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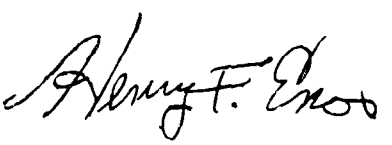

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Preparation Date:

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Abel, Daniel C., Christopher C. Koenig, and William P. Davis. 1987. Emersion in the Mangrove Forest Fish Rivulus marmoratus: A Unique Response to Hydrogen Sulfide. Environ. Biol. Fishes. 18(1):67-72. (ERL,GB 554).

The mangrove forest fish Rivulus marmoratus (Cyprinodontidae) has frequently been observed out of water, a phenomenon generally attributed to habitat drying. We tested the hypothesis that hydrogen sulfide, a substance characteristically found in their environment, can serve as a stimulus for emersion. In the field we found R. marmoratus in water with low to moderate levels (less than 250 ppb) of H2S. In the laboratory, R. marmoratus leaped from water contaminated with H2S at ecologically relevant concentrations (median response at 123 ppb). Aquatic hypoxia did not induce emersion, but prey capture did. Oxygen consumption by both juveniles and adults decreased significantly in air (27 and 25%, respectively). Our results suggest that avoidance of H2S and the ability to survive terrestrial conditions enable this species to permanently occupy an area of the forest unavailable to other fishes. Furthermore, because a variety of stimuli lead to emersion in R. marmoratus, terrestriality in this species is likely a generalized response to environmental stress as well as a means of exploiting terrestrial resources.

Ahearn, D.G., and S.A. Crow. In review. Fungi and Hydrocarbons in the Marine Environment. In: Proceedings of the 4th International Marine Mycology Symposium. S.T. Moss, editor, Cambridge University Press, London. (ERL,GB X507*).
Avail. from NTIS, Springfield, VA: PB86-109964

Hydrocarbons from various sources--anthropogenic pollution, marine seeps, marine algae, atmospheric fallout and terrestrial runoff--enter the ocean daily. These complex hydrocarbon mixtures are dispersed and degraded by abiotic and biogenic processes. The rate of degradation and the significance of microbial activities in the fate of oceanic hydrocarbons vary with environmental conditions and the type of hydrocarbon. Most commonly, bacteria are considered the primary degraders, with algae and fungi having minor roles. Although implied in a number of cases, the degradation of complex hydrocarbon mixtures by a successional microflora containing temporally isolated populations of bacteria and fungi, has been inadequately studied.

Alexander, Martin. In press. Anomalous Effects of Concentration on Biodegradation of Organic Chemicals. Appl. Environ. Microbiol. 19p. (ERL,GB X481*).

The purpose of this review is to show that erroneous conclusions may be reached from studies or routine tests done with organic chemicals at the levels often employed for predicting chemical fate in nature. These errors in extrapolation from high to low concentration may occur in routine evaluations of biodegradation, careful assessments of kinetics or the establishment of products formed in waters, soils or sediments.

Barkay, Tamar. In review. Adaptation of Aquatic Microbial Communities to Hg^{2+} Stress. Appl. Environ. Microbiol. 23p. (ERL,GB 608*).

The mechanism of adaptation to Hg^{2+} in four aquatic habitats was studied by correlating microbially mediated Hg^{2+} volatilization with the adaptive state of the exposed communities. Structural and functional parameters indicated that adaptation of all four communities was stimulated by exposure to Hg^{2+}. In saline water communities, adaptation was associated with rapid volatilization after an initial lag period. This mechanism, however, did not promote adaptation in as freshwater environment, where Hg^{2+} volatilized slowly, regardless of the adaptive state of the microbial community. Distribution of the mer operon among represenetative colonies of the communities was not related to adaptation to Hg^{2+}. Thus, although volatilization is a mechanism which enables some microbial communities to sustain their functions in Hg^{2+} stressed environments, it is not coded for by the gene system that mediates this mechanism in pure cultures.

Barkay, T., D. Chatterjee, S. Cuskey, R. Walter, F. Genthner, and A. Bourquin. In review. Bacteria and the Environment. In: Revolution in Biotechnology. International Council of Scientific Unions. 22p. (ERL,GB 604*).

Microorganisms with new functions can be constructed in the laboratory by gene cloning. This paper discusses the potential of a powerful tool for environmental management: new strains to control pests, to increase yields, and to degrade noxious pollutants. Approaches and methods are described for risk assessment based on the experiences and findings in microbial ecology. However, risk assessment criteria have yet to be established due to the unknown and potentially harmful effects of the introduced organisms on the receiving environments.

Barkay, Tamar, and Gary Sayler. In review. Gene Probes as a Tool for the Detection of Specific Genomes in the Environment. Presented at the 10th ASTM Symposium on Aquatic Toxicology and Hazard Assessment, May 4-6, 1986, New Orleans, LA. 16p. (ERL,GB 578*).
Avail. from NTIS, Springfield, VA: PB87-102505.

Gene probes hold a great promise as a tool in environmental sciences. They may be used to detect specific genotypes, to follow gene flow process, to delineate complex taxonomic aggregates and to monitor genetically engineered organisms in the environment. The sensitivity of the method is currently limited by experimental procedures and its specificity depends on the nature of the DNA sequences used as probes and the efficacy of lysing methods. Variable genetic determinants which code for the same trait determine the universality of gene probes. Finally, the method is highly feasible in terms of cost, speed and expertise. Current and future developments in molecular microbial ecology are likely to contribute toward the improvement of the probing methodology for the full realization of its potential in environmental sciences.

Borthwick, Patrick W. 1986. Effects of Salinity Change on Acute Lethal Responses of Bay Mysids (Mysidopsis bahia) to Three Insecticides. EPA/600/X-86/272*, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 7p.

In this study we investigated the influence of salinity change on the acute toxicities of three insecticides to the bay mysid (Mysidopsis bahia). Salinity is a principal environmental variable in estuarine waters. Bay mysids subjected to a 50-minute salinity change and high (32 o/oo) and low (10 o/oo) salinity seawater were approximately three times more sensitive to fenvalerate (synthetic pyrethroid insecticide) compared to mysids exposed at the reference (culture) salinity (21 o/oo). At low salinity, mysids exposed to endosulfan (chlorinated insecticide) were almost 4 times more sensitive; high salinity caused a slight sensitivity increase. Fenthion (organophosphate insecticide) toxicity, least affected by salinity, was unchanged at high salinity, and only slightly more toxic at low salinity. Evidence from this study indicated that salinity change may reduce an organism's tolerance to toxic insecticides.

Bourquin, Al W. 1986. Biotechnology Aquatic Risk Assessment Research. EPA/600/X-86/235*, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 346p.

This internal status report and attached publications summarizes the initial research efforts in the Aquatic Biotechnology Risk Assessment Research Program at ERL/Gulf Breeze. The research was undertaken in response to an OPTS need to more fully understand the ecological impact of the deliberate release of genetically engineered microorganisms into the aquatic environment. The research covered in this report emphasizes the most recent developments in detection and enumeration technology for specific microorganisms and specific DNA segments. The report summarizes the development of immunoepifluorescence microscopy techniques for detecting viable but nonculturable microbes and the use of DNA probes for detecting specific segments of genetic materials in environments and the development of a laboratory method to determine genetic exchange in environmental systems. The report summarizes and contains 17 peer-reviewed publications and reports produced by the ERL/Gulf Breeze program during the period from June 1985 to September 1986.

Brayton, P.R., and R.R. Colwell. In review. Fluorescent Antibody Staining Method for Enumeration of Viable Environmental Vibrio cholerae. J. Microbiol. Methods. 12p. (ERL,GB X521*).

A membrane filtration method has been developed which is useful for enumeration of viable Vibrio cholerae 01 in environmental water samples by immunofluorescent staining. The samples are incubated with yeast extract and nalidixic acid. Substrate responsive cells, i.e. viable cells, elongate and after staining with specific antiserum and fluorescein conjugate, viable V. cholerae cells appear as long, peripheral fluorescent green banded bacilli when viewed under the microscope. Using an ocular reticule, the number of viable cells per ml can be calculated. The procedure has been adapted for use with other bacterial species if specific antisera is employed.

Brusca, J., M. Summers, J. Couch, and L. Courtney. 1986. Autographa californica Nuclear Polyhedrosis Virus Efficiently Enters but Does Not Replicate in Polikilo Thermic Vertebrate Cells. Intervirology. 26:207-222. (ERL,GB 558*).

The host range of the insect virus Autographa californica nuclear polyhedrosis virus (AcMNPV) was examined. AcMNPV could not initiate a productive infection in frog, turtle, trout, or moth cell lines. After exposure to AcMNPV, neither viral DNA nor RNA synthesis could be detected in these cell lines when assayed by dot-blot hybridizations. Entry of viral DNA to the nucleus, however, was as efficient in the nonpermissive cell lines as it was in a permissive insect cell line. Electron microscopy revealed numerous AcMNPV nucleocapsids in the cytoplasm of the nonpermissive cell lines is therefore at a stage subsequent to viral entry to the nucleus.

Capuzzo, Judith M. In review. Development of Physiological Indices to Predict the Effects of Chronic Pesticide Exposure on Zooplankton Populations. Aquat. Toxicol. 41p. (ERL,GB X511*).

The effects of the pyrethroid pesticide fenvalerate and the organophosphate pesticide fenthion on planktonic crustaceans were investigated in continuous-flow bioassays. Fenvalerate was more toxic than fenthion in acute bioassays with 96-h LC50 values ranging from 5.4 ng/l for Stage I larvae of Homarus americanus to 46.0 ng/l for adult Heteromysis formosa; LC50 values for adult Acartia tonsa were 14.7 ng/l, fenvalerate and 102.5 ug/l, fenthion. Metabolic changes paralleled delays in development for larvae of H. americanus and reductions in egg production and larval viability of A. tonsa. The most sensitive indicators of acute toxic response of Acartia to both pesticides were gross and net growth efficiencies (K1 and K2) and instantaneous birth rates, parameters which integrate metabolic responses, survival, and reproduction. Reductions in both bioenergetic parameters and birth rates were evident with sublethal exposure to both contaminants. Disruptions in reproduction and development were also observed in chronic assays of fenvalerate on Acartia tonsa. Chronic exposure to 0.6 ng/l also resulted in reproductive and developmental impairment, although some second generation copepods developed to maturity.

Chatterjee, Deb K., and A.W. Bourquin. 1987. Metabolism of Aromatic Compounds by Caulobacter crescentus. J. Bacteriol. 169(5):1993-1996. (ERL,GB 591).

Cultures of Caulobacter crescentus were found to grow on a variety of aromatic compounds. Degradation of benzoate, p-hydroxybenzoate and phenol was found to occur via B-ketoadipate. Induction of the degradative enzymes such as benzoate, 1,2-dioxygenase, the ring cleavage enzyme, catechol 1,2-dioxygenase, and cis, cis-muconate lactonizing enzyme appeared similar to the control mechanism present in Pseudomonas. Both benzoate 1,2-dioxygenase and catechol 1,2-dioxygenase seem to have stringent specificities as revealed by their action towards substituted benzoates and substituted catechols respectively. The potential degradative abilities of Caulobacters is discussed.

Clark, James R., James M. Patrick, Jr., James C. Moore, and Jerrold Forester. 1986. Accumulation of Sediment-Bound PCBs by Fiddler Crabs. Bull. Environ. Contam. Toxicol. 36:571-578. (ERL,GB 533).

Polychlorinated biphenyls (PCBs) have been, and continue to be, an ecological problem because of their environmental persistence. In aquatic systems, PCBs sorb to organic matter, accumulate in sediments, and contaminate food chains. Because of the potential for causing reproductive impairment, PCBs in aquatic food chains pose a threat to human and other predators that consume fish and shellfish. Fiddler crabs accumulate PCBs from contaminated sediments and detritus and can transfer them to aquatic, avian, and terrestrial food webs when preyed upon by fishes, birds, and small mammals. The primary objective of our research was to characterize rates of PCB uptake and depuration by fiddler crabs in a simulated spoil bank habitat that contained PCBs in weathered sediment. Also, we examined whether the concentration of PCBs in substrates affected bioaccumulation by mixing PCB-laden sediments with clean sand. In a pilot study, we tested Uca pugilator, an inhabitant of relatively dry and sandy areas, and U. minax, which inhabits wetter and muddier substrates, to determine if species differ in PCB uptake and depuration rates.

Clark, James R., Patrick W. Borthwick, Larry R. Goodman, James M. Patrick, Jr., Emile M. Lores, and James C. Moore. 1987. Comparison of Laboratory Toxicity Test Results with Responses of Caged Estuarine Animals Exposed to Fenthion in the Field. Environ. Toxicol. Chem. 6:151-160. (ERL,GB 545).

Acute, lethal effects of fenthion (an organophosphate insecticide) on mysids (Mysidopsis bahia), grass shrimp (Palaemonetes pugio), pink shrimp (Penaeus duorarum), and sheepshead minnows (Cyprinodon variegatus) were determined in laboratory tests and after field applications. Exposure at four field sites ranged from short-term exposures (equal to or less than 12 h) of rapidly decreasing fenthion concentrations to extended intervals (greater than 72 h) with slowly increasing or decreasing fenthion concentrations. Laboratory-derived LC50s provided a reliable benchmark for predicting acute, lethal effects of fenthion on caged animals in the field when exposures persisted for 24 h or more but overestimated the toxicity for exposures less than 24 h. Laboratory pulse-exposure tests with rapidly changing concentrations for 12 h were predictive of nonlethal and lethal effects observed for short-term field exposures.

Clark, J.R., P.W. Borthwick, L.R. Goodman, J.M. Patrick, Jr., E.M. Lores, and J.C. Moore. In review. Effects of Aerial Thermal Fog Applications of Fenthion on Caged Pink Shrimp, Mysids, and Sheepshead Minnows. J. Am. Mosq. Control Assoc. 14p. (ERL,GB 602*).

Mosquito control applications of fenthion by aerial thermal fog equipment were studied at two sites in Collier County, FL, for sprays that occurred on 20 and 23 June 1984. Acute, lethal effects of fenthion deposited in these estuarine habitats were assessed for caged pink shrimp (Penaeus duorarum), mysids (Mysidopsis bahia), and sheepshead minnows (Cyprinodon variegatus). At Site 1, along a bay with substantial dilution and tidal mixing, fenthion concentrations of 1.5 ug/l and 0.29 ug/l were recorded immediately after both sprays. Concentrations decreased to less than or equal to 0.020 ug/l 12 h post-spray and no mortality was observed for caged pink shrimp and mysids. Site 2 was along a residential canal system that offered limited dilution and mixing. Measurable concentrations (greater than 0.038 ug/l) of fenthion persisted at this site for 4 days. Fenthion concentrations in surface waters were toxic to caged pink shrimp and mysids after both sprays; maximum concentrations were 2.6 ug/l and 0.51 ug/l. Caged sheepshead minnows were not affected by the sprays at either site.

