,** please forward my FREEDOM OF INFORMATION ACT REQUEST to the EPA.

☐ To help CITIZENS AGAINST TOXIC WASTE FOUNDATION continue this vitally important FREEDOM OF INFORMATION campaign and help speed the cleanup of our nation's toxic wastes, I am enclosing my check for:

☐ $100 ☐ $50 ☐ $25 ☐ $15 ☐ $ ______

I understand for my contribution of at least $25 I can expect to receive a summary report of this FREEDOM OF INFORMATION campaign, plus a quarterly newsletter detailing our efforts to protect our groundwater and our health from toxic waste contamination.

Obviously, since you live near an official EPA listed toxic waste site, you have a very personal interest in all of this. So, I'm especially hopeful you'll realize our need to ask you for a donation. It costs us at least $3.50 just to handle, process and then follow-up each Freedom of Information Act Request with the EPA. Could you possibly add this $3.50 to your contribution amount to help defray our administrative costs? Any amount you can give will be greatly appreciated and put to good use.

John V. Albertella

Multispecies Toxicity Testing

Edited by **John Cairns, Jr.**, University Center for Environmental Studies, Virginia Polytechnic Institute and State University, Blacksburg, VA

"The editor has done much to illuminate the dark and murky field of the behavior of natural species under stress. His approaches, conceptually, to understanding of behavior are deep and are gradually revealing practicable surrogate parameters—long sought in the important understanding of the universe of ambient water flora and fauna."

Abel Wolman,
Emeritus Professor
Department of Sanitary Engineering & Water Resources,
The Johns Hopkins University

The purpose of this volume is to discuss in detail the scientific, regulatory and industrial use and problems associated with the development of multi-species toxicity tests. Some case histories are included to provide the reader with examples of how multispecies toxicity tests work. Also included are sections on such problems as quality assurance and replication, as well as the variability and complexity of river ecosystems as an illustration of the kinds of systems currently being protected by multispecies tests.

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Environmental Hazard Assessment of Effluents

Edited by **Harold L. Bergman**, Department of Zoology and Physiology, The University of Wyoming, Laramie, WY; **Richard A. Kimerle**, Monsanto Industrial Chemicals Co., St. Louis, MO; **Alan W. Maki**, Research and Environmental Health Division, Exxon Corporation, East Millstone, NJ

"This outstanding volume, which is the result of the contributions of 41 professionals from government, industry and academia, presents, for the first time, a complete overview of the complex water quality problems presented by effluents. . . . The wealth of information in this book makes it a valuable addition to the library of anyone involved in water quality work, and its well rounded presentation makes it an excellent source book for students."

—Robert E. Reinert
Professor of Fisheries
The University of Georgia

This volume has been prepared to meet the need for improved methods to assess the hazards of chemicals in the aquatic environment. The subject deals with effluents, that is, the complex mixtures of chemicals that can include liquid wastes, dredged materials, sewage and industrial sludges, solid waste leachates, and other complex materials that may enter aquatic ecosystems.

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ARE SINGLE SPECIES TOXICITY TESTS ALONE ADEQUATE FOR ESTIMATING ENVIRONMENTAL HAZARD? *

JOHN CAIRNS, JR.

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(Received 23 February, 1983)

Abstract. Most biologists agree that at each succeeding level of biological organization new properties appear that would not have been evident by even the most intense and careful examination of lower levels of organization. These levels might be crudely characterized as subcellular, cellular, organ, organism, population, multispecies, community, and ecosystem. The field of ecology developed because even the most meticulous study of single species could not accurately predict how several such species might interact competitively or in predator-prey interactions and the like. Moreover, interactions of biotic and abiotic materials at the level of organization called ecosystem are so complex that they could not be predicted from a detailed examination of isolated component parts. This preamble may seem platitudinous to most biologists who have heard this many times before. This makes it all the more remarkable that in the field of toxicity testing an assumption is made that responses at levels of biological organization above single species can be reliably predicted with single species toxicity tests. Unfortunately, this assumption is rarely explicitly stated and, therefore, often passes unchallenged. When the assumption is challenged, a response is that single species tests have been used for years and no adverse ecosystem or multispecies effects were noted. This could be because single species tests are overly protective when coupled with an enormous application factor or that such effects were simply not detected because there were no systematic, scientifically sound studies carried out to detect them. Probably both of these possibilities occur. However, the important factor is that no scientifically justifiable evidence exists to indicate the degree of reliability with which one may use single species tests to predict responses at higher levels of biological organization. One might speculate that the absence of such information is due to the paucity of reliable tests at higher levels of organization. This situation certainly exists but does not explain the lack of pressure to develop such tests. The most pressing need in the field of toxicity testing is not further perfection of single species tests, but rather the development of parallel tests at higher levels of organization. These need not be inordinately expensive, time consuming, or require any more skilled professionals than single species tests. Higher level tests merely require a different type of biological background. Theoretical ecologists have been notoriously reluctant to contribute to this effort, and, as a consequence, such tests must be developed by this and other organizations with similar interests.

