United States
Environmental Protection
Agency

Environmental Research
Laboratory
Athens GA 30613

Research and Development

EPA-600/S3-82-093 Feb. 1983

EPA

Project Summary

Community Structure, Nutrient Dynamics, and the Degradation of Diethyl Phthalate in Aquatic Laboratory Microcosms

T. L. Hall, J. A. Hamala, P. F. Hendrix, H. P. Kollig, J. A. Krewer, L. C. Langner, and W. R. Payne, Jr.

An investigation was conducted of the environmental fate of diethyl phthalate (DEP) in the continuous-flow channel microcosms housed in the U.S. Environmental Protection Agency's (EPA) Environmental Research Laboratory, Athens, GA. The microcosms were designed to enable testing of the Exposure Analysis Modeling System (EXAMS), a theoretical-type predictive model for the determination of the fate of toxic compounds in freshwater systems. The objectives of the investigations were to determine (1) whether a definable stable state could be achieved in which to test the model, (2) the effects of different nutrient treatments on ecosystem structure and function and on the fate of DEP, and (3) the degree of similarity between replicate microcosms.

Aufwuchs assemblages in the microcosms reached fairly stable levels of biomass, metabolic activity, and similar species composition within two or three months after inoculation. Communities receiving direct nutrient inputs appeared to stabilize first, followed by downstream communities.

A highly significant relationship between phosphorus inputs and aufwuchs chlorophyll *a* was established, suggesting that the relatively stable input concentrations of inorganic nutrients into any given compartment were among the primary factors controlling maximum development of aufwuchs.

Replicate microcosms were statistically indistinguishable with respect to nutrient concentrations for most of the experimental period. Compartments receiving direct inputs of inorganic nutrients had the most consistent replicability. Although non-taxonomic community structure was generally similar in replicate compartments, some differences were observed in relative species abundance.

Sorption, volatilization, and photolysis were insignificant processes in the fate of DEP. Alkaline hydrolysis at pH 10 showed only a slight effect. Microbial degradation was the dominant process. First-order degradation rates were all within an order of magnitude, even though there were significant differences in both chemical environments and biological communities.

This Project Summary was developed by EPA's Environmental Research Laboratory, Athens, GA, to announce key findings of the research project that is fully documented in a separate report of the same title (see Project Report ordering information at back).

Introduction

The fate of toxic compounds in aquatic ecosystems is influenced by a number of biotic and abiotic factors. Experimental analysis of such factors in laboratory ecosystems provides one means of deriving information for use in predictive

models of environmental fate of toxic compounds. For this purpose, it is important that experimental systems meet the assumptions of the model and the requirements of the parameterization procedure. This report presents the results of an experimental study of the fate of the plasticizer, diethyl phthalate (DEP), in a set of continuous-flow microcosms subjected to a range of nutrient enrichment levels. Primary emphasis is on the degree to which the experimental systems fulfilled the parameterization requirements of the Exposure Analysis Modeling System (EXAMS). Those requirements are that (1) physical, chemical, and biological processes that influence the environmental fate of a compound be at steady state during the time interval under study; (2) manipulation of chemical inputs into the microcosms creates a set of different environments in which to study the fate of the compound; and (3) identically manipulated microcosms behave as statistically indistinguishable replicates. These requirements are addressed in terms of several physical, chemical, and biological variables that were measured routinely throughout the experiment.

Materials and Methods

The experimental system consisted of a 19.5-m-long, 46-cm-wide, 51-cm-deep "U-shaped" Plexiglas flume lined with Teflon film. The flume was divided into two independent channels, each subdivided into eight 250-liter compartments equipped with outlet weirs such that the effluent from an upstream compartment constituted the influent for the next one downstream. Uniform mixing in each compartment was accomplished by the use of Teflon-covered Plexiglas paddlewheels suspended longitudinally in each compartment and adjusted to a rotation speed of 2.0 rpm. The system was based in an environmental chamber that provided control of temperature (21°C), water flow (500 l/day), relative humidity (50%), and light (2000 fc of fluorescent light on a 12-h light/dark cycle).

The first two compartments in each channel were not treated with nutrients or biotic inoculum to allow for the study of alkaline hydrolysis, uv photolysis, sorption and volatilization under abiotic conditions. The remaining six downstream compartments in each channel were inoculated from local ponds and streams, and nutrient chemicals were continuously added to selected compartments to create gradients of nutrient enrichment.