Clark, J.R., L.R. Goodman, P.W. Borthwick, J.M. Patrick, Jr., J.C. Moore, and E.M. Lores. 1986. Field and Laboratory Toxicity Tests with Shrimp, Mysids, and Sheepshead Minnows Exposed to Fenthion. In: Aquatic Toxicology and Environmental Fate: Ninth Volume, ASTM STP 921. T.M. Poston and R. Purdy, editors, American Society for Testing Materials, Philadelphia, PA. pp. 161-176. (ERL,GB 539).

We conducted a series of laboratory pulse-exposure experiments to model short-term field exposures of two representative estuarine crustaceans, Penaeus duorarum and Mysidopsis bahia, to the organophosphate insecticide fenthion. These tests established acutely lethal and nonlethal concentrations during pulse exposures. The data are necessary for interpretation of responses of test animals in the field when fenthion concentrations changed rapidly with time. The toxicity of fenthion to caged pink shrimp, mysids, and sheepshead minnows (Cyprinodon variegatus) was determined in the field following two aerial applications separated by 72 h, to control adult saltmarsh mosquitoes. At one estuarine site, initial concentrations of fenthion in water were 1.5 ug/L following Spray 1 and 0.29 ug/L after Spray 2. Within 12 to 24 h, however, fenthion was not detectable (less than 0.01 ug/L) because of rapid tidal flushing and high dilution at this site. Although initial exposures approached or exceeded laboratory 24-h LC50s for pink shrimp (0.40 ug/L) and mysids (0.42 ug/L), no mortality occurred among caged animals. At a second site along a residential saltwater canal with limited tidal flushing and dilution, initial concentrations of fenthion were 2.6 ug/L (Spray 1) and 0.5 ug/L (Spray 2). Within 12 to 24 h post-spray, fenthion decreased to 0.4 ug/L (Spray 1) and 0.14 ug/L (Spray 2) and continued to diminish during the next 48 to 72 h. These concentrations approximated the 48- and 72-h LC50s for pink shrimp (0.22 ug/L and 0.15 ug/L) and mysids (0.37 ug/L and 0.18 ug/L). All exposure concentrations were three orders of magnitude below the 24-h LC50 for sheepshead minnows (1900 ug/L) and no mortality occurred among caged fish. By deploying caged pink shrimp and mysids daily, before and after each spray, in situ exposure regimes varied for each group and resulted in responses among caged test populations that ranged from no observed effect to 100% mortality. The responses of caged pink shrimp and mysids exposed to slowly changing concentrations of fenthion in the field were similar to what would have been predicted based on laboratory tests that established 24-, 48-, and 72-h LC50s. Laboratory pulse-exposure tests were predictive of no-effect and effect pulse exposures in the field. These comparisons demonstrated that predictions of fenthion toxicity based on laboratory test results were valid when field and laboratory exposure regimes were similar.

Clark, James R., and James M. Patrick, Jr. In review. Toxicity of Sediment-Incorporated Drilling Fluids to Lancelets (Branchiostoma caribaeum). Mar. Pollut. Bull. 9p. (ERL,GB 607*).

The 24, 96, or 168-h LC50s of four used drilling fluids or barite incorporated into sediment were determined in toxicity tests with lancelets (Branchiostoma caribaeum), a benthic chordate. The number of lancelets that did not burrow into contaminated sediments was used to calculate EC50s at the same times that LC50s were determined. Observations of the burrowing behavior allowed quantitation of effects after 24-h exposures to each of the drilling fluids whereas lancelet mortality was sufficient to calculate 24-h LC50s for only one drilling fluid. Drilling fluids were less toxic to lancelets when incorporated into sediments than to mysids (Mysidopsis bahia) or benthic invertebrate communities in water-column exposures.

Clark, James R., James M. Patrick, Jr., James C. Moore, and Emile M. Lores. 1987. Waterborne and Sediment-Source Toxicities of Six Organic Chemicals to Grass Shrimp (Palaemonetes pugio) and Amphioxus (Branchiostoma caribaeum). Arch. Environ. Contam. Toxicol. 16:401-407. (ERL,GB 575*).

Grass shrimp (Palaemonetes pugio) were exposed to either waterborne or sediment-source concentrations of fenvalerate, cypermethrin, 1,2,4-trichlorobenzene (TCB), tributyltin oxide (TBTO), triphenyltin oxide, and di-n-butylphthalate in static or flow-through test systems. Similarly, amphioxus (Branchiostoma caribaeum) were tested with fenvalerate, TCB, and TBTO. The LC50 and no-effect and 100% mortality concentrations are reported from 96-hr and 10-day tests. The toxicity of contaminated sediments could be explained by chemical partitioning into overlying or interstitial water. Amphioxus is not recommended as a routine test species because of (1) difficulty in distinguishing severely affected from dead animals, (2) inability to determine the status of burrowed animals without disrupting sediment, (3) their relative lack of sensitivity in acute exposures to toxic chemicals, and (4) difficulty in routine collection of sufficient numbers of animals. Grass shrimp, however, are useful as an epibenthic test species for waterborne and sediment-source toxicants.

Colwell, R.R., and D.J. Grimes. 1986. Evidence for Genetic Modification of Microorganisms Occurring in Natural Aquatic Environments. In: Aquatic Toxicology and Environmental Fate: Ninth Volume, ASTM STP 921. T.M. Poston and R. Purdy, editors, American Society for Testing and Materials, Philadelphia, PA. pp. 222-230. (ERL,GB X518*).

Recent work at a deep ocean dump site off the coast of Puerto Rico has shown that changes in the microbial populations of the receiving waters can be detected, that is, changes in bacterial community structure, over and above seasonal effects, have been documented. Microbial impact of the dumping of wastes occurs at three levels which can be measured. These include the initial effects at the time of dumping, followed by sustained community structural changes, and finally genetic modification of the natural population evidenced by increased incidence of plasmids. The ocean dumping studies were augmented by examination of the incidence of plasmids in bacteria isolated from samples collected at other locations in the Atlantic Ocean, including outfall samples collected at Barceloneta, PR, off shore samples collected at an outfall off Ocean City, MD, and a clean unpolluted site. The incidence of plasmids could be significantly and dramatically related to influx of sewage. Thus, environmental changes already occur as a result of entrance of allochthonous material into the marine environment. It is clear that baseline measurements are necessary to determine genetic alteration already taking place, before effects of entry of genetically engineered organisms to the marine environment can be determined.

Colwell, Rita R. In review. Release of Genetically Engineered Microorganisms into the Environment. Microbiol. Sci. 19p. (ERL,GB X517*).

The survival, fate, and effects of GEM in the environment are discussed. Because organisms, when released, cannot be recalled or always controlled, it is imperative that a full understanding of the risks be known. Predictive ecology must include the new subdiscipline of molecular microbial ecology, if the need for information prior to release of GEM is to be met. One important aspect of deliberate release to be considered is the ability to detect and monitor GEM in the environment. It has been discovered that microorganisms can undergo "dormancy" i.e., enter a viable but difficult or non-recoverable stage. New techniques have been developed, employing immunofluorescent/epifluorescent microscopy, coupled with 5S rRNA sequencing, which allow accurate nongenetic detection of GEM. These techniques have been employed in aquatic systems.

Connolly, John P., Mary E. Cleveland, and Parmely H. Pritchard. In review. Validity of Partition Coefficient as the Adsorption Descriptor in Exposure Concentrations Predictions: Studies with Kepone and Methyl Parathion. Water Res. (ERL,GB 415).

This work investigates three major assumptions implicit in the use of partition coefficient as sole adsorption descriptor: (1) adsorption kinetics are unimportant to fate and transport of the toxic chemical because they are rapid; (2) adsorption is a reversible process; and (3) equilibrium conditions are independent of the individual concentrations of toxic chemical and adsorbing solid, depending only on their ratio. Adsorption of Kepone and methyl parathion was found to be rapid and two-step, a fast adsorption for approximately 5 min. followed by a slower adsorption to equilibrium at 1 to 2 hr. Kinetics of adsorption indicated adsorption rate was controlled by mass transport mechanisms. Kinetics of methyl parathion adsorption were identical for sterile and biologically active systems to the point of sterile system equilibrium. Continued decrease of dissolved 14C and total mass recovery in the active system suggested degradation to an irreversibly adsorbed compound. The results indicate that kinetics can be ignored for small particle size sediments but that reversibility of adsorption cannot be assumed. Equilibrium adsorption of both compounds at constant sediment concentration was described by a linear isotherm. Partition coefficient was, however, an inverse function of sediment concentration, decreasing by as much as an order of magnitude between sediment concentrations representative of suspended sediment and sediment concentrations representaive of bed sediment. Therefore, a single partition coefficient is inadequate for exposure concentration predictions.

Couch, John A. In press. Carcinogenicity Tests: Utilization of Ectothermic Organisms. Presented at "Alternative Approaches to Toxicity Testing" held at Battelle Laboratories, Columbus, Ohio, November 11-13, 1986. 27p. (ERL,GB 599*).

Certain ectothermic species, particularly some teleost fishes, reveal promise as carcinogen assay organisms and as carcinogen sentinel and indicator species in the environment. Reptiles, amphibians, fishes, and bivalve mollusks have been studied in terms of their responsiveness to chemical carcinogen exposures; of these species, fishes have been studied in greatest detail in the last 20 years. Seven to eight species of teleosts have been studied in enough detail to be recommended as laboratory carcinogen assay subjects. These are the rainbow trout, Mekaka, guppy, Rivulus sp., Poeciliopsis sp., sheepshead minnow and the brown bullhead. Bivalve mollusks such as oysters and clams should be studied further as possible models. Many different test systems have been developed for use of aquatic species in carcinogen studies. Neoplasms have been induced in 12 to 14 tissues in different species of fishes. Between 50-60 chemical compounds have been tested in fishes for their carcinogenic potential. Though these areas of research are relatively new, considerable data and information are available on metabolism, pathologic, and environmental effects of carcinogens in ectothermic animals.

Couch, J.A., and L.A. Courtney. In press. DENA-Induced Hepatocarcinogenesis in the Estuarine Sheepshead Minnow (Cyprinodon variegatus): Neoplasms and Related Lesions with Comparisons to Mammalian Lesions. J. Natl. Cancer Inst. (ERL,GB 589*).

Groups of estuarine sheepshead minnows (Cyprinodon variegatus) were exposed to 50-60 mg/l N-nitrosodiethylamine (DENA) for five to six weeks. Exposure was stopped and the fish were then transferred to clean, flowing seawater. Induced liver lesions were studied in periodic samples of fish taken during the next 67 weeks of holding. Most of these lesions were compared to their counterpart lesions in the rat. Certain lesions such as hepatocellular carcinomas, cholangiolar carcinomas, spongiosis hepatis (SH), and cholangiofibrosis in our fish have apparent similar cellular origins and morphogenesis to those lesions in rats, and perhaps in other mammals. SH in the sheepshead minnow apparently arises from perisinusoidal cells and may be a neoplasm of this cell type. The general similarity of response to DENA in sheepshead minnows and rats suggests that this fish has promise as an assay subject for identifying some hepatocarcinogens, and as a sentinal organism for detecting hepatocarcinogens in contaminated coastal waters.

Couch, John A. In review. Enclosed Systems for Testing Microbial Pest Control Agents. U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. (ERL,GB X526*).

This report stems from a workshop held at the EPA, Environmental Research Laboratory, Gulf Breeze, Florida on February 18 and 19, 1986. The workshop and report were requested by the Hazard Evaluation Division of the Office of Pesticide Programs. The report consists of descriptions and documentation of some enclosed, multispecies systems that may be used for laboratory testing of both natural and genetically altered microbial pest control agents (MPCA's--viruses, bacteria, fungi, and protozoa) for possible effects in nontarget species, and ecosystems.

Cripe, Geraldine M., David J. Hansen, Stephanie F. Macauley, and Jerrold Forester. 1986. Effects of Diet Quantity on Sheepshead Minnows (Cyprinodon variegatus) During Early Life-Stage Exposures to Chlorpyrifos. In: Aquatic Toxicology and Environmental Fate: Ninth Volume, ASTM STP 921. T.M. Poston and R. Purdy, editors, American Society for Testing and Materials, Philadelphia, PA. pp. 450-460. (ERL,GB 538).

The influence of food quantity on the effects of chlorpyrifos was determined in early life-stage (ELS) toxicity tests with estuarine sheepshead minnows (Cyprinodon variegatus). Three 28-day ELS tests were conducted simultaneously, each with a different feeding rate: approximately 20, 110 or 550 Artemia nauplii/fish per feeding. In the first group of three tests, growth was reduced significantly (p less than or equal to 0.001) at nearly all feeding rates and chlorpyrifos concentrations tested (3.1 to 52 ug/L). Therefore, a second group of three tests was conducted at lower chlorpyrifos concentrations (0.4 to 6.8 ug/L) and the same feeding rates used in the first series. Chlorpyrifos concentrations that significantly decreased fish growth were greater than or equal to 3.0 ug/L, regardless of feeding rate. Weights of fish at the end of all tests were directly associated with concentration and food. Fish receiving the greatest amount of food weighed 10 times more than those receiving the least and were three times heavier than those in the intermediate feeding rate. In treatments where growth was affected, mean percentage survival ranged from 67% at 52 ug/L to 99% at 3.0 ug/L. The standard deviations for this survival varied from 14 at the lowest feeding rate for fish exposed to 52 u/L to 2.8 for fish fed 550 Artemia per cup in 3.0 ug/L. Bioconcentration factors (amount of chlorpyrifos in tissue divided by average measured water concentrations) and chlorpyrifos in whole fish at exposure concentrations greater than or equal to 3.0 ug/L generally increased with increasing feeding rates and increased chlorpyrifos concentrations. Within the feeding range tested, the quantity of available food was not an important factor controlling differences in growth of Cyprinodon variegatus exposed to chlorpyrifos. However, when food quantity restricted growth, survival of sheepshead minnows was not as reproducible, and variability (standard deviation) increased with decreased food.

Cripe, C.R., E.J. O'Neill, M.E. Woods, W.T. Gilliam, and P.H. Pritchard. In review. Fate of Fenthion in Salt-Marsh Environments: 1. Factors Affecting Biotic and Abiotic Degradation Rates in Water and Sediment. Environ. Toxicol. Chem. (ERL,GB 583*).

Fenthion (Baytex), an organophosphate insecticide, is frequently applied to salt-marsh environments to control mosquitoes. Shake-flask tests were used to study rates of abiotic and biotic degradation of fenthion and the environmental parameters that affect these rates. Water or water-sediment (500 mg dry weight/L) slurries from salt marshes located along the Northwest Florida Gulf Coast were used. Flasks contained 200 ug fenthion/L, and degradation rates were determined by following decrease of fenthion over time. Hydrolysis and biodegradation in water were relatively insignificant fate processes; fenthion disappeared from flasks containing water, formalin-sterilized water, or formalin-sterilized sediment very slowly (half-life equal to or greater than 2 weeks). The presence of nonsterile sediment resulted in a rapid exponential disappearance of fenthion (half-life equal to or greater than 3.8 days). Biodegradation was assumed since sterile sediment systems showed a much slower decrease of fenthion, and the production of polar compounds (hexane-unextractable) from radiolabeled fenthion was greater in the presence of sediment than sterilized sediment.