1. Introduction

Although this discussion may appear hostile to single species toxicity testing efforts, it is not intended to be. Single species tests are exceedingly useful and are presently the major and only reliable means of estimating probable damage from anthropogenic stress. Furthermore, a substantial majority, perhaps everyone at this meeting, is certainly aware of the need for community and system level toxicity testing. How then does one account for the difference between awareness and performance? As an illustration of why such a difference exists, consider this scenario from a hypothetical workshop entitled 'The Contributions of Theoretical Ecology to Pollution Assessment'. On the first day of this workshop, reassuring exchange occurs among participants on ideas of energy flow, ecosystem dynamics, multiple aggregate variables, niche packing, and the like. On

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the second day, the group who must make use of this information confronts the theoretical ecologist with one or more site-specific problems and asks specifically how theoretical ecology can be used in a particular situation. It usually becomes abundantly clear that no system level measurements exist on which a concensus occurs among theoretical ecologists on use, interpretation, validity, and predictive value! Following this exercise, a retreat ensues to measurements associated with single species or at least those that are clearly not ecosystem level parameters. This is usually accompanied by a call for more research. Probably well over half the participants at this present symposium have attended such workshops; many have probably attended a number of such meetings with roughly similar scenarios.

The call for more research before specific recommendations can be made usually falls on deaf ears. This is a pity because truly more research is needed. I greatly fear that ecologists will lose credibility among practioners of pollution assessment because they have correctly called attention to rather vast and significant problems without following through with a professional concensus on which system level tests to carry out, measurements to make, methods to formally approve, and so on. There is even danger in calling attention to deficiencies in single species tests in predicting system level effects because it may cause some practitioners and regulators to doubt the efficacy of any biological measurements. In fact, single species tests have proven remarkably effective to estimate responses at high levels of biological organization despite considerable theoretical deficiencies in using them. Nevertheless, if the field of environmental toxicology and chemistry is to continue to evolve, these deficiencies must be identified and corrective measures taken.

Another problem is the inescapable conclusion that research (to provide system level responses of the type just described) is not sufficiently theoretical for some funding agencies and too theoretical for others. There is some evidence that this problem has been recognized and has been addressed in a minor way. Other blocks to development of adequate ecosystem level tests include difficulties in getting specialists in necessary disciplines to collaborate when their salary increases, and/or tenure and promotion may be judged by specialists with an uncharitable view toward group research.

A fundamental problem with present toxicity testing protocols is that they often estimate effects on an ecosystem as if the ecosystem were merely a collection of species exposed to a single pure compound under constant conditions. The need to go beyond single species testing to evaluate hazard to the environment posed by toxic chemicals is gaining momentum. A parallel thrust involving the study of increasingly complex systems for evaluating environmental fate of chemicals is also in progress. Although the outcome of these developments is not evident, it is abundantly clear that the need to examine both toxicity and environmental fate of chemicals in a more environmentally realistic way is now a *sine qua non*. As is the case with most developing fields, it would be unfortunate if these new approaches and new methods were used for regulation before a substantial and sound data base validating their efficacy has been produced. It would be equally foolish to retard development of such methods because they are not of immediate practical benefit. Both society and industry have much to gain from the production of more accurate means of estimating hazard of chemicals in the environment. While single species tests are far from perfect, their development has far outstripped development of determination of toxicological responses at higher levels of biological organization. Although this article addresses higher levels of biological organization than single species, it is not intended to denigrate the research at lower levels (e.g., enzyme) which might enable predictive capabilities for mechanisms of toxicity to be disclosed.

Robert MacArthur (1975) said 'Scientists are perennially aware that it is best not to trust theory until it is confirmed by evidence. It is equally true, as Eddington pointed out, that it is best not to put too much faith in facts until they have been confirmed by theory'. Since Hart *et al.* (1945) produced a method for toxicity testing that was soon endorsed by a committee of the Water Pollution Control Federation Doudoroff (1951), quite a large number of facts have been generated in the field of aquatic toxicology: water quality often may markedly mediate the expression of toxicity, some chemicals will interact synergistically or antagonistically, life history stages of a single species may not be comparably sensitive to different toxicants, and different species may alter their relationships to each other in terms of sensitivity to toxicants so that knowing response relationships to chemical A does not ensure prediction of relative sensitivity to chemical B. As a consequence, predictive value of toxicity tests has remained low, and transferability of information from one species to another and from one level of biological organization to another has not been satisfactory. Use of application factors to compensate for areas of ignorance or absence of data has not reached a stage of development where substantive scientific justification is available for the efficacy of the factors presently used. A certain degree of safety can be achieved by making the figures as large as the worse possible case demands, but sanitary engineers attempting to achieve these levels have found them technologically or economically impossible. In short, the period since 1945 might be characterized as an era where much evidence accumulated but little integrating theory surfaced. No integrating hypothesis was produced to pull these facts together or explain their relationship to each other.