A concentrated solution DEP was added through an all glass, constant-head system at 2.0 ml/min to achieve a final concentration of approximately 194 μg/l when diluted with the incoming water. The DEP input was moved consecutively from one replicate pair of compartments to the next on a weekly basis to allow for the study of DEP degradation under conditions existing in each individual pair.

Water samples were collected for analysis weekly from each compartment. Dissolved nutrients (nitrate, nitrite, ammonia, Kjeldahl nitrogen, total phosphorus, and orthophosphorus) were analyzed on glass fiber filtered aliquots in a Technicon Autoanalyzer. Total and dissolved organic carbon were analyzed in a Beckman TOC analyzer. DEP was extracted in iso-octane and analyzed by electron capture in a gas-liquid chromatograph. Dissolved oxygen and pH were measured with electronic meters *in situ*; these values were used to estimate community metabolic activity.

Biological analyses (algal and bacterial enumeration, total ATP concentration, chlorophyll *a* content, ash-free dry weight, DEP concentration, and total organic carbon) were performed on samples collected weekly from artificial substrates attached to the sides of the compartments. Chlorophyll *a*, ATP, and bacterial numbers were also measured in water column samples.

Results and Conclusions

1. Aufwuchs communities appeared to reach a definable steady state within two to three months of inoculation, based on available data for taxonomic and non-taxonomic community structure (relative abundance of algal species, similarity indices, chlorophyll *a*, ash-free dry weight, ATP, and total organic carbon) and community metabolic activity (relative changes in dissolved oxygen and pH). Also, based on metabolic activity estimates, communities receiving direct nutrient inputs appeared to stabilize first, followed by downstream communities.
2. A significant linear regression ($r = 0.88$) between phosphorus loading and aufwuchs chlorophyll *a* suggested that phosphorus inputs were important in controlling non-taxonomic structure. A significant curvilinear relationship ($r = 0.91$) between the same two parameters indicated that spatial limitations may have become important in controlling aufwuchs development at higher phosphorus loading rates.

   Non-taxonomic parameters associated with living aufwuchs (chlorophyll *a*, ATP, and bacterial numbers) provided greater resolution in distinguishing structurally distinct communities than did those that also included detritus (ash-free dry weight and total organic carbon). As expected, microcosms receiving nutrient enrichment displayed significant increases in non-taxonomic structure for all parameters examined.
3. Replicate microcosms were generally similar with respect to both chemical and biological variables. Nutrient concentrations (PO_4, NO_3, NH_3) were statistically indistinguishable for most of the experimental period; the differences that did occur could not be attributed to experimental manipulations. Non-taxonomic structure also was similar between replicate microcosms. High light intensity, temperature, and nutrient loading combined with low carbonate alkalinity appeared to select for euryecious organisms typical of eutrophic systems. Algal species composition was generally more similar than dissimilar (based on SIMI indices) among the different microcosms. The differences that were observed usually resulted from large variations in relative abundances of one or two algal species. Shannon-Wiener species diversity and evenness estimates indicated relatively complex algal aufwuchs communities.
4. Sorption, volatilization, photolysis, and chemical hydrolysis were insignificant processes in the fate of DEP. Microbial degradation of DEP resulted in the disappearance of 36 to 90 percent of the compound. Although aufwuchs bacterial numbers varied by several orders of magnitude among the microcosms, first-order degradation rates were similar in all except those receiving low inorganic and high organic nutrient inputs. Therefore, only a fraction of the total bacteria present may have participated in DEP degradation.

T. L. Hall, J. A. Hamala, P. F. Hendrix, J. A. Krewer, and L. C. Langner are with The Bionetics Corporation; the EPA authors ***H. P. Kollig*** *(also the EPA Project Officer, see below) and* ***W. R. Payne, Jr.****, are with the Environmental Research Laboratory, Athens, GA 30613.*

The complete report, entitled "Community Structure, Nutrient Dynamics, and the Degradation of Diethyl Phthalate in Aquatic Laboratory Microcosms," (Order No. PB 83-136 341; Cost: $14.50, subject to change) will be available only from:

National Technical Information Service
5285 Port Royal Road
Springfield, VA 22161
Telephone: 703-487-4650

The EPA Project Officer can be contacted at:

Environmental Research Laboratory
U.S. Environmental Protection Agency
College Station Road
Athens, GA 30613