Cripe, Geraldine M. In press. Occurrence of Mysidopsis bahia (Mysidacea,Mysidae) on the Atlantic Coast of Florida. Crustaceana (Leiden). 2p. (ERL,GB 560*).

A collection of mysids was taken from Link Port Channel, Ft. Pierce, Florida on December 6, 1984, at 20 salinity and 24 degrees C and returned to our laboratory for culture and identification. All twenty-two individuals were identified as Mysidopsis bahia: 15 females, 4 males, and 2 juveniles (sex undetermined). Gravid females averaged 7 mm length (base of eyestalk to posterior ends of uropods, excluding setae) and had a mean brood of 5.4 young (range 2 to 10). Mature males ranged from 6 to 7 mm length (mean 6.5 mm). A sample of these mysids was sent to Dr. Thomas E. Bowman at the National Museum of Natural History and identified as M. bahia.

Cripe, C.R., W.W. Walker, P.H. Pritchard, and A.W. Bourquin. In review. Shake-Flask Test for Estimation of Biodegradability of Toxic Organic Substances in the Aquatic Environment. Ecotoxicol. Environ. Saf. 24p. (ERL,GB 603*).

Disadvantages of current biodegradation tests are examined: the need for high substrate concentrations, lack of parent compound concentration measurements, no estimation of sediment effects, failure to indicate compounds to which microbial populations must adapt to degrade and lack of site-specificity in innocula selection. A modified river die-away test is proposed for determining biodegradability of organic compounds and testing for toxic degradation products. Our test uses shake flasks containing sterile (2% formalin) and nonsterile site water: both with, and without, site sediment (500 mg/liter). Concurrent toxicity testing with mysids or daphnids provides a sensitive assay for the detection of toxic metabolites. Examples of three test compounds are given: methyl parathion, which undergoes rapid, sediment-mediated biodegradation; dibutylphthalate, to which some microbial communities exhibit an adaptation phenomenon; and methoxychlor, which has a relatively low water solubility and high sediment partition coefficient. The relative merits of this test procedure are discussed.

D'Asaro, Charles N. 1986. Egg Capsules of Eleven Marine Prosobranchs from Northwest Florida. Bull. Mar. Sci. 39(1):76-91. (ERL,GB X527*).
Avail. from NTIS, Springfield, VA: PB87-169207.

Egg capsules of eleven prosobranchs are described and illustrated, including Strombus alatus, Murex fulvescens, Urosalpinx perrugata, Favartia cellulosa, Eupleura sulcidentata, Calotrophon ostrearum, Cantharus cancellarius, C. multangulus, Fasciolaria lilium hunteria, Conus floridanus floridensis, and C. jaspideus stearnsi. Enumerations of capsules and embryos, and capsular dimensions, developmental pattern, and observations on reproductive behavior are given.

D'Asaro, Charles N. 1986. Laboratory Spawning, Egg Membranes, and Egg Capsules of 14 Small Marine Prosobranchs from Florida and Bimini, Bahamas. Am. Malacol. Bull. 4(2):185-199. (ERL,GB X533).
Avail. from NTIS, Springfield, VA: PB87-178729.

Specific substrata or locations used for oviposition and external and internal structure of egg capsules produced by small prosobranchs from seagrass beds and coastal splash pools are described. Included are Tricolia affinis affinis (C.B. Adams, 1850), T. thalassicola Robertson, 1958, T. bella (M. Smith, 1937), Puperita pupa (Linne, 1767), Smaragdia viridis viridemaris Maury, 1917, Littorina mespillum (Muhlfeld, 1824), Alvania auberiana (Orbigny, 1842), Rissoina catesbyana (Orbigny, 1842), R. bryerea (Montagu, 1803), Zebrina browniana (Orbigny, 1842), Rissoella caribaea Rehder, 1943, Caecum nitidum Stimpson, 1851, Marginella aureocincta Stearns, 1872, and Granulina ovuliformis (Orbigny, 1841).

Davis, William P. 1986. Role of Rivulus marmoratus in Research on Aquatic Pollutants. J. Am. Killifish Assoc. 19(1):70-80. (ERL,GB 556).

The role of Rivulus marmoratus in research in environmental aquatic research is described. The unique biology of R. marmoratus provides the aquatic toxicologist with the following advantages: 1. Ability to thrive in small volume of water throughout life span; 2. Reproduction through internal self-fertilization; 3. Isogenic clones that allow interclonal and specific intraclonal tissue transplants; 4. Reproductive process in which eggs are laid on a weekly basis throughout the year; 5. Semiamphibious adaptations that contribute to rapid uptake of waterborne and even some airborne compounds.

DeWeerd, Kim A., Joseph M. Suflita, Tim Linkfield, James M. Tiedje, and P.H. Pritchard. In review. Relationship Between Reductive Dehalogenation and Other Aryl Substituent Removal Reactions Catalyzed by Anaerobes. FEMS (Fed. Eur. Microbiol. Soc.) Microbiol. Ecol. 42p. (ERL,GB X529*).

Anaerobic bacteria are known to catalyze the removal of a variety of aromatic substituents including -COOH, -OH, -OCH3, and -CH3, and halogens. We investigated whether reductive dehalogenation was related to other types of aryl substituent removal reactions. A dehalogenating bacterial consortium was tested for its ability to use benzoic acids substitute in the 3 position with the functional groups listed above. In addition to dehalogenation, the enrichment (as well as the dehalogenating pure culture) was able to transform 3-methoxybenzoic acid to 3-hydroxybenzoic acid without a lag. This reaction exhibited Michaelis-Menten kinetics with an apparent Km of 5 uM. To test the hypothesis that the two reactions were related, we developed a mathematical model incorporating a competitive inhibition term to account for the influence of one substrate on the degradation of the other. However, experimental evidence showed no significant difference in the rates of 3-chlorobenzoic acid or 3-methoxybenzoic acid degradation in either the presence or absence of the other substrate. The isolated dechlorinating organism strain DCB-1 was able to transform 3-methoxybenzoic acid in the presence of 1 mM thiosulfate, but the dehalogenation of 3-chlorobenzoic acid under such conditions was inhibited. Therefore, it is unlikely that a relationship exists between dehalogenation and other anaerobic aromatic substituent removal mechanisms.

Diaz, R.J., M. Luckenbach, S. Thornton, R.J. Livingston, C.C. Koenig, G.L. Ray, and L.E. Wolfe. 1987. Field Validation of Multi-Species Laboratory Test Systems for Estuarine Benthic Communities. EPA/600/3-87/016*, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 81p.

The major objective of this report was to determine the validity of using multi-species laboratory systems to evaluate the response of estuarine benthic communities to an introduced stress. In a 5-year period, experiments in Apalachicola Bay, Florida, and the York River, Virginia sought to (1) develop criteria for microcosm tests to evaluate the capacity of microcosms to model natural communities in the presence and absence of pollution-induced stress and (2) assess the validity of extrapolating test results of one location to another. Individual species response patterns in the microcosms were highly variable and seldom showed good agreement with patterns in the field. Species richness in the microcosms and field sites showed good temporal agreement and provided a conservative indicator of community response to a toxic stress. An ecologically based guild approach to grouping species proved to be a powerful and reliable method of extrapolating from microcosm test results to responses of field communities.

Duke, T.W., and P.R. Parrish. In press. Drilling Fluid Test Procedure: Participation, Data Comparison and Implementation. Presented at the Ninth Annual Analytical Symposium Sponsored by EPA Office of Water Programs, June 19-20, 1986, Norfolk, VA. 6p. (ERL,GB 570*).

The proposed Best Available Technology (BAT) guidelines for discharge of drilling fluids from off-shore oil and gas platforms require that a toxicity test be conducted on certain drilling fluids. This paper describes participation of the Environmental Research Laboratory, Gulf Breeze, in evaluating the toxicity test methods and conducting the tests. Practical aspects (availability of animals, suitable facilities, effort required) of conducting such tests are discussed. Also, interpretation of the results of the tests with reference to biological variation and regulatory needs is presented.

Duke, Thomas W., and Donald I. Mount. In press. Toxic Effects on Individuals, Populations and Aquatic Ecosystems and Indicators of Exposure to Chemicals. Presented at the WHO Workshop on Methodologies for the Safety Evaluation of Chemicals, August 11-17, 1985, Mexico City, Mexico. 21p. (ERL,GB 550*).
Avail. from NTIS, Springfield, VA: PB85-237428

This paper presents two research approaches that address problems encountered in evaluating the effects of complex mixtures of chemicals on aquatic systems. The concept of ambient toxicity testing is applied to the impact of effluents in freshwater receiving waters (the concept also applies to saltwater systems), where measurement of toxicity is made without attempting to identify the toxics. Another approach develops structural and functional indices that can be used to evaluate impact of chemicals on communities maintained under controlled conditions in the laboratory. One approach is concerned with chemicals already in the environment; the other, with developing ecosystem level indices used to evaluate chemicals before they reach the environment. Also, applicability of laboratory-derived data to field situations is discussed.

Environmental Research Laboratory, Gulf Breeze, FL. 1987. Gulf Breeze Laboratory Titles and Abstracts: 1986, 1987, in Press and in Review Publications. U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 79p. (ERL,GB SR-104).

This report represents an effort to provide Agency administrators, managers, and scientists with the most timely information about availability and content of the Gulf Breeze Laboratory research program. Full text, a report copy, or a reprint can be provided on request to: Elizabeth Pinnell (904) 932-5311 or (FTS) 686-9011. This format is intended as a service to Agency users who may wish not only to examine the title and abstract of a publication or report, but who also have a need to know of the availability of technical documentation. To facilitate usage, publications are indexed by title keywords and author.

Environmental Research Laboratory, Gulf Breeze, FL. 1986. Publications: Gulf Breeze Laboratory. SR-107, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 250p. (ERL,GB SR-107).

This report lists all published in-house and extramural reports, publications and journal articles issued by the Environmental Research Laboratory, Gulf Breeze, during the years 1970 through the present time. It is divided into 3 sections: standardized citations grouped by year of issue, a first author index, and a key-word title index.

Federle, Thomas W., Robert J. Livingston, Duane A. Meeter, and David C. White. In press. Quantitative Comparison of Microbial Community Structure of Estuarine Sediments from Microcosms and the Field. Can. J. Microbiol. 24p. (ERL,GB X467*).

Estuarine mud-flat sediments in microcosms and the field were compared with regard to microbial community structure. Community structure was determined by analyzing the fatty acids derived from the microbial lipids in the sediments. Fatty acid profiles were compared using a multivariate statistical approach. Experiments were performed using sediments from St. George Sound and Apalachicola Bay, Florida. The community structure of St. George Sound sediments was controlled by epibenthic predators. In Apalachicola Bay, the dominant influences were physical factors related to the flow of the Apalachicola River. In the St. George Sound experiment, microbial communities in the microcosms differed from those in the field after only two weeks, and the degree of this difference increased substantially as time progressed. In the Apalachicola Bay experiment, although microbial communities in the microcosms were detectably different from those in the field, the degree of this difference was not large nor did it increase with time. This differential behavior of sediment communitie from different sites may be related to the different ecological factors regulating community composition at these sites.

Fisher, D.J., J.R. Clark, M.H. Roberts, Jr., J.P. Connolly, and L.H. Mueller. 1986. Bioaccumulation of Kepone by Spot (Leiostomus xanthurus): Importance of Dietary Accumulation and Ingestion Rate. Aquat. Toxicol. 9(2,3):161-178. (ERL,GB 580*).

The relative extent of dietary accumulation and bioconcentration of Kepone by spot (Leiostomus xanthurus) was quantitatively evaluated at food rations of 4, 8 or 20% of the average wet weight of fish. [14C]Kepone was utilized to determine bioconcentration and dietary accumulation separately, while [14C]Kepone-contaminated food (grass shrimp, Palaemonetes pugio) and unlabeled Kepone in water were used to determine simultaneously accumulation from both sources. Grass shrimp and spot were exposed to the same aqueous Kepone concentration (0.04 ug/l). A first-order pharmacokinetic equation was used to model Kepone accumulation kinetics during the 19-day uptake and 28-day clearance phases. A doubling of contaminated food ration caused a doubling of the whole-body Kepone concentration in spot. Spot fed 8% ration of uncontaminated food and exposed to aqueous Kepone did not bioconcentrate significantly greater amounts of the pesticide than fish fed 4% ration and exposed to the same aqueous concentration. When spot were exposed to contaminated water and food, Kepone contributions from each source were additive. Feeding rate, however, was very important in determining final Kepone body burdens in spot. The dietary source of Kepone represented approximately 9, 18 and 37% of the total body burden bioaccumulated by fish fed 4, 8 and 20% food rations, respectively, but assimilation efficiencies of Kepone from the food source were low. The laboratory results further suggest that dietary accumulation of Kepone by spot may play an important role in determining final Kepone concentrations in spot in the James River, Virginia.

Flemer, David A., Virginia K. Tippie, Gail B. Mackiernan, Robert B. Biggs, Willa Nehlsen, and Kent S. Price. In press. Characterizing the Chesapeake Bay Ecosystem and Lessons Learned. Presented at the Tenth National Conference, The Coastal Society, New Orleans, LA, Oct. 12-15, 1986. 22p. (ERL,GB 594*).
Avail. from NTIS, Springfield, VA: PB87-166930.

During the scientific study phase, the U.S. Chesapeake Bay Program examined the complex ecological structure and processes of the Bay estuary in a coherent and manageable framework. The framework was supported by a rational spatial scaling or segmentation, with an implicit temporal scale. The historic geological, physical, chemical (water quality), and biological data were analysed within this framework to determine trends, correlations and, where appropriate, causal relationships. The overall process resulted in a synthesis or statement on the environmental condition of the Chesapeake Bay ecosystem. We provide an explanation of the strengths and weaknesses of the approach and suggest improvements in future efforts of this type.

Flemer, David A., Thomas W. Duke, and Foster L. Mayer, Jr. 1986. Integration of Monitoring and Research in Coastal Waters: Issues for Consideration from a Regulatory Point of View. In: IEEE Oceans '86 Conference Proceedings. pp. 980-992. (ERL,GB 581).