Over the 30-yr period from 1945 to 1975, an enormous toxicological data base for aquatic organisms had been generated. This data base was so large that it was beyond the capability of any individual to fully comprehend all details, even when the information was subdivided into a single group of organisms such as fish. However, an event occurred that showed that this very substantial data base was inadequate in several notable aspects: (1) the amount of information on a particular chemical was probably inadequate, (2) the kinds of information generated on a particular chemical were generally inadequate for making a scientifically justifiable estimate of hazard, and (3) the transfer of information on one chemical to estimate with precision the hazard of another did not appear feasible.

Under the provisions of the June 7, 1976, consent decree, the U.S. Environmental Protection Agency (USEPA) was directed to issue Federal Water Pollution Control Act effluent limitations and guidelines, new source standards of performance, and three treatment standards for 65 identified toxic pollutants. Implementation of the directives of this decree began when the USEPA drafted Water Quality Criteria Documents for each of the individual 65 pollutants. These criterion documents reviewed all pertinent literature available on the particular chemical and attempted to determine acceptable limits not to be exceeded for the protection of aquatic life and human drinking water supplies. The Water Quality Committee of the USEPA Science Advisory Board was charged with evaluating the 65 criteria documents. The report of this committee to the Science Advisory Board stated that no documents had conclusions that were scientifically justifiable. This report was accepted by the Executive Committee of the Science Advisory Board and transmitted to the Administrator of USEPA (then Douglas Costel). It is worth emphasizing that the court issuing the consent decree did not allow sufficient time for the USEPA to generate its own data base, and, therefore, USEPA was forced to prepare the criterion documents with data already available in the literature or documents in the open literature that generally were prepared for some other purpose. This event provided unmistakable proof that data to be used for the hazard evaluation must be systematically generated for that purpose.

Another event that had a major influence on toxicity testing and hazard evaluation was the passage of the Toxic Substances Control Act (TSCA) that became law with President Ford's signature on October 11, 1976. This act represents an attempt to establish a mechanism whereby the hazard of a chemical substance to human health and the environment can be assessed before the substance is introduced into the environment. The enactment of TSCA served as a powerful new stimulant to development of testing procedures to evaluate hazard associated with potentially toxic substances to human health and the environment.

These events and others clearly indicated the need for the development of a strategy for hazard evaluation that led to the production of a series of books now commonly referred to in the profession as Pellston I, II, III, and IV. Pellston I (Cairns *et al.*, 1978) advocated linkage of the environmental concentration of a chemical (the term is meant to include such things as partitioning, transformation processes, persistance, etc.) with the concentration producing no adverse biological effects. The degree of uncertainty or lack of confidence in estimating these two concentrations was a function of their proximity to each other. This view is summarized graphically in Figure 1. Pellston II (Dickson *et al.*, 1979) examined protocols used in various industrialized countries for systematically generating the data base necessary for a scientifically justifiable hazard evaluation. Pellston III (Maki *et al.*, 1980) examined biotransformation processes and their role in estimating environmental concentration of a chemical. Pellston IV (Dickson *et al.*, 1982) dealt with modeling the fate of chemicals in the environment.

These and other publications had as a primary goal the development of an underlying strategy for estimating hazard. This strategy must be based on sound science and professional judgement and should be as cost-effective as possible. The most important consequence of these events just described has been to direct attention to the *information content* of data being generated (i.e., the facts) and *ways in which data will be used*! This will, in turn, add the additional requirement that data not only be precise, reliable, reproducible, and so on, but also *be suitable for the use of estimates proposed*! If the

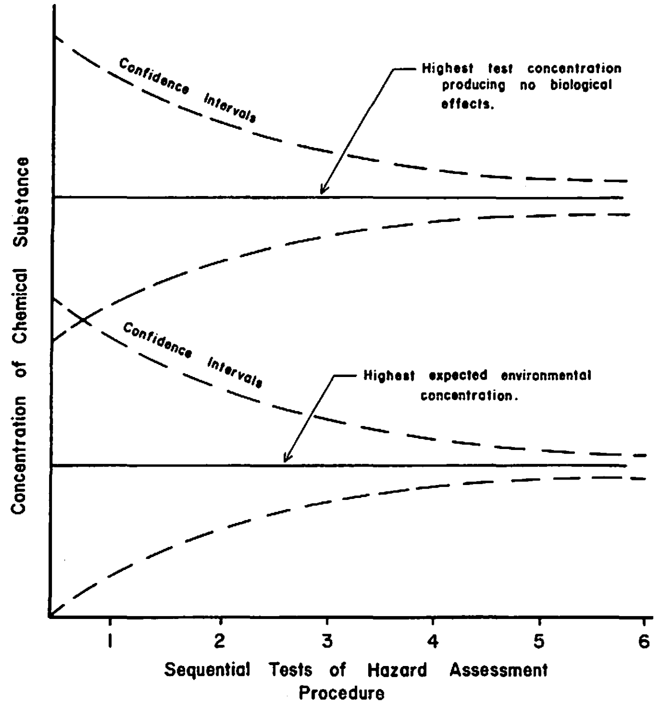

Fig. 1. Diagrammatic representation of a sequential hazard-assessment procedure demonstrating increasingly narrow confidence limits of estimates of no-biological-effect concentration and actual-expected-environmental concentration. (Reprint with permission from ASTM STP 657, *Estimating the Hazard of Chemical Substances to Aquatic Life*, Copyright ASTM, 1916 Race St., Philadelphia, Pa., 19103).

problem is viewed in this fashion, it becomes abundantly clear that the types of toxicity data now being generated are qualitatively deficient for their intended purpose! Toxicity tests should provide information that will facilitate predictions of the concentrations that will not harm living things in the environment *at all levels of biological organization*! The purpose of this manuscript is to present the view that single species tests alone are inadequate for this purpose.