Coastal marine ecosystems are characterized by a high degree of natural variability. The weak resolving power of marine science to differentiate between effects ascribable to natural factors versus human intervention often leads to unrealistic expectations of "goods and services" that these ecosystems can provide. This high uncertainty often contributes to faulty communication among scientists, resource managers and the public. We believe that this problem is further enhanced by misunderstandings of the need to integrate monitoring and research. We explain why monitoring is a retrospective activity and the principal way it can become a prospective activity is through hypothesis framing, testing,and modeling. We describe the logic that underpins a program designed to characterize the limits of applicability of extrapolation from laboratory data to the field. This interactive, iterative process couples concepts of monitoring and research so that the research question and method are linked to spatial and temporal scales of ecological variability. Without such considerations, important ecological relationships remain unspecified, thus precluding meaningful approaches to management of such complex but valuable ecosystems.

Foss, Steven S., Lee A. Courtney, and John A. Couch. 1986. Evaluation of a Fungal Agent (Lagenidium giganteum) Under Development as an MPCA for Nontarget Risk. EPA/600/X-86/229, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 12p.

This report deals with the development of methods for nontarget testing of fungal microbial pest control agents (MPCAs). The investigations of Lagenidium giganteum, a natural pathogen of mosquito larvae, a potential registrant as an MPCA, and our selected prototype fungal agent, are presented and discussed. Methods for testing various life stages of shrimp (e.g., Palaemonetes pugio) and embryonic fish (e.g., Menidia beryllina) are outlined and evaluated. Also, salinity tolerance testing of the freshwater fungus is summarized. The methods presented provide relatively simple procedures for single species testing of aquatic fungi and incorporate a positive control assay to confirm the infectivity of the MPCA at the time of testing. Results to date indicate the systems to be viable, inexpensive, and reliable. Preliminary data suggest that the selected nontarget species are not affected by L. giganteum. Future studies and refinements of the L. giganteum systems are under consideration. Additionally, a multispecies test system including plant and animal nontarget species is under development. Future testing will include at least one additional MPCA, a registered postemergent herbicide, Colletotrichum gloeosporioides.

Fredrickson, H., and J. Refermat. In review. Microbial 14CO2 Release and Lipid Biosynthesis from Acetate, Lactate and Glucose in a Spartina Rhizosphere and a Nonvegetated Tidal Flat. Appl. Environ. Microbiol. 25p. (ERL,GB 537*).

The microbial biomass, community structure and community metabolism of sediment cores collected from the rhizosphere of Spartina alterniflora were compared to those of an adjacent nonvegetated tidal flat. Lipid cores injected with 14C-acetate, -lactate, -glucose or -p-cresol were characterized by column and thin layer chromatography, and capillary gas-liquid chromatography/mass fragmentography. The rhizosphere contained three times more lipid (neutral lipid, glycolipid and phospholipid) than the tidal flat sediment. The rhizosphere contained more bl5:0 and less oleic acid than did the tidal flat sediment indicating a predominately anaerobic bacterial rhizosphere community. [35S]-sulfate reduction rates were at least three times faster in the rhizosphere. Acetate, glucose and p-cresol were mineralized faster in rhizosphere cores. Lactate was mineralized four times faster and 14C from lactate was incorporated preferentially into glycolipids and specific neutral lipids in the tidal flat cores. This study shows microbial biomass, community structure and community metabolism were heterogeneous within sediments less than 15 m apart and benthic microbial communities showed substrate preferences for lipid biosynthesis. The catabolic rate of a particular compound in a sediment is not necessarily directly related to: (1) biomass, (2) the rate of catabolism of the compound in a different sediment or (3) the rate of catabolism of a different compound in the same sediment.

Gaetz, Charles T., and Collard B. Sneed. In review. Laboratory Culture and Observations on the Reproductive Biology of the Marine Pelagic Isopod, Idotea metallica (Crustacea; Isopoda). Mar. Biol. (ERL,GB 153*).

Laboratory culture of the marine pelagic isopod, Idotea metallica, is described. I. metallica was reared through multiple generations and observations were made on its reproductive biology. These data are compared with those obtained by others for this and related species. Female I. metallica are capable of producing sequential broods in the laboratory without passing through intervening non-reproductive intermolt periods. Mean brood size is 33 and the mean period between egg fertilization and juvenile release is 16 to 17 days. Juveniles emerge from the marsupium 1.5 to 2.0 mm in length and begin feeding immediately. Sexual dimorphism is evident in 25 to 30 days at which time isopods are 6.0 To 7.5 mm in length. Sexual maturity is attained when isopods reach 10 to 12 mm, resulting in a generation time of 80 to 85 days.

Gaetz, Charles T., Richard Montgomery, and Thomas W. Duke. 1986. Toxicity of Used Drilling Fluids to Mysids (Mysidopsis bahia). Environ. Toxicol. Chem. 5(9):813-821. (ERL,GB X392).

Static, acute toxicity tests were conducted with mysids (Mysidopsis bahia) and 11 used drilling fluids (also called drilling muds) obtained from active drilling platforms in the Gulf of Mexico in U.S. waters. Each whole mud was tested, along with three phases of each mud: a liquid phase with particulate materials removed, a suspended particulate phase composed of soluble and lighter particulate fractions and a solid phase composed mainly of drill cuttings and rapidly settling particulates. These muds represented seven of the eight generic mud types described by the U.S. Environmental Protection Agency for use on the U.S. Outer Continental Shelf. Based on volume:volume preparations of the drilling muds in seawater, the lowest 96-hour LC50s obtained were 26 ul/l for whole mud, 11,400 ul/L for the liquid phase, 726 ul/L for the suspended particulate phase and 1,456 ug/l for the solid phase. The toxicity of the 11 muds tested was apparently increased by the presence of aliphatic components.

Genthner, Fred J., Pramita Chatterjee, Tamar Barkay, and Al W. Bourquin. In review. Genetic Stability of Plasmid DNA in Aquatic Bacteria. Appl. Environ. Microbiol. 20p. (ERL,GB 595*).

Sixty-nine randomly selected, gram negative, freshwater bacterial isolates were screened for their ability to receive and express plasmids from Pseudomonas aeruginosa donors, using a plate mating technique. The plate mating technique identified 26 of the isolates as recipient-active for the self-transmissible, wide host-range plasmid R68, 14% were recipient-active, by RP4 mobilization, for the wide host-range plasmid cloning vector R1162. Frequencies of transfer were compared by using 3 conjugal transfer procedures: broth mating, plate mating, and filter mating. With every recipient tested a solid environment was superior to liquid for transfer. The broth mating technique failed to demonstrate R68 transfer in 63% of the recipient-active isolates. Filter mating, in general, yielded the highest transfer frequencies. The more rapid plate mating procedure, however, was just as sensitive for testing the capacity of natural isolates to participate in conjugal plasmid transfer.

Goodman, Larry R., Geraldine M. Cripe, Paul H. Moody, and Darrel G. Halsell. In review. Acute Toxicity of Malathion, Tetrabromobisphenol-A, and Tributyltin Chloride to Mysids (Mysidopsis bahia) of Three Different Ages. Arch. Environ. Contam. Toxicol. 17p. (ERL,GB 598*).

Mysids (Mysidopsis bahia) of three ages (less than or equal to 1-, 5-, and 10-d-old at test initiation) were confined within the same aquaria and exposed to measured concentrations of malathion, tetrabromobisphenol-A, and tributyltin chloride in separate 96-hr acute toxicity tests. Sensitivities of the three age groups were very similar. Ninety-six hour LC50 values ranged from 2.6 to 3.1 ug/L for malathion and from 1.1 to 2.2 ug/L for tributyltin chloride. The 96-hr LC50 for less than or equal to 1-d-old mysids exposed to tetrabromobisphenol-A was 860 ug/L, and approximately 50% of the 5-and 10-d-old mysids died at 1150 ug/L.

Goodman, Larry R., and Geraldine M. Cripe. 1987. Cage for Use with Small Aquatic Animals in Field Studies. J. Am. Mosq. Control Assoc. 3(1):109-110. (ERL,GB 579).

A cage was developed and used with small sheepshead minnows (Cyprinodon variegatus) and mysids (Mysidopsis bahia) in estuarine field studies. The cages float on their sides and can be deployed at the water's surface or submerged at various depths. Construction materials are noncorrosive, relatively inert, and will withstand cleaning with acetone and a mild bleach solution.

Grimes, D.J., C.C. Somerville, W. Straube, D.B. Roszak, B.A. Ortiz-Conde, M.T. MacDonell, and R.R. Colwell. In review. Plasmid Mobility in the Ocean Environment. Presented at the 10th Symposium on Aquatic Toxicology and Hazard Assessment, ASTM, New Orleans, LA, May 4-6, 1986. 15p. (ERL,GB X523*).

Evidence of plasmid selection and genetic exchange in natural aquatic environments, including the ocean, includes: (I) high incidence of plasmid containing strains in polluted areas, (II) presence of free DNA in natural environments, (III) co-existence of identical plasmids in different co-habiting strains, and (IV) data from in situ plasmid transfer experiments. Current research in our laboratory regarding plasmid mobility in the ocean centers around viable but non-culturable bacteria, cloning of ecologically significant genes, genetic exchange between deep sea bacteria under pressure at low temperature, and development of a 16S ribosomal DNA probe for tracking genetically engineered microorganisms that are released to natural environments.

Grizzle, John M. 1986. Lesions in Fishes Captured near Drilling Platforms in the Gulf of Mexico. Mar. Environ. Res. 18(4):267-276. (ERL,GB X514*).

Fish were collected near two actively drilling, petroleum-well platforms and from control areas near the Flower Garden Banks, a natural reef area in the northwestern Gulf of Mexico. Hepatomegaly (enlargement of the liver) occurred in gray triggerfish Balistes capriscus, creole-fish Paranthias furcifer, wenchman Pristipomoides aquilonaris and southern hake Urophycis floridana collected near platforms. Compared with control fish, creole-fish and vermillion snapper Rhomboplites aurorubens collected near platforms had more frequent gill lesions. Southern hake from platform stations had an increased prevalence of hepatic fatty change. Pathogens were not observed in association with the lesions that were more common in fish collected near platforms. The toxicants causing these lesions cannot be determined from this study because the lesions could have been caused by a wide variety of chemicals.

Hansen, David J., Larry R. Goodman, Geraldine M. Cripe, and Stephanie F. Macauley. 1986. Early Life-Stage Toxicity Test Methods for Gulf Toadfish (Opsanus beta) and Results Using Chlorpyrifos. Ecotoxicol. Environ. Saf. 11:15-22. (ERL,GB 549).

Gulf toadfish (Opsanus beta) were continuously exposed as embryos, sac fry and juveniles to technical chlorpyrifos in two 49-day early life-stage toxicity tests. Survival was significantly (a = 0.05) reduced only in 150 ug/liter. However, toadfish exposed to chlorpyrifos concentrations from 3.7 to 150 ug/liter weighed significantly less than control fish: 9% lower in 3.7 ug/liter to 62% lower in 150 ug/liter. The 96-hr LC50 for juvenile fish was 520 ug/liter. Concentrations of chlorpyrifos in toadfish and bioconcentration factors increased with increasing exposure concentration, a condition not generally observed with other marine fishes and other test chemicals. These results demonstrated the procedures for, and the practicality of, early life-stage tests with this marine species. We recommend the use of the gulf toadfish for comparative toxicity testing and for evaluating the toxicity of substances in conjunction with ontogenetical, physiological and histological investigations of this considerably studied genus. We do not recommend it for routine effects testing.

Hinton, David E., John A. Couch, Swee J. Teh, and Lee A. Courtney. In review. Cytological Changes During Progression of Neoplasia. Aquat. Toxicol. 23p. (ERL,GB X539*).

Cytological changes during progression of hepatic neoplasia in fishes were reviewed with emphasis on recent findings in Cyprinodon variegatus and Oryzias latipes. Hepatocytes are particularly sensitive to toxic changes during early phases of response to carcinogens reflecting both lethal and sublethal alterations. Enzyme histochemical studies reveal marked deficiency of glucose-6-phosphate dehydrogenase, glucose-6-phosphatase and adenosine triphosphatase. Surviving hepatocytes are either enlarged, encircled by cells with small nuclear to cytoplasmic rations, and have altered nuclear morphology suggestive of an inability to divide, or, are smaller, apparently rapidly dividing, and have basophilic cytoplasm. In both species, development of spongiosis hepatis occurred following cytotoxic phases. This lesion apparently provides abundant space for cellular remodeling during neoplastic progression leading to eventual multinodular change. Foci of altered hepatocytes included basophilic, eosinophilic (both species) and clear cells (Cyprinodon variegatus only). Enzyme alterations preceded other morphologic alterations and were seen in cells of foci and tumors suggesting lineage of phenotypic alteration. Cytologic changes within other resident cell populations during neoplastic progression were reviewed.

Kelly, John R., Thomas W. Duke, Mark A. Harwell, and Christine C. Harwell. In review. Ecosystem Perspective on Potential Impacts of Drilling Fluid Discharges on Seagrasses. Environ. Manage. 52p. (ERL,GB X528*).

Potential effects of oil drilling fluid discharges upon Thalassia seagrass ecosystems were examined to provide general insights and to raise specific ecotoxicological issues concerning ecological effects of anthropogenic actions. Microcosm experiments demonstrated effects upon both autotrophic and heterotrophic species, and the processes of primary productivity and decomposition. Significant ecological changes may result from disturbance effects related to the physical presence of higher particle loads, in addition to effects from toxic constituents of drilling fluids. We argue that estimating effects upon both ecosystem processes and biotic composition, and developing broader ecological understanding of the particular ecosystem of concern, are required for environmental assessments seeking to provide a scientific basis for judging the acceptability of environmental changes likely to ensue from human activities.

Klein, Theodore M., and Martin Alexander. 1986. Bacterial Inhibitors in Lake Water. Appl. Environ. Microbiol. 52(1):114-118. (ERL,GB X516). Avail. from NTIS, Springfield, VA: PB87-152617.

The populations of six bacterial genera fell rapidly after their addition to sterile lake water but not after their addition to buffer. The decline in numbers of two species that were studied further, Klebsiella pneumoniae and Micrococcus flavus, occurred even when the buffer was added to sterile lake water. The inhibition of K. pneumoniae by substances in lake water varied with the season of the year, and the rate and extent of decline of both species were different in sterile samples of different lakes. The extent of reduction in the density of K. pneumoniae was independent of initial population size and was diminished by the addition of 10 ug of glucose per ml of lake water. The toxin was removed from lake water by dialysis and by a cation-exchange resin but not by an anion-exchange resin, and it was destroyed by heating. The inhibition of K. pneumoniae was not evident in lake water buffered at a pH value above 8.0. We suggest that toxins may be important in determining the composition of the bacterial community of lakes.

Kokjohn, Tyler A., and Robert V. Miller. In review. Characterization of recA Mutants of Pseudomonas aeruginosa: rec-102 is a Mutant Allele of the Pseudomonas aeruginosa PAO recA Gene. J. Bacteriol. 32p. (ERL,GB X535*).

Several recombination deficient mutations have been isolated in Pseudomonas aeruginosa PAO. None has been shown to be in a recA-like function. A fragment of the P. aeruginosa PAO chromosome which complements Escherichia coli recA mutations was used to probe chromosomal digests of isogenic Rec+ and Rec- strains of P. aeruginosa. When strains containing the rec-102 allele (R. Fruh, J.M. Watson, and D. Haas. Mol. Gen. Genet. 191:334-337, 1983) were compared to rec-102+ strains, a restriction endonuclease polymorphism was observed in DNA showing homology to the recA-complementing plasmid.

Kokjohn, Tyler A., and Robert V. Miller. In review. Characterization of the Pseudomonas aeruginosa PAO recA Analogue and Identification of Its Protein Product. J. Bacteriol. 36p. (ERL,GB X534*).

We have cloned a 2.3 kilobase pair fragment of the Pseudomonas aeruginosa PAO chromosome which is capable of complementing recA mutations of Escherichia coli. The recA-complementing activity was further localized to a 1.5 kilobase pair PvuII-HindIII fragment. The direction of transcription was determined. Southern analysis under conditions of high stringency indicated that DNA sequence homology is shared by the E. coli recA gene and the P. aeruginosa recA analogue. The cloned recA analogue was shown to restore resistance to methyl methane-sulfonate, nitrofurantoin and ultraviolet irradiation to E. coli recA mutants.

Lores, Emile M., James C. Moore, and Paul Moody. 1987. Improved Silica Gel Cleanup Method for Organophosphorous Pesticides. Chemosphere. 16(5):1065-1069. (ERL,GB 571).

Quantitative recovery of some organophosphorous pesticide residues has not been possible with existing silica gel-cleanup procedures. We have developed a modification that permits quantitative recovery of all organophosphorous pesticides tested, except those with a carbamate functional group. The method uses a 3.5 g silica gel column with a 1% acetic acid wash to condition the column prior to the addition of the sample. Percentage recovery and standard deviation of compounds such as phorate and disulfoton are 96 (5.6) and 98 (1.0), respectively. Recoveries range from 92 to 101% for the 11 compounds tested.

MacDonell, M.T., S.C. Morris, B.A. Ortiz-Conde, C.J. Pillidge, and R.R. Colwell. In press. Application of Ion-Exchange High-Performance Liquid Chromatography in the Purification of 5S rRNAs Suitable for Sequence Analysis. J. Chromatogr. 6p. (ERL,GB X520*).

A simple, dependable size-exclusion or ion-exchange method for the liquid chromatographic separation of tRNAs and 5S rRNA is not available. Indeed, the method of choice for purification of small RNA species consists of electrophoretic separation on denaturing polyacrylamide gels. Methods for purifying small oligoribonucleotides using either conventionall,2 or thiol-soluble3 polyacrylamide gels are well developed. In this paper we describe a rapid and reliable HPLC method for purifying of 5S rRNA from biological samples with sufficient homogeneity of the preparations for sequence analysis.

MacDonell, M.T., B.A. Ortiz-Conde, G.A. Last, and R.R. Colwell. In review. Distribution of Mutations in Gram Negative Eubacterial 5S rRNAs and Significance for Sequence Analysis. J. Microbiol. Methods. (ERL,GB X519*).

Alignments of 72 5S rRNAs from Gram negative Eubacteria were used to derive a position-wise frequency distribution of mutations along the 5S rRNA molecule. These empirically derived, position-wise frequencies were used as coefficients for preparation of difference matrices and construction of evolutionary trees. Significance of the observed distribution of mutations in the 5S rRNAs, prepared for the Gram negative eubacteria, as well as its relationship to secondary structure are discussed.

Mayer, Foster L., Jr. 1986. Acute Toxicity Handbook of Chemicals to Estuarine Organisms. EPA/600/8-87/017, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 274p.

All acute toxicity data developed by the Gulf Breeze Environmental Research Laboratory, U.S. Environmental Protection Agency, since 1961 were evaluated for quality. A data base was established for 1,175 tests with 197 chemicals and 52 species of estuarine organisms. The chemicals represent all major groups of pesticides, as well as numerous industrial and inorganic chemicals. The compilation of data presented here is unique in that the research was conducted within one laboratory system by methods that, for the most part, were based on or were the consensus methods in use today. It should serve as a useful data base for the many agencies and organizations concerned with the impact of chemicals on estuarine and marine environments.

Mayer, Foster L., Jr., Kathleen S. Mayer, and Mark R. Ellersieck. 1986. Relation of Survival to Other Endpoints in Chronic Toxicity Tests with Fish. Environ. Toxicol. Chem. 5(8):737-748. (ERL,GB 577).
Avail. from NTIS, Springfield, VA: PB-87-171138.

Hazard assessment of chemicals in aquatic organisms often include chronic toxicity testing. The evaluation of exposure duration and of the life stages tested according to standard test methods has led to the development of shorter chronic toxicity tests. A similar evaluation of biological endpoints (i.e., survival, growth and reproduction) could result in tests that are more economical. We analyzed endpoints for 28 chemicals and seven fish species in 34 chronic toxicity studies. When all endpoints were compared, survival was equal to or more sensitive than all other endpoints 56 to 69% of the time. Individual endpoints were more sensitive than survival 19 to 61% of the time, except for reproduction, which was always more sensitive (although there were few observations). The no observed effect concentration (NOEC) for growth could be predicted from the NOEC for survival by using interendpoint correlations ($r = 0.949$ to 0.974). Ratios of NOECs for survival to those for all other endpoints examined were 5 or less in 93 to 96% of the comparisons (specific endpoint comparisons ranged from 80 to 100%). The determination of the survival endpoint requires less time and money than does the determination of most other endpoints, and it appears adequate for hazard assessments in the initial stage of estimating chronic toxicity. However, a factor of at least 0.2 should be applied to the estimated no-effect concentrations for survival to include other potentially biologically significant effects at least 95% of the time. The factor of 0.2 is based on frequency analyses that resulted in the NOECs for survival being 5 times or less than the NOECs for most other endpoints about 95% of the time. Univariate analyses, however, indicated a range of 0.13 to 0.22 for the factor. A thorough evaluation of other published studies that contain endpoints other than survival should be conducted to define the appropriate factor more accurately.

McKenney, Charles L., Jr. 1986. Critical Responses of Populations of Crustacea to Toxicants. EPA/600/M-86/004, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, Fl. 8p.

The objective of the research summarized herein was to provide information necessary to determine appropriate responses for assessing the long-term effects of various classes of pesticides on estuarine crustacean populations. Dose-response relationships of pesticide toxicity and individual physiological functions were examined and compared for various life stages of estuarine mysids (Mysidopsis bahia), grass shrimp (Palaemonetes pugio), and mud crabs (Eurypanopeus depressus). Correlations between physiological dysfunction of discrete life stages and alterations in the ecological fitness of the population should aid in the selection of sensitive, rapid, and inexpensive monitoring tools for predicting chronic effects of pesticides on pesticide-sensitive estuarine populations.

McKenney, Charles L., Jr. 1986. Influence of the Organophosphate Insecticide Fenthion on Mysidopsis bahia Exposed During a Complete Life Cycle: I. Survival, Reproduction, and Age-Specific Growth. Dis. Aquat. Org. 1(2):131-139. (ERL,GB 552).
Avail. from NTIS, Springfield, VA: PB87-171104.

Survival, growth, and various measures of reproductive performance were examined for an estuarine mysid, Mysidopsis bahia, throughout its life cycle during exposure to the organophosphate insecticide, fenthion (0,0-dimethyl 0-[3-methyl-4-(methylthio) phenyl] phosphorothioate). Concentrations of fenthion responsible for lethality (300 ng reciprocal of 1) did not vary significantly between that observed after 4 d exposure of newly released juvenile mysids and that produced with continuous exposure through maturation and production of young. Exposure of maturing juveniles to 166 ng fenthion reciprocal of 1 postponed the onset of reproduction by 4 d. Both individual fecundity of females and total population production of young were reduced by fenthion concentrations of 79 ng reciprocal of 1 and higher. Suppression of mysid growth rates was evident after only 4 d exposure of juvenile mysids to sublethal fenthion concentrations of 166 ng reciprocal of 1; a lower concentrations (79 ng reciprocal of 1) retarded growth rates of the more rapidly growing advanced juveniles after approximately 2 wk exposure. Reduced survival capacity, retarded growth rates, and diminished reproductive success of mysid populations with chronic, low-level exposure to fenthion would result in lowered production rates of an important prey population for commercially important fish that utilize the estuary as a nursery.

McKenney, Charles L., Jr. 1986. Methods for Determining the Influence of Biochemical Biological Control Agents on Metamorphosis of Marine Crustacea. EPA/600/X-86/234, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 18p.

A historical and rational basis for the use of biochemical biological control agents (BCAs) as insect growth regulators in the control of insect pests is presented. The various major types of BCAs used in this capacity are identified and their unique modes of action are described. Procedures required for testing the influence of the most extensively used BCA to date, a juvenile hormone analog, on the complete larval development and metamorphosis of a marine crustacean are presented in detail. Results utilizing these procedures with the registered juvenile hormone analog, methoprene, are presented. These results are discussed in conjunction with previous studies with this compound and their implications regarding appropriate testing procedures for other biochemical BCAs.

McKenney, Charles L., Jr. In press. Optimization of Environmental Factors During the Life Cycle of Mysidopsis bahia. U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 14p. (ERL,GB X541*).

When considering both survival capacity of Mysidopsis bahia through a complete life cycle and time required for juvenile mysids to become reproductively mature, salinity- temperature conditions of 20 o/oo S and 25 degrees Celsius appear optimal for this estuarine crustacean. Optimization of growth and reproduction in this species requires a feeding density of 2-3 Artemia nauplii per ml of seawater. For M. bahia this food density results in maximum growth, shortest duration prior to initiation of reproduction, and maximum young production.

Middaugh, Douglas P., Michael J. Hemmer, and Daniel E. Penttila. In review. Embryo Ecology of the Pacific Surf Smelt, Hypomesus pretiosus (Pisces: Osmeridae). Pac. Sci. 22p. (ERL,GB 557*).

A study of the ecology of developing embryos of the Pacific surf smelt, Hypomesus pretiosus, was conducted. Embryos were maintained in the laboratory at 7.6, 12.1 and 17.6 degrees C and the time to specific embryonic stages determined. Embryos held at 7.6 degrees C developed to stage 24, 18 days after collection; those held at 12.1 degrees C hatched after 13 days; at 17.6 degrees C hatching occurred 8.5 days after collection. Embryos maintained at 15 degrees C and salinities of 20, 25 and 30 salinity averaged 84% survival. There was no significant difference in survival between the groups (ANOVA, $p = 0.53$). Field observations indicated that embryos are spawned in patches in the upper intertidal zone near the time of high tide. They are attached to gravel substrates by the zona radiata membrane which ruptures and quickly turns inside out at the time embryos are fertilized. After several days of development, stage 18 to 22 embryos detach from the original spawning substrates and are washed seaward and down into the gravel substrate in the intertidal zone. However, there was no significant difference (ANOVA, p is greater than or equal to 0.09) in the number of eggs found at each of 4 depth strata in the upper, middle and lower intertidal zones.

Middaugh, D.P., and M.J. Hemmer. In press. Influence of Environmental Temperature on Sex-ratios in the Tidewater Silverside, Menidia peninsulae. Copeia. (ERL,GB 568*).

The sex-ratios of Menidia peninsulae from Santa Rosa Island, Florida were studied during a 13 month survey. Weekly samples revealed significant deviations from the expected sex-ratio of 1:1. During May-October, young-of-the-year (YOY) females comprised 70 to 94% of the individuals collected in the 32.5 to 62.4 mm SL size class. These females are the presumptive progeny of reproduction during cold to cool fluctuating temperatures, 15.5 to 22.5 degrees C, during February-April. In contrast, collections of YOY Menidia during November-April yielded 34 to 60% females. These individuals are the presumptive progeny of reproductive activity and sexual differentiation in May-August at warm fluctuating temperatures of 25.0 to 29.0 degrees C. The pattern in sex-ratios of older Menidia 62.5 - 102.4 mm SL paralleled that of YOY individuals. The annual (13 month) sex-ratio for collections of YOY and older Menidia was identical at 68% females.

Middaugh, Douglas P., Michael J. Hemmer, and Yara Lamadrid-Rose. 1986. Laboratory Spawning Cues in Menidia beryllina and M. peninsulae (Pisces, Atherinidae) with Notes on Survival and Growth of Larvae at Different Salinities. Environ. Biol. Fishes. 15(2):107-117. (ERL,GB 508).
Avail. from NTIS, Springfield, VA: PB86-208543.

Spawning patterns of inland silversides, Menidia beryllina, and tidewater silversides, Menidia peninsulae, were examined in the laboratory under several combinations of 'tidal' and diel light cycle cues. M. beryllina showed a high frequency of spawning throughout the day when held under constant conditions (24L: OD, current velocity 8 cm sec-1) and when 'tidal' and diel light cycles were presented singly or in combination. In contrast, M. peninsulae demonstrated a high frequency of spawning only when presented a combination of 'tidal' and diel light cycle cues and spawned predominantly at night. Menidia beryllina embryos were euryhaline. Hatching ranged from 73 to 78% at salinities of 5, 15 and 30 0/00. M. peninsulae embroyos showed an inverse relationship between the percentage hatch and the incubation salinity, 90% at 5 0/00 and only 65% at 30 0/00. Survival and growth of larval M. beryllina from the day of hatching through 16 days old was optimal at 15 0/00. Although survival of M. peninsulae larvae was optimal at 30 0/00, no trend was apparent in growth of larvae held for 16 days at 5, 15, or 30 0/00 salinity.

Middaugh, Douglas P., Michael J. Hemmer, and Larry R. Goodman. 1987. Methods for Spawning, Culturing and Conducting Toxicity Tests with Early Life Stages of Atherinid Fishes. EPA/600/8-87/004*, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 56p.
Avail. from NTIS, Springfield, VA: PB87-174934.

Procedures are presented for spawning, culturing and conducting acute and chronic toxicity tests with four atherinid fishes: the inland silverside, Menidia beryllina, Atlantic silverside, M. menidia, tidewater silverside, M. peninsulae, and California grunion, Leuresthes tenuis. Guidelines also are provided for growing of food organisns (Chlorella sp., Brachionus plicatilis, and Artemia sp.) that are required for successful culture and testing of the atherinid fishes.

Middaugh, Douglas P., and Michael J. Hemmer. In press. Reproductive Ecology of the Tidewater Silverside, Menidia peninsulae (Pisces: Atherinidae) from Santa Rosa Island, Florida. Copeia. 26p. (ERL,GB 561*).