2. Discussion

Some very important questions are related to testing at different levels of biological organization that deserve serious attention:

(1) Can single species tests be used to predict responses reliably at other levels of biological organization?

(2) In estimating the effects of chemicals on populations, multispecies assemblages, communities, and ecosystems, what are the limitations of laboratory science? In other words, are different degrees of environmental realism possible in the laboratory or under laboratory conditions at different levels of biological organization?

(3) What should be the balance of toxicity tests at different levels of biological organization in order to make a valid estimate of hazard?

(4) How should tests at different levels of biological organization be sequenced?

(5) What criteria should be used to validate laboratory predictions in 'real world' field situations? The complexity and uniqueness of each ecosystem has mitigated against ready transfer in general information from one site to another. Thus, many field studies are situation bound and highly site specific. Can the transferability of information from one site to another be enhanced by the development of mathematical models?

2.1. Can single species tests be used to predict responses reliably at other levels of biological organization?

One primary justification for using single species tests as a basis for estimating concentrations that will not prove harmful to communities and ecosystems is that if the most sensitive species is selected and concentration standards are set on that basis, then all other species will be protected. Since only a small percentage (probably less than 1%) of all freshwater species can be maintained in the laboratory sufficiently well to satisfy the requirement that no more than 10% of the control organisms expire during the course of tests, it seems quite likely that the most sensitive species will be selected for testing. In virtually every instance, the most sensitive species is being selected from a limited array of test species and extrapolating is being done from those results. Lest the discussion that follows be misunderstood, it is not intended to be an attack on single species toxicity testing. Such tests are essential for obtaining information on concentrations and durations of exposures to chemicals that result in changes in survival, reproduction, physiology, biochemistry, and behavior of individuals within particular species. One can question the scientific justification of using single species tests to predict changes in competition, predation, community function, ecosystem energy flow, and nutrient cycling. These are only a few of the many characteristics of ecosystems that either cannot be predicted from single species tests or for which there is insufficient evidence that the prediction is scientifically justifiable. Although practitioners of single species toxicity testing may not state that the results of these tests can be used to protect biological systems of greater complexity than single species, the implication is definitely present. The public believes that when a concentration is proported to produce no adverse biological effects, then the effects so designed go beyond the kinds of effects that are characteristic of single species responses. If the field of environmental toxicology and chemistry is to prosper, 'truth in packaging' is mandatory in terms of the limitations, as well as the strengths, of single species toxicity tests now so widely used.

One common argument advanced by people who favor continuation of primary reliance on single species testing is that no significant ecological disasters have occurred when carefully carried out single species tests were used. Of course, this could be merely due to the fact that single species tests are rarely validated by extensive, carefully carried out ecological investigations. It is not surprising that no adverse effects were noted because no extensive investigations were carried out to support this statement. In short, it is a statement based more on absence of information than on supporting information. It is quite likely that no dramatic events, such as a major fish kill, would be associated with waste discharge practices based on carefully carried out single species tests because, if the species were carefully selected and the test conducted by experienced professionals and a large application factor used, this should certainly not occur. On the other hand, changes in the ecosystem that might reduce fish population by impaired spawning rather than lethality would be less likely to be noticed by casual observers because no dramatic, highly visible evidence would be present to suggest major changes were occurring. It would be extremely helpful if predictions made with single species tests were validated by extensive field studies that would show whether or nor both ecosystem structure and function were impaired at concentrations considered to have no adverse biological effects based on single species evidence alone.

There is also another intriguing possibility – single species tests are vastly overprotective. Ecologists have made statements for years that ecosystems are fragile because of their extraordinary complexity. The intuitively reasonable argument that such highly complex systems may be put into disequilibrium by disturbing any component of the system has been quite prevalent. The reasoning is that such an interdependent, interlocking system is fragile because of these abundant linkages. This complexity and multitudinous first, second, third, etc. order interactions are so well accepted by ecologists that any statement along these lines would be regarded as platitudinous. However, very little substantive evidence exists that supports the statement that complexity is necessarily associated with fragility. Some of the most complex ecosystems known to man are periodically subjected to major natural disturbances that they are