The reproductive ecology of the tidewater silverside, Menidia peninsulae, was studied during February 1982 through February 1983 along the shoreline of Santa Rosa Island, Florida. Adult Menidia were observed at low tide spawning on a red alga, Ceramium byssoideum, which was growing in the cracks and crevices of a rocky substrate just below the low tide line. Pinfish, Lagodon rhomboides, were noted preying upon newly spawned Menidia eggs; gut analyses revealed a mean number of 191 eggs in five of the predators. The annual reproductive cycle of Menidia extends from February through July or August with the highest spawning activity during March through June at water temperatures of 16.7 to 30.8 degrees C. A single female with ripe ova was collected in November. On eight occasions, minima in female gonadal indices occurred in association with recurring 3- to 4-day periods of tropic tides, suggesting a tidally mediated spawning cycle attuned to periods of very low tidal amplitude and thus low tidal current velocities. Analysis of young-of-the-year Menidia (6-28 mm SL) revealed several distinct length classes indicating that spawning and subsequent hatching of larvae occurred in periodic pulses throughout the spring and early summer.

Morton, R. Dana, T.W. Duke, J.M Macauley, J.R. Clark, W.A. Price, S.J. Hendricks, S.L. Owsley-Montgomery, and G.R. Plaia. 1986. Impact of Drilling Fluids on Seagrasses: An Experimental Community Approach. In: Community Toxicity Testing, ASTM STP 920. John Cairns, Jr., editor, American Society for Testing and Materials, Philadelphia, PA. pp. 199-212. (ERL,GB 546). Avail. from NTIS, Springfield, VA: PB87-166807.

Effects of a used drilling fluid on an experimental seagrass community (Thalassia testudinum Konig et Sims) were measured by exposing the community to the suspended particulate phase (SPP) in laboratory microcosms. Structure of the macroinvertebrate assemblage, growth and chlorophyll content of grass and associated epiphytes, and rates of decomposition as indicated by weight loss of grass leaves in treated and untreated microcosms were compared. Health of the plants and structure of the macroinvertebrate assemblage maintained in the laboratory were compared periodically with the seagrass community from which the plants and attendant sediment were taken. Treated microcosms were exposed to either 190 parts per million (ppm), volume to volume, of SPP or an equivalent amount of montmorillonite clay. Untreated microcosms received only flowing water from Santa Rosa Sound. Sixteen replicates were provided for each treated and untreated set. There were statistically significant differences in community structure and function among untreated microcosms and those receiving the clay and drilling fluid. For example, drilling fluid and clay caused a significant decrease in the numbers of the ten most numerically abundant (dominant) macroinvertebrates, and drilling fluid decreased the rate at which Thalassia leaves decomposed.

Nelson, M.J., P.H. Pritchard, and A.W. Bourquin. 1986. Aerobic Biodegradation of Trichloroethylene. U.S. Department of the Air Force, Engineering and Science Center, Tyndall Air Force Base, FL. 36p. (ERL,GB 600*).

Samples, suspected of having a capability to biologically transform trichloroethylene (TCE), were provided by Tyndall Air Force Base for verification and characterization of activity. Biological transformation of TCE was not observed in these samples. Other soil and water samples, obtained from the Pensacola area, were therefore screened for TCE degradation activity. One sample was found to have this ability and a gram-negative bacillus, which appeared to be responsible for the metabolic activity was isolated. The isolated organism degrades TCE (up to 3.4 uM) to less than 0.02 uM within 24 hours. TCE degradation occured only when water from the original site of isolation and O2 were in the medium. The isolate converted TCE into CO2 and unidentified nonvolatile products. Phenol, toluene O- and m-cresol were found to replace the site water requirement for TCE metabolism.

Nelson, Michael J.K., S.O. Montgomery, E.J. O'Neill, and P.H. Pritchard. 1986. Aerobic Metabolism of Trichloroethylene by a Bacterial Isolate. Appl. Environ. Microbiol. 52(2):383-384. (ERL,GB 572).
Avail. from NTIS, Springfield, VA: PB87-152609.

A number of soil and water samples were screened for the biological capacity to metabolize trichloroethylene. One water sample was found to contain this capacity, and a gram-negative, rod-shaped bacterium which appeared to be responsible for the metabolic activity was isolated from this sample. The isolate degraded trichloroethylene to CO2 and unidentified, nonvolatile products. Oxygen and water from the original site of isolation were required for degradation.

Nelson, Michael J.K., Stacy O. Montgomery, William R. Mahaffey, and P.H. Pritchard. 1987. Biodegradation of Trichloroethylene and the Involvement of an Aromatic Biodegradative Pathway. Appl. Environ. Microbiol. 53(5):949-954. (ERL,GB 593*).

Biodegradation of trichloroethylene (TCE) by the bacterial isolate strain G4 resulted in complete dechlorination of the compound as indicated by the production of inorganic chloride. A component of the water from which strain G4 was isolated that was required for TCE degradation was identified as phenol. Strain G4 degraded TCE in the presence of chloramphenicol only when preinduced with phenol. Toluene, o-cresol and m-cresol could replace the phenol requirement. Two of the inducers of TCE metabolism, phenol and toluene, apparently induced the same aromatic degradative pathway that cleaved the aromatic ring by meta-fission. Cells induced with either phenol or toluene had similar oxidation rates for several aromatic compounds and had similar levels of catechol-2,3-dioxygenase. The results indicate one or more enzymes of an inducible pathway for aromatic degradation in strain G4 are responsible for the degradation of TCE.

Novick, Norman J., Reba Mukherjee, and Martin Alexander. 1986. Metabolism of Alachlor and Propachlor in Suspensions of Pretreated Soils and in Samples from Ground Water Aquifers. J. Agric. Food Chem. 34(4):721-725. (ERL,GB X530*).

Suspensions of soils treated in the field with alachlor [2-chloro-2',6'-diethyl-N-(methoxymethyl)acetanilide] and propachlor (2-chlor-N-isopropylacetanilide) were tested for their ability to metabolize these herbicides. Less than 8% of 14C ring-labeled alachlor was mineralized in 30 days at concentrations of 10 and 0.073 ug/mL. The soil suspensions mineralized 16-61% and 0.6-63% of ring-labeled propachlor in 30 days at concentrations of 0.025 and 10 ug/mL of suspension, respectively. Although soils converted alachlor to organic products, microorganisms able to mineralize the pesticide could not be isolated. Samples from ground water aquifers mineralized less than 1% of the herbicides at the lower concentrations, but four organic products were formed from alachlor. A mixture of two bacteria mineralized 57.6% of ring-labeled propachlor in 52.5 h. A product of the microbial metabolism of propachlor was identified as N-isopropylaniline. These findings suggest that mineralization is a major means for the destruction of propachlor but not for alachlor in soil.

O'Brien, Mark, and Rita R. Colwell. In review. Rapid Indirect Test for Chitinase Activity Using 4-Methylumbelliferyl-N-Acetyl-B-D-Glucosaminide. Appl. Environ. Microbiol. 11p. (ERL,GB X522*).

One hundred and one strains of bacteria from environmental and clinical sources, most of which were Gram negative, were tested for N-acetyl-B-D-glucosaminidase activity using a filter paper spot test with 4-methylumbelliferyl-N-acetyl-B-D-glucosaminide (4-MNABG) as substrate. The results were compared to those obtained by a conventional plate method for chitinase activity using colloidal chitin as substrate. There was excellent agreement in results for both methods. The filter paper spot test with 4-MNABG has the advantage of being rapid, simple-to-perform and inexpensive. This method should be adaptable to a wider range of microorganisms, particularly those with unusual growth requirements.

O'Connor, Joseph M., and John C. Pizza. In press. Pharmacokinetic Model for the Accumulation of PCBs in Marine Fishes. In: Ocean Pollution, Vol. 1. Krieger Publishing. (ERL,GB X501*).

Pharmacokinetic studies were carried out with striped bass in order to determine assimilation and elimination rate constants for polychlorinated biphenyl (PCB) uptake from dietary sources. Efficient assimilation (85 percent of PCBs in a single dose) and a long elimination half-life (120 h) make it apparent that dietary sources of PCBs are important components of the overall body burden in striped bass. The pharmacokinetics of PCB uptake from food were incorporated into a model designed to predict body burdens. The model presented here takes into account several factors of significance in predicting PCB burdens in cold-blooded organisms with indeterminate growth. These are: (1) changes in diet associated with growth, (2) changes in elimination rate constant due to age-related decreases in metabolism, and (3) migratory movements that may cause changes in exposure to PCBs.

Ogram, Andrew, Gary S. Sayler, Denise Gustin, and Russell L. Lewis. In review. DNA Sorption to Soils and Sediments. Environ. Sci. & Technol. 14p. (ERL,GB X540*).

Deoxyribonucleic acid (DNA) adsorption of five soils, an acid-washed sand, and a lake sediment was investigated. All DNA at environmentally relevant concentrations was adsorbed by soils containing a significant amount of montmorillonite at low to neutral pH values. Studies on the effects of DNA molecular size on adsorption to sand and a sandy soil were described by the Freundlich isotherm model (r2 >0.85), and revealed that the higher the molecular weight, the more the adsorption. The effects of ionic strength (as sodium phosphate buffer) on adsorption showed that adsorption decreases as [PO4=] increases. Organic carbon was found to play a relatively minor role in the adsorption of DNA to these soils. A scheme for the extraction of DNA from soils was also developed.

Parrish, P.R., K.L. Dickson, J.L. Hamelink, R.A. Kimerle, D.J. Macek, F.L. Mayer, Jr., and D.I. Mount. In press. Aquatic Toxicology: Ten Years in Review and a Look at the Future. Presented at the Tenth Annual ASTM Symposium on Aquatic Toxicology and Hazard Assessment, May 4-6, 1986, New Orleans, LA. 30p. (ERL,GB X393*).

This Symposium marks the tenth time that we have gathered as a group of professional scientists who share common goals and ideas concerning the protection of our Nation's aquatic resources. This 10th Symposium seems like a fitting time to reflect on our origins, our successes, and our plans for the future. To that end, several people who have been instrumental in shaping the science of aquatic toxicology and hazard (risk) assessment were invited to present their views on the growth of this science and their ideas about its future. This paper is, then, a collection of those view points which are set down in writing so that others may benefit from the experience of the authors and so that newcomers to this field may benefit by knowing about the roots of aquatic toxicology and hazard assessment. The fact that the science has persisted and grown over the past ten years is a tribute to all those who have contributed their time, energy, and intellect.

Parrish, Patrick R., and Thomas W. Duke. In press. Effects of Drilling Fluids on Marine Organisms. In: Proceedings: 5th International Ocean Disposal Symposium. Robert Krieger Publishing Co., Melbourne, FL. 43p. (ERL,GB 507*).

Drilling fluids, also called drilling muds, are essential to drilling processes in the exploration and production of oil and gas from the U.S. Outer Continental Shelf (OCS). These fluids are usually discharged from drilling platforms into surrounding waters of the OCS and are regulated by the U.S. Environmental Protection Agency (EPA). In a program carried out by the EPA Environmental Research Laboratory at Gulf Breeze, Florida, diverse marine species, as well as microbiotic and macrobiotic communities, were studied. Drilling fluids were toxic to marine organisms in certain concentrations and exposure regimes. Furthermore, the fluids adversely affected benthos physically by burying them or by altering substrates. Toxicity of drilling fluid components, used drilling fluids from active Gulf of Mexico sites, and laboratory-prepared drilling fluids varied considerably. For example, 96-h LC50s were from 25 ul/l-1 to greater than 1,500 ul/l-1 for clams, larval lobsters, mysids, and grass shrimp. In most instances, mortality was significantly (a = 0.05) correlated with "diesel" oil content of the fluids collected from the Gulf of Mexico. Data and model simulations suggest rapid dilution of drilling fluids released into OCS waters, resulting in concentrations below the acute effect concentration for water column organisms tested. Accumulation of fluids and cuttings on the bottom within a few hundred meters of the discharge could adversely affect benthic organisms. There is concern that the potential hazard of drilling fluids may be underestimated in some instances because results of short-term toxicity tests may not reveal subtle effects that could occur at the ecosystem level of biological complexity.

Parrish, P.R., and T.W. Duke. In press. Variability of the Acute Toxicity of Drilling Fluids to Mysids (Mysidopsis bahia). In: Proceedings of the Symposium on Chemical and Biological Characterization of Municipal Sludges, Sediments, Dredge Spoils, and Drilling Muds. American Society for Testing and Materials, Philadelphia, PA. 15p. (ERL,GB 596*).

Numerous factors affect the variability of the acute toxicity of drilling fluids (muds) to mysids (Mysidopsis bahia). Source, composition, and age of drilling fluid sample; preparation of test material; condition of test animals; and skill and experience of the people conducting the tests can influence test results. Despite these confounding factors, our intralaboratory variation of median lethal concentrations (96-h LC50s) for six tests with a laboratory-prepared generic drilling fluid was within a factor of two; interlaboratory variation for seven commercial laboratories that tested the same generic drilling fluid was within a factor of four, the same as reported in the literature for acute toxicity tests with single chemicals. The presence of petroleum hydrocarbons in drilling fluids greatly increases toxicity and, because toxic, volatile fractions may be lost, variability of results from tests with petroleum hydrocarbon-contaminated drilling fluids may be greater than that stated above.

Pettigrew, Charles A., and Gary S. Sayler. 1986. Use of DNA:DNA Colony Hybridization in the Rapid Isolation of 4-Chlorobiphenyl Degradative Bacterial Phenotypes. J. Microbiol. Methods. 5:205-213. (ERL,GB X525*).

DNA:DNA colony hybridization techniques were used to select isolates from freshwater sediment samples that contain genes homologous to plasmid pSS50, coding for 4-chlorobiphenyl biodegradation. A high degree of resolution was achieved in which target organisms representing 0.3% of the total population were discerned. Initially, eight positive cultures were obtained, these were found to exist as consortia populations. Pure cultures, from the consortia, were then isolated and screened for 4-chlorobiphenyl degradative genes by DNA:DNA colony hybridization. Each strain demonstrating positive hybridization was subsequently shown to biodegrade 4-chlorobiphenyl to 4-chlorobenzoate. Following phenotypic characterization of the pure cultures it was found that three different organisms were repeatedly isolated from the various consortia populations. Field sampling to isolation of positive strains was accomplished within one week and completely avoided primary enrichment cultivation.

Price, W. Allen, John M. Macauley, and James R. Clark. 1986. Effects of Drilling Fluids on Thalassia testudinum and Its Epiphytic Algae. Environ. Exp. Bot. 26(4):321-330. (ERL,GB 555).
Avail. from NTIS, Springfield, VA: PB87-178661.

A flow-through microcosm system was developed to assess the potential influence of drilling fluids on Thalassia testudinium and its epiphytic algae. Two treatments (drilling fluid and a montmorillonite clay) and a control were used for seven tests: two 10-day, 200 ul/l exposures; two 10-day, 1000 ul/l; and three six-week, 190 ul/l. Six-week exposure to drilling fluid reduced epiphyte biomass (measured as ash free dry weight/cm2), but surviving algae did not differ (measured as chlorophyll a/g epiphyte ash free dry weight) from controls. Thalassia productivity (carbon uptake and growth rate) was reduced by 10-day exposure to drilling fluid concentrations of 200 ul/l and 1000 ul/l. Thalassia productivity was reduced by drilling fluid exposure in summer and fall but not in spring. The variation in response is attributed to seasonal changes in Thalassia allotment and storage of carbohydrates. The effect of montmorillonite clay exposure varied inconsistently among all tests for both Thalassia and epiphytes.

Pritchard, P.H. In review. Assessing the Biodegradation of Sediment Associated Chemicals. In: Workshop Proceedings: Toxicity and Fate of Chemicals in Sediments. 49p. (ERL,GB 530*).
Avail. from NTIS, Springfield, VA: PB86-11657/AS.

Investigations of the fate of xenobiotic chemicals in laboratory systems that accommodate the microbial ecology of sediments are described. These systems permit examination of biochemical activities in the sediment bed with particular emphasis at the sediment-water interface. Sediment may contain thousands of microcommunities, each containing the same genotypic array of metabolic potential. Each community, however, will demonstrate, depending on the surrounding conditions, a certain phenotypic response that reflects a small portion of its total metabolic potential.

Pritchard, P.H., C.R. Cripe, W.W. Walker, J.C. Spain, and A.W. Bourquin. In press. Biotic and Abiotic Degradation Rates of Methyl Parathion in Freshwater and Estuarine Water and Sediment Samples. Chemosphere. (ERL,GB 513).

Statistical analysis of degradation rates of methyl parathion samples from two Gulf Coast estuaries over a three-year period indicated that biodegradation occurred in the presence of sediment but was insignificant in water. Sediment rates always showed the same relative five-fold difference at a primary site within each estuarine area. Samples from 11 ancillary sites indicated biodegradation rates in sediments can be subdivided into two groupings which were independent of seasonal differences (excluding temperature). Spatial variations in rates, therefore, may be of minor environmental significance for this chemical in estuarine areas.

Pritchard, P.H., L.H. Mueller, J.C. Spain, and A.W. Bourquin. 1986. Degradation of Jet and Missile Fuels by Aquatic Microbial Communities. U.S. Air Force, Tyndall AFB, Panama City, FL. 177p. (ERL,GB 590*).

The fate of jet fuel (JP-4) in aquatic sediments was studied concomitantly in laboratory test systems and in the field. Sediments from an estuarine pond were dosed with jet fuel and then reapplied to the pond as well as into plexiglass trays on the sediment bed and quiescent bottle tests in the laboratory. Thirty-three selected hydrocarbons in the jet fuel were followed chemically to quantitate relative hydrocarbon losses. Several hydrocarbons which biodegraded or rapidly volatilized in the bottle tests were much slower to disappear in the field and the plexiglass trays. In general, mixing of the jet fuel with sediments increased the persistence of the associated hydrocarbons. The high density missile fuels RJ-5 and JP-9 resisted biodegradation when incubated with water/sediment suspensions collected from aquatic habitats. RJ-5 and JP-9 were not toxic to the microbial communities at concentrations of 400 mg per liter, but RJ-5 was toxic to Mysidopsis bahia in 96-hour acute tests (LC50 88 ug/l).

Pritchard, P.H. 1986. Extrapolation of Laboratory Biodegradation Information to Microcosms and Field Studies: A Summary of Research Results. EPA/600/X-86/223, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 31p.

The ability to extrapolate biodegradation information from the laboratory to the field has been examined. Site-specific extrapolations were performed by comparing the fate of p-cresol, p-nitrophenol, fenthion, p-chlorophenol and a complex mixture of hydrocarbons in sterile and nonsterile shake flasks and microcosms tests with the fate of each chemical following dosing into a field site. The field sites included a freshwater stream, an estuarine salt marsh and a freshwater pond. Success in extrapolating laboratory data to the field was a function of the environmental complexity that could be modeled in laboratory systems. Microcosms proved to be, within certain limits, excellent analogs to field studies. Physical parameters, such as turbulent mixing and gaseous exchange in the water column, were the most difficult parameters to model.

Pritchard, P.H. 1986. Fate of Pollutants. J. Water Pollut. Control Fed. 58(6):635-645. (ERL,GB 582).

Published literature on the environmental fate of pollutants published during 1984 are reviewed. Short excerpts are presented from each reference covering such areas as photolysis, biodegradation, hydrolosis, sorption, and volatility for pollutants including pesticides, hydrocarbons, heavy metals, polynuclear hydrocarbons, and other toxic organic chemicals.

Pritchard, P.H., C.R. Cripe, L.H. Mueller, and E.J. O'Neill. 1987. Metabolism of Fenthion by Aquatic Microbial Communities. In: Pesticide Science and Biotechnology: Proceedings of the Sixth International Congress of Pesticide Chemistry, IUPAC International Union of Pure and Applied Chemistry, Ottawa, Canada, August 10-17, 1986. R. Greenhalgh and T.R. Roberts, editors, Blackwell Scientific Publications, Boston, MA. pp. 505-508. (ERL,GB 592). Avail. from NTIS, Springfield, VA: PB87-102430.

The microbial metabolism of the mosquito control agent, fenthion, has been studied in shake flask systems containing water and sediment from a salt marsh. The usefulness of this information in describing the fate of fenthion in microcosms and in a field dosing experiment was determined. Our results show that microbial communities associated with the sediment, the presence of invertebrate animals in the sediment bed, and the anaerobic conditions of the sediment contribute significantly to the fate of fenthion under natural conditions.

Pritchard, Parmely H., Carol A. Monti, Ellen J. O'Neill, John P. Connolly, and Donald G. Ahearn. 1986. Movement of Kepone (Chlordecone) Across an Undisturbed Sediment-Water Interface in Laboratory Systems. Environ. Toxicol. Chem. 5(7):647-657. (ERL,GB 487).
Avail. from NTIS, Springfield, VA: PB87-169645.

The distribution of Kepone (chlordecone) in a sediment bed after various periods of continuous toxicant input to the overlying water column was determined in a laboratory system. Most of the Kepone was found to accumulate in the top 0.6 to 1.5 cm of sediment. A mathematical model was developed to predict Kepone concentrations with depth over time in the sediment. An equilibrium partition coefficient was determined from batch sorption tests and a molecular diffusion coefficient for Kepone was estimated from an empirical relationship between diffusivity and molecular weight. A computed Kepone distribution based on diffusion rates that decreased with depth and with incubation time gave the best fit to the observed data. We attribute the apparently faster rates in the upper sediment to mixing between interstitial and overlying water. Our results illustrate the value of models in conjunction with laboratory studies in defining the interactions of pollutants with sediment beds.

Pritchard, Parmely H., Ellen J. O'Neill, Carol M. Spain, and Donald G. Ahearn. In press. Physical and Biological Parameters That Determine the Fate of p-Chlorophenol in Laboratory Test Systems. Appl. Environ. Microbiol. 25p. (ERL,GB 609*).

Shake flask and microcosm studies were conducted to determine the fate of para-chlorophenol (p-CP) in water and sediment systems and the role of sediment and nonsediment surfaces in the biodegradation process. Biodegradation of p-CP in estuarine water samples in shake flasks was slow over incubation periods of 300 hours. The addition of detrital sediment resulted in immediate and rapid degradation evidenced by the production of 14CO2 from [14C] p-CP. The addition of sterile sediment, glass beads or sand resulted in an approximately 4 to 6 times faster biodegradation than observed in the water alone. Densities of p-CP degrading bacteria associated with the detrital sediment were 100 times greater than those enumerated in water. Bacteria in the water and associated with the sediment after preexposure of both water and sediment to p-CP demonstrated enhanced biodegradation. In some microcosms, p-CP was degraded completely in the top 1.0 cm of intact sediment beds. Sediment reworking activities by benthic invertebrates from one site were sufficient to mix p-CP deep into the sediment bed faster than biodegradation or molecular diffusion. p-CP was persistant at lower depths of the sediment, possibly a result of reduced oxygen conditions preventing aerobic biodegradation.

Rao, Kothapalli Ranga, and Philip J. Conklin. 1986. Molt-Related Susceptibility and Regenerative Limb Growth as Sensitive Indicators of Aquatic Pollutant Toxicity to Crustaceans. In: Indian Ocean: Biology of Benthic Marine Organisms: Techniques and Methods as Applied to the Indian Ocean. M. Thompson, R. Sarojini, and R. Nagabhushanam, editors, Oxford & IBH Publishing Co., New Delhi, India. pp. 523-534. (ERL,GB X472*).
Avail. from NTIS, Springfield, VA: PB86-213741.

This study evaluated the comparative toxicity of various pollutants to intermolt and molting grass shrimp (Palaemonetes pugio). Most of the tested materials (pentachlorophenol, tetrachlorophenols, trichlorophenols, methylenebis dichlorophenol, dibutyl phthalate, chromium, and drilling mud) were more toxic to molting shrimp than to intermolt shrimp. Radiotracer studies with 2,4,5-trichlorophenol and pentachlorophenol indicated that the increased susceptibility of newly molted shrimp is linked to increased pollutant uptake. Additional work showed that certain chlorophenols, dithiocarbamates, dibutyl phthalate, and chromium cause inhibition of regenerative limb growth in grass shrimp without affecting the molt cycle duration. The median effective concentrations (EC50s) for inhibition of limb regeneration were well below the medial lethal concentration (LC50s) for intermolt shrimp. Thus, limb regeneration assays with intermolt shrimp as well as toxicity tests with molting shrimp serve as sensitive indicators of aquatic pollutant toxicity.

Ritchie, Scott A., and William P. Davis. 1986. Evidence for Embryonic Diapause in Rivulus marmoratus: Laboratory and Field Observations. J. Am. Killifish Assoc. 19(1):103-108. (ERL,GB 567).

Among North American killifish species, diapause, or arrested embryonic development, has been infrequently noted. Marsh sods sampled from natural vegetated swales yield hatched F. confluentus 15-30 minutes after immersion in the laboratory following 2-3 months in the field. Time-series observations in the Collier County (Florida) Mosquito Control District are reported. The uniform size of the fry collected from the test site, and the brevity of immersion (November 1984-April 1985) before onset of rainy season, mitigate against the survival of previously hatched fish, and argue in favor of embryonic diapause.

Russell, G.A., D.P. Middaugh, and M.J. Hemmer. In review. Reproductive Rhythmicity of the False Grunion, Colpichthys regis, from Estero del Soldado, Mexico. Calif. Fish Game. 12p. (ERL,GB 586*).

The reproductive rhythmicity of the false grunion, Colpichthys regis, was observed in the Estero del Soldado, Mexico during October 1982 through January 1983. Spawning runs occurred at approximately 2-week intervals during daytime high tides. These high tides coincided with new and full moons. Spawning only occurred when predicted tidal heights were equal to or greater than 0.73 m above MLW. Eggs were deposited in the upper intertidal zone in locations that appeared to provide protection from predators, thermal stress and desiccation.

Saye, D.J., O. Ogunseitan, G.S. Sayler, and R.V. Miller. In review. Potential for Transduction of Plasmids in Pseudomonas aeruginosa in a Natural Freshwater Environment. Appl. Environ. Microbiol. 34p. (ERL,GB X536*).

The transduction of Pseudomonas aeruginosa plasmid Rms149 by the generalized transducing phage phi DS1 was shown to occur during a nine day incubation of environmental test chambers in a freshwater reservoir. Plasmid DNA was transferred from a nonlysogenic plasmid donor to a phi DS1 lysogen of Pseudomonas aeruginosa that served both as the source of the transducing phage and as the recipient of the plasmid DNA. Transduction of the plasmid in the presence of the natural microbial community of the reservoir was below the limits of detection employed. The results demonstrate that a potential exists for the transduction of plasmid DNA in aquatic habitats.

Saye, D.J., O. Ogunseitan, G.S. Sayler, and R.V. Miller. In review. The Effect of Plasmid Donor Concentration and a Natural Freshwater Community on Transduction in Pseudomonas aeruginosa. Appl. Environ. Microbiol. 27p. (ERL,GB X538*).

A series of environmental test chambers containing sterile lake water were inoculated with nonlysogenic plasmid-containing Pseudomonas aeruginosa and a lysogen which served as both a source of generalized transducing phage and as a recipient of transduced DNA. A comparable series of test chambers was set up and included the natural microbial community. The concentration of donors introduced into the chambers was varied while the recipient concentration in each chamber was at a level equivalent to natural concentrations of Pseudomonas. The transduction of the plasmid Rms149 in P. aeruginosa was shown to occur in the environmental test chambers during seven days of incubation in a freshwater reservoir. Transduction was observed both in the absence and in the presence of the natural microbial community. The presence of the natural community resulted in a rapid decrease in the numbers of the introduced donors and recipients and a decrease in the number of transductants recovered. The concentration of plasmid-containing donor cells introduced was shown to significantly effect the frequency of transduction. These results demonstrate the potential for naturally occurring transduction in aquatic environments and indicate that donor load may be an important parameter in assessing this potential.

Sayler, Gary S., Rakesh K. Jain, Andrew Ogram, Charles A. Pettigrew, Laura Houston, James Blackburn, and William S. Riggsby. In review. Applications for DNA Probes in Biodegradation Research. Presented at the 4th International Symposium on Microbial Ecology, Ljubljana, Yugoslavia, Aug. 24-29, 1986. 34p. (ERL,GB X531*).
Avail. from NTIS, Springfield, VA: PB87-145322.

The use of DNA:DNA hybridization technology in biodegradation studies is investigated. The rate constants for sediments exposed to synthetic oils could be calculated from the NAH+ genotypes and this approach would be useful in predicting the kinetics of aromatic hydrocarbon degradation. Gene probes prepared from NAH7 plasmid were also used to monitor and enumerate the naphthalene-degrading populations in a continuous mixed culture bioreactor and this analysis demonstrated at least one order of magnitude difference in the naphthalene-degrading population over the conventional plate analysis. It was also shown that using pSS50 (a chlorobiphenyl mineralizing plasmid) as probe DNA, other polychlorinated biphenyls degrading organisms can be identified from the environment. Further, the maintenance and stability of a genetically modified Pseudomonas putida (carrying plasmids TOL and RK2) over an 8-week period in chemically contaminated groundwater aquifer material was established. Results demonstrate the wide applications, signficances, sensitivity, and accuracy of DNA probes in environmental biodegradation research.

Schoor, W.P., D.E. Williams, and J.J. Lech. In review. Combined Use of Biochemical Indicators to Assess Sublethal Pollution Effects on Fundulus grandis, the Gulf Killifish. Mar. Environ. Res. 16p. (ERL,GB 565*).