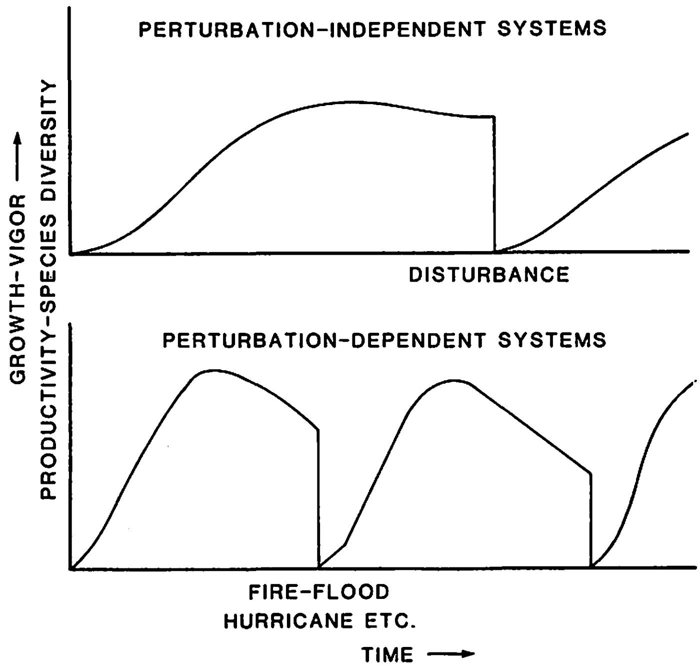

Fig. 2. Disturbances in general ecosystems create vegetational setbacks and complete recovery is slow, whereas disturbances in perturbation-dependent ecosystems usually stimulate pulses of growth which rapidly decline unless disturbed again.

either able to resist or, if displacement occurs, recover. In fact, Vogl (1979) and others have pointed out that some ecosystems actually deteriorate if striking disturbances do not occur at periodic intervals. The difference between disturbance-dependent and disturbance-independent ecosystems is given in Figure 2. An alternative hypothesis equally tenable is that ecosystems are tough because they are complex and that damage to one of several similar pathways may result in shunting to alternative pathways of nearly comparable function. In this case, complexity would increase rather than decrease resistance to disturbance. Functional redundancy in ecosystems has been recognized for years. There may by several predators on a single prey species. The river continuum hypothesis (Vannote *et al.*, 1980) indicates that certain processes, such as leaf degradation, may be carried out by different taxonomic groups in the upper and lower reaches of a stream. The end functional result, namely increased availability of nutrients and energy in the leaf, is unchanged.

In addition, the low environmental realism of the very simple, common toxicity test (where organisms are tested in a container with water but with no mud, rocks, vegetation, etc.) means that transformation of hazardous chemicals might occur less rapidly than in the 'real world'. Rapid transformation in the latter might produce secondary products less harmful than the original and result in decreased ecosystem vulnerability. Similarly, various types of environmental sinks for chemicals are not incorporated into most commonly used single species tests.

A final argument given by those who accept the need for going beyond single species testing will be that these are sufficient in instances when the estimated environmental concentration of the chemical is so far below the estimated no adverse effects concentration that it would be ridiculous to go beyond simple and inexpensive single species screening tests. Kimmerle (1979) has noted, however, that the actual environmental concentration might be far higher than was estimated from simple laboratory screening tests and that the no adverse biological effects concentration might be far lower in the 'real world' than was estimated from simple screening laboratory tests (Figures 3–4). In short, the screening tests did not accurately predict 'real world' events! In one case, the concentration was vastly increased and in the other (the environmental concentration) vastly decreased (Figure 3). The end result in two concentrations that appeared comfortably separated in the laboratory were in the 'real world' quite close together. Of course, the errors could be in the opposite direction in both cases and end up with two concentrations that appeared quite close together from laboratory evidence being quite distant from each other in the real world (Figure 4). These possibilities provide support for doing toxicity testing at more than one level of biological organization even for screening purposes.

From an economic standpoint, the soundest possible evidence on which to base management and regulatory decisions must be demanded. At the present time, sufficient evidence is not available to determine how accurately predictions can be done of toxicological response from one level of biological organization to another, but both theoretical biology and the rapidly accumulating data base on this subject seem to indicate that such predictions are relatively weak.

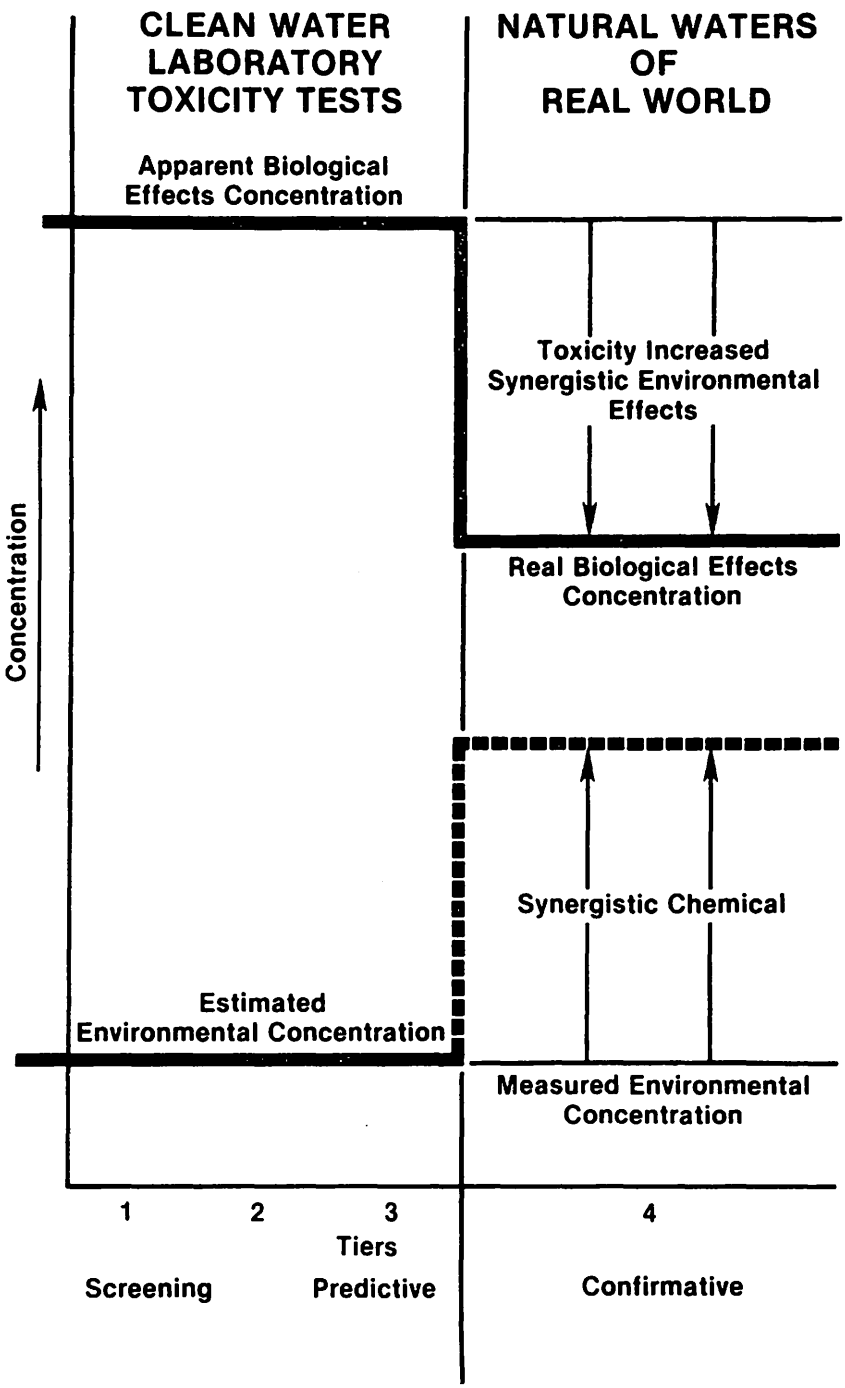

Fig. 3. Hypothetical situation of an apparent small margin of safety from clean water laboratory toxicity data actually being much greater because of mitigating effects of natural waters. (From Kimerle, 1979, in *Workshop on Hazard Assessment.*)

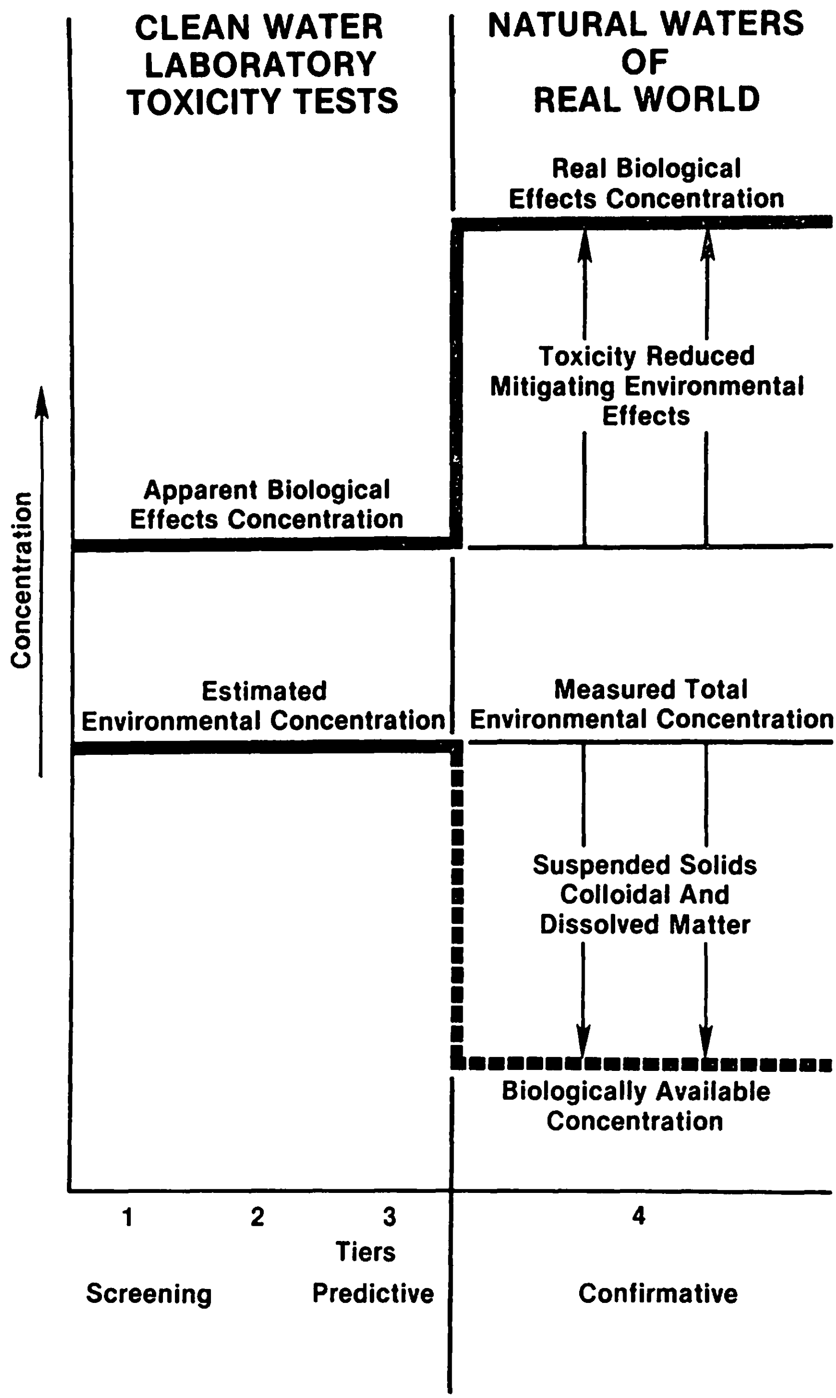

Fig. 4. Hypothetical situation of an apparent large margin of safety from clean water laboratory toxicity data actually being much smaller because of synergistic effects of natural waters. (From Kimerle, 1979, in *Workshop on Hazard Assessment*.)

2.2. In estimating the effects of chemicals on populations, multispecies assemblages, communities, and ecosystems, what are the limitations of laboratory science?

Are there different degrees of environmental realism possible under laboratory conditions at different levels of biological organization? In a visiting scholar address given at the Mountain Lake Biological Station, Virginia, Odum (1981) gave an address entitled 'The Limitations of Laboratory Science' that beautifully illustrates some areas in which laboratory investigations, however skillfully carried out, will not suffice. It seems intuitively reasonable that environmental realism is more easily achieved in the laboratory at lower levels of biological organization (e.g., species) than at higher levels or organization (e.g., community or ecosystem). The report of the National Research Council Committee on Ecotoxicology (Cairns *et al.*, 1981) takes note of the fact that situations occur where laboratory evidence will not be adequate and field testing will be mandatory. The report even makes a distinction between field situations in which the exposure is contained and those in which it is not. This merely recognizes that both effects of scale and time as well as degree of complexity must be considered that are not amenable to laboratory study. If this hypothesis is correct, then predictions of toxicological effects from one level of biological organization to another are not scientifically justifiable. In this event, an array of toxicity tests at different levels of biological organization are necessary for scientifically justifiable estimates of hazard. If the hypothesis is accepted that the limitations of laboratory science become greater as the complexity or level of biological organization increases, then both microcosms and field tests will become more common than they are now. Of course, both these hypotheses need careful and detailed testing so that the decision that the judgment of their soundness can be based on solid evidence. In addition, evidence must be obtained on the limitations of laboratory science in making predictions at different levels of biological organization. In other words, to what degree can more complex biological systems be simulated in the laboratory?

2.3. What should be the balance of toxicity tests at different levels of biological organization in order to make a valid estimate of hazard?

A rigid or specific balance of tests at each level of biological organization would not serve equally well for all situations and all categories of chemical substances. Therefore, protocols need to incorporate procedures for adjusting the balance of tests at different levels of biological organization as information on the level of complexity most likely to be affected is determined. Presumably, the most even distribution would be at the outset and become less and less even as critical sensitive components are identified.

The determination of balance, particularly as one proceeds through a sequential protocol at different levels of organization (i.e., a sequence for each of the major levels), will pose some interesting problems. For example, suppose that a particular single species test shows that deleterious effects would occur with that species but that major ecosystem functions would continue undisturbed. An additional condition, not explicity stated but implied in the previous statement, could be made that alternative species were available to carry out similar ecological functions if the species suffering the deleterious effects represented a significant portion of the biomass. This would still pose a problem requiring considerable professional judgment and analysis because the loss of functional redundancy (i.e., reducing the number of species carrying out a particular function) is a deleterious ecosystem effect. An even more interesting decision would be forced by an effect on a transitory species that would soon be lost anyway because of normal successional processes with no other effects being discernible. A number of other interesting arrays of mixed test results could be furnished that would illustrate the point that by adding more levels of biological organization to the test system that the increased complexity of the situation requires much more professional judgment than has ever been necessary. However, this merely emphasizes the point that judgments were probably being made on evidence that was far too simplistic.

2.4. How should tests at different levels of biological organization be sequenced?

Sequencing in a toxicity testing protocol can serve various purposes: (1) adding entirely different information from that already gathered, (2) expanding on information shown to be critical by earlier tests, (3) validating evidence gathered in previous tests. In many of the existing toxicity testing protocols, multispecies and system level tests are carried out only when the probability appears great that some deleterious effects might occur. In cases where the no adverse biological effects concentration (based on single species tests) was very markedly higher than the estimated environmental concentration of the chemical, carrying out additional tests at higher levels of biological organization was usually considered unnecessary.

If several levels of biological organization are tested at the outset of a toxicity testing protocol, several alternative courses of action are possible: (1) if some or all the tests at various levels in the first part of the sequence show a probability of adverse biological effects or there is uncertainty about this probability, additional tests would be carried out at all levels of biological organization in the early part of the sequence unless compelling evidence exists for omission of one or more levels, (2) the most critical level of biological organization could be selected for these tests and additional tests could be conducted only in that sequence designed for that particular level (i.e., a sequence would be designed for each level of biological organization), (3) the levels of biological organization likely to give the least useful information could be eliminated but several levels each with its own sequence could be retained as a means of validating the presumed relationship identified in earlier parts of the sequence.

Since so few tests are now routinely utilized for hazard evaluation at levels of organization higher than the species, providing a detailed scenario on how they might be used is difficult. Some factors that would influence sequencing in the alternative system proposed would be the amount of information redundancy, the predictive capability within a particular level of organization from one function to another, and so on. Since so little information exists now on these factors, speculation on details of sequencing is difficult.

2.5. What criteria should be used to validate laboratory predictions in 'real world' field situations?

If predictions from one level of biological organization to another are unsound, validations of predictions made using one level with a higher or lower level of biological organization would not work well. On the other hand, if a prediction is being validated, then it does not matter what level or biological organization was used in the test but only what level of biological organization is being protected by the prediction. Thus, in the first case the accuracy of a test in terms of the degree of environmental realism incorporated into the laboratory test is being validated. In the second case, the accuracy of the prediction based on the laboratory test in being validated. The prediction may be that ecosystem integrity will not be impaired at or below a certain concentration of a chemical. At the very least, putting this procedure into place would introduce a note of caution into the predictions being made, particularly where the protection of ecosystems is concerned. Furthermore, the basic assumptions underlying most present practices would be more rigorously examined. Finally, ecologists would be forced to play a more active role as problem solvers and to endorse professionally those methods now available for making realistic ecosystem measurements and predictions. If none are immediately suitable for formal endorsement by professional ecologists, strenuous efforts would be made to determine why this situation exists.

3. Concluding Remarks

When I entered the field of pollution assessment in 1948, the burning issue was whether biological testing had any role to play in pollution assessment. Students in my classes find it difficult to believe that anyone would doubt the value of biological evidence, but an examination of the literature of the period from 1945 to 1950 will show this to be true (Patrick, 1949). This was the period when the newly published simple batch toxicity testing method (Hart *et al.*, 1945) was being acknowledged by a committee of the organization now called the Water Pollution Control Federation (Doudoroff *et al.*, 1951) and was eventually incorporated as an American Society for Testing and Materials standard method. Had Hart *et al.* (1945) or the Doudoroff committee (Doudoroff *et al.*, 1951) called for the various toxicity tests now commonly used with continuous flow requirements, embryo larvae tests, generational tests, tests at different trophic or functional levels, etc., they would have been regarded as hopelessly visionary. This is merely a consequence of the explosive and rapid development of a field that had only a few practioners in the late 1940s. However, the last 10 yrs have shown remarkable changes in both attitudes and methodology. Almost all the advances have occured in single species toxicity testing. Testing at higher levels of biological organization has not kept pace with advances in single species testing, and an uncharitable person might say that a practitioner using ecological methods of the late 1940s and early 1950s could still get by today.

There is abundant evidence, however, that a period of explosive development is already beginning in the use of laboratory microcosms, as well as in the use of artificial streams and larger stimulation units which Odum has called *mesocosms*. Papers are beginning to appear in the professional literature validating laboratory tests in natural systems with a frequency that is in notable contrast to the virtual absence of such publications only a few years ago.

I recognize the considerable temerity of calling attention to the need for going beyond single species testing when such tests are just now being commonly used and no system level tests have been formally endorsed as standard methods. However, precise predictions of ecosystem effects will not be possible until methodologies and capabilities not now available have been developed. It has been said that looking into the future is equivalent to peering through a brick wall. Nevertheless, it seems intuitively reasonable that the following will be landmarks in the development of toxicity tests at higher levels of biological organization than single species:

(1) The first professionally endorsed ecosystem level method appearing as a standard method in one of the presently recognized systems for doing so.

(2) The first protocol in which system structure and function are given equal attention.

(3) The first protocol in which tests at higher levels of biological organization than single species play a major role in generating the initial information on which subsequent testing is based.

(4) The first formally endorsed field method for validating laboratory tests of any kind.

The November 1981 issue of *Science* announces as a matter of general interest the development of a sealed microcosm which has stable characteristics and species composition for over a year. Granted that this is a rather simple system, it nevertheless displays characteristics long sought by those who wish to carry out chronic microcosm tests under controlled conditions. Presumably now that this initial breakthrough has led the way, additional methods will quickly appear as is often the case when a major new field begins to open.

Acknowledgments

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