Sublethal biochemical markers were used to identify liver enzyme induction in fish from a bayou in Pensacola, FLorida. Gulf killifish, Fundulus grandis, from a nonpolluted site were used in the study which included laboratory-induced fish and their various controls as well as the fish captured in the bayou. The biochemical markers tested were liver to body weight ratios, total content of cytochrome P-450, mixed function oxygenase and ethoxyresorufin-0-deethylase activities, and the specific induction of the LM4b isozyme of the cytochrome P-450 system. The findings suggest that enzyme induction occurs at a sublethal level, indicating the presence of liver enzyme inducing substances in the bayou.

Shirley, Michael A., and Charles L. McKenney, Jr. 1987. Influence of Lindane on Survival and Osmoregulatory/Metabolic Responses of the Larvae and Adults of the Estuarine Crab, Eurypanopeus depressus. In: Physiology Pollution of Marine Organisms. Winona B. Vernberg, editor, University of South Carolina Press, Columbia, SC. pp. 275-297. (ERL,GB 562*).

Short-term exposure to sublethal concentrations of the organochlorine insecticide, lindane, caused alterations in ionic and osmotic regulatory abilities and related compensatory metabolic mechanisms in the xanthid crab Eurypanopeus depressus. A lindane exposure concentration of 1.45 ug/L reduced hemolymph osmotic concentrations in adult crabs; however, chloride ion regulation was more sensitive, being disrupted at a lindane exposure concentration of 0.07 ug/L. Larval stages proved to be more sensitive to lindane exposure than adults. A lindane exposure concentration of 0.01 ug/L increased larval mortality and altered larval respiration and ammonia excretion rates. Zoeae, megalopae and adults of the crab, E. depressus, appear to possess different response patterns to hypoosmotic stress and lindane exposure.

Sinclair, James L., and Martin Alexander. In review. Effect of Bacterial Growth on Protozoan Predation in the Presence of Alternative Prey. Appl. Environ. Microbiol. 19p. (ERL,GB X537*).

A study was conducted on the influence of growth rate and initial population size on the survival of bacteria subjected to grazing by protozoa. In a mixture containing Tetrahymena thermophila and a streptomycin-resistant Bradyrhizobium sp., the growth rate of Salmonella thompson was varied by adding differing concentrations of streptomycin. S. thompson initially increased in number, but the population density fell as grazing pressure increased. The organisms that grew the fastest in culture declined to a smaller extent than the slow growers. The decline occurred in sewage containing protozoa but not in samples from which protozoa had been eliminated. In sewage inoculated with 70 to 190 cells per ml of the test species, the densities of two of the three fast growing bacteria increased, but the numbers of the slow growing test organisms declined. In protozoa-free sewage, the abundance of the three fast growing but not the slow growing species declined. In cultures containing T. thermophila, a test bacterium, and a high desity of Enterobacter aerogenes cells as alternative prey, only a fast growing Pseudomonas sp. of three test bacteria increased appreciably in abundance. Based on these data, we suggest that in environments supporting active predation by protozoa, bacterial species that grow quickly and reach high densities will be dominant among the surviving prey species.

Spain, J.C., and C.C. Somerville. In review. Biodegradation of Jet Fuel by Aquatic Microbial Communities. In: Proceedings: 2nd International Symposium on Microbial-Enhanced Oil Recovery, Georgia State University, Atlanta, GA, August 16, 1984. Georgia State University, Atlanta, GA. 23p. (ERL,GB X485*).
Avail. from NTIS, Springfield, VA: PB85-191971.

This paper describes laboratory experiments that studied the fate of jet fuel in several types of situations that could be encountered in the field. Benzene, toluene, and p-xylene were the only components of the fuel that dissolved in the water to significant concentrations. All three compounds volatilized within 24 h and, thus, did not remain in the water long enough for microbial degradation to affect their fate. Inclusion of sediment (500 mg/l dry weight) did not retard the disappearance of the fuel components, and rates of disappearance were identical in controls sterilized with HgCl2.

Tagatz, M.E., R.S. Stanley, G.R. Plaia, and C.H. Deans. 1987. Responses of Estuarine Macrofauna Colonizing Sediments Contaminated with Fenvalerate. Environ. Toxicol. Chem. 6:21-25. (ERL,GB 569).

Macrobenthic animal communities that colonized uncontaminated and fenvalerate-contaminated sand (0.1, 1 and 10 ug/g dry weight, nominal) in boxes placed for 8 weeks in an estuary were compared to assess effects of fenvalerate on community structure. As much as 27% of initial concentrations of this synthetic pyrethrin persisted in sediment at the end of the test. The average number of species (35.6) in communities in five replicates exposed to 10 ug/g was significantly less than that in the control (47.8) and lower concentrations (45.0 and 46.2). Of the dominant phyla collected (Annelida, Mollusca, Chordata, and Arthropoda), abundance of chordates only (primarily lancelets, Branchiostoma caribaeum) was reduced by 10 ug fenvalerate/g. Biological indices applied to the data showed the greatest structural differences for communities exposed to the highest concentration, but these did not differ substantially from those for the control. Effective concentration for exposure via the sediment was five orders of magnitude greater than that for waterborne exposure determined in earlier benthic community studies.

Tagatz, Marlin E., and Roman S. Stanley. 1986. Results of Acute Toxicity Testing of Single Species Dominant in Benthic Community Testing at Gulf Breeze, Florida. EPA/600/X-86/325, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 36p.

This report gives the 96-hr acute toxicities of pentachlorophenol, fenvalerate, dibutyl phthalate, or 1,2,4-trichlorobenzene to various mollusks (Laevicardium mortoni, Ensis minor, Mulinia lateralis) and annelids (Armandia maculata, Neanthes succinea, Capitella capitata) and to an arthropod (Corophium acherusicum), echinoderm (Leptosynapta inhaerens), and a chordate (Molgula sp.). Species were selected from those numerically dominant in earlier benthic community toxicity tests in order to allow comparison between results of single species and community toxicity tests. Acute test results are presented in a series of tables.

Tagatz, Marlin E. 1986. Some Methods for Measuring Effects of Toxicants on Laboratory- and Field-Colonized Estuarine Benthic Communities. In: Community Toxicity Testing, ASTM STP 920. John Cairns, Jr., editor, American Society for Testing and Materials, Philadelphia, PA. pp. 18-29. (ERL,GB 529).

Effects of toxicants on estuarine macrobenthic animals that developed in sand-filled boxes in the laboratory and field during eight weeks were determined by comparing community structures in control boxes and in boxes treated with a toxicant. Boxes were colonized in the laboratory by planktonic larvae in continuously supplied unfiltered seawater and in the field by animals that occurred naturally. Field boxes were placed in estuarine waters, either near the laboratory or at salt-marsh sites subjected to contamination by mosquito control pesticide applications. Eight separate studies were conducted using the same test materials in laboratory and field tests. Communities that developed were diverse and averaged 1441 individuals, 30 species, and 6 phyla for laboratory tests and 933 individuals, 51 species, and 8 phyla for field tests. Toxicants were introduced via water, air, or sediment and before, during, or after colonization. Tests with laboratory- and field-colonized communities provided corroborating data as well as date unique to each test. Various structural attributes among laboratory, experimental field, and natural field communities were similar, indicating that data derived from the laboratory and field toxicity tests can have good environmental applicability.

Tagatz, Marlin E., Gayle R. Plaia, and Christine H. Deans. 1986. Toxicity of Dibutyl Phthalate-Contaminated Sediment to Laboratory- and Field-Colonized Estuarine Benthic Communities. Bull. Environ. Contam. Toxicol. 37(1):141-150. (ERL,GB 547).
Avail. from NTIS, Springfield, VA: PB87-152815.

Dibutyl phthalate (DBP), one of a large class of alkyl esters of 1,2-benzene dicarboxylic acid, is used widely in the United States and other countries as a plasticizer for epoxy and PVC resin. Significant amounts of DBP commonly occur in the aquatic environment, including the sediment. Its octanol-water partition coefficient of 5.2 (US EPA 1979) indicates that sorption of DBP by sediment could be substantial in waters polluted by this chemical. Concentrations as high as 89 ppb have been reported in sediment samples from Chesapeake Bay and up to 15.5 ppm in those from the Rhine River. To obtain information on the effects of DBP on estuarine communities exposed via the sediment, we investigated the responses of macrobenthic animals that colonized sand contaminated with this chemical in the laboratory and field.

Tan, B., and Melius P. 1986. Polynuclear Aromatic Hydrocarbon Metabolism in Fishes. Comp. Biochem. Physiol. C Comp. Pharmacol. Toxicol. 83C(2):217-224. (ERL,GB X513*).

The metabolism of PAHs in fishes is reviewed. Oxygenated and hydroxylated Phase I metabolites of PAHs in fish are compared with each other and with those of other mammals. The review emphasizes the metabolites and not the metabolizing enzymes in many fish species. Some implications of chemical carcinogenesis also are discussed.

Trevors, J.T., T. Barkay, and A.W. Bourquin. In review. Gene Transfer Among Bacteria in Soil and Aquatic Environments: A Review. Can. J. Microbiol. 27p. (ERL,GB 584*).

The exchange of genetic material between microorganisms in soil and aquatic environments is considered in light of the potential of foreign gene desamination from engineered organisms to indigenous bacteria. Abundant indirect evidence suggests that natural isolates can serve as donors and recipients of genetic material. Studies have mostly documented such transfer of plasmid coded antibiotic and metal resistances. However, the scarce information which is available indicates that in situ gene transfer occurs at very low frequencies due to biological and physical parameters of the soil and aquatic environments.

Walker, W.W., C.R. Cripe, P.H. Pritchard, and A.W. Bourquin. In press. Biological and Abiotic Degradation Rates of Xenobiotic Chemicals in In Vitro Estuarine and Sediment/Water Systems. J. Agric. Food Chem. (ERL,GB 504*).

Three herbicides, two fungicides, four organophorphorus insecticides, and one miticide (acaricide) were characterized with respect to degradation rate in estuarine water and sediment/water systems using a simple shake flask test. Decay rates for each chemical could generally be described by a first order model. The degradation of hoelon, bravo, bolstar, fenthion, and bolero was biologically mediated. The fastest biodegradation rates occurred when sediment was present. The degradation of trifluralin, dursban, phorate, EPN and pentachloronitrobenzene were primarily by abiotic means. Relative to the other test materials, phorate and bravo, pentachloronitrobenzene, trifluralin, and bolstar reflected intermediate degradation rates. Variability in rates from replicate flasks suggested that a difference in rate within treatments (sterile/active, with and without sediments) of a factor of two or less was probably not significant.

Walsh, Gerald E., Christine H. Deans, and Leslie L. McLaughlin. In press. Comparison of Four Methods for Calculating the EC50 from Algal Population Growth. Environ. Toxicol. Chem. 8p. (ERL,GB 588*).

EC50s (calculated concentrations that would inhibit growth by 50%) of 21 pesticides in unicellular algal toxicity tests were calculated by straight-line graphical interpolation, moving average interpolation, probit analysis and the binomial method. EC50s of 18 tin compounds were calculated by graphical interpolation, moving average and probit methods. A total of 187 tests was analyzed. Values of the EC50 were essentially indentical when calculated by each method, and it is concluded that straight-line graphical interpolation, the simplest and most rapid method, can be used to estimate relative toxic effect on algal population growth.

Walsh, Gerald E., Leslie L. McLaughlin, Michael K. Louie, Christine H. Deans, and Emile M. Lores. 1986. Inhibition of Arm Regeneration by Ophioderma brevispina (Echinodermata, Ophiuroidea) by Tributyltin Oxide and Triphenyltin Oxide. Ecotoxicol. Environ. Saf. 12(1):95-100. (ERL,GB 528).

Effects of water-bourne toxicants on regeneration of arms by the brittle star, Ophioderma brevispina, are described. Regeneration was inhibited by 0.1 ug liter-1 bis(tri-n-butyltin)oxide and bis(triphenyltin)oxide. Both substances are known to act upon the nervous system, and it is suggested that inhibition was caused by neurotoxicological action of the tin compounds or by their direct effect upon tissue at the breakage point. The former is most likely because regeneration is mediated by the radial nerves of brittle stars.

Walsh, Gerald E., Michael K. Louie, Leslie L. McLaughlin, and Emile M. Lores. 1986. Lugworm (Arenicola cristata) Larvae in Toxicity Tests: Survival and Development When Exposed to Organotins. Environ. Toxicol. Chem. 5(8):749-754. (ERL,GB 521).
Avail. from NTIS, Springfield, VA: PB87-171120.

A test is described for the exposure of the lugworm Arenicola cristata to toxicants. Embryos of A. cristata were exposed for 96 and 168 h to bis(triphenyltin) oxide (TPTO), triphenyltin chloride (TPTC), bis(tri-n-butyltin) oxide (TBTO) and tributyltin acetate (TBTA). The toxic effects were death and abnormal development of larvae. Concentrations that killed all animals were 4 ug L-1 (96 h) and 2 ug L-1 (168 h) TPTO; 10 ug L-1 (96 h) and 5 ug L-1 (168 h) TPTC; 4 ug L-1 (96 h) TBTO; and 10 ug L-1 (96 h) and 5 ug L-1 (168 h) TBTA. Abnormal morphology was caused by 0.75 ug L-1 TPTO, 1 ug L-1 TPTC and 5 ug L-1 TBTA. Several developmental stages, from embryo to swimming larvae, were exposed to TPTO. The most sensitive stages were early trochophore and early settled stage. The range of concentrations between 100% survival and 100% mortality was narrow in all tests. The exposure system is simple and detects teratogenicity.

Walsh, Gerald E. 1986. Organotin Toxicity Studies Conducted with Selected Marine Organisms at EPA's Environmental Research Laboratory, Gulf Breeze, Florida. In: IEEE Oceans '86 Conference Proceedings. pp. 1210-1212. (ERL,GB 585).
Avail. from NTIS, Springfield, VA: PB87-102539.

Studies on effect of bis(tri-n-butyltin)oxide (TBTO) and other organotins on marine species have been conducted at the U.S. Environmental Protection Agency's laboratory at Gulf Breeze, Florida, since 1983. First studies were done on two species of algae, Skeletonema costatum and Thalassiosira pseudonana, where 72h EC50s for tributyltins and population growth were 0.35 and 1.16 ug/l, respectively. Two developmental stages of the lugworm, Arenicola cristata, were sensitive to TBTO (96h LC50=4 ug/l). Only 0.1 ug/l inhibited arm regeneration by the brittle star, Ophioderma brevispina. TBTO was less toxic to the grass shrimp, Palaemonetes pugio, (96h LC50=20 ug/l). Continuing studies include research on effects of TBTO on 1-, 4- and 10-day-old mysids and estuarine seagrass communities.

Walsh, Gerald E. In review. Principles of Toxicity Testing with Marine Unicellular Algae. Environ. Toxicol. Chem. 23p. (ERL,GB 606).

Toxicity testing with unicellular algae requires application of the principles of phycology and microbiology to culturing, handling, and exposing the organisms. This brief review describes major aspects of algal toxicity testing, including growth curves, factors that influence population growth in culture (light, temperature, medium composition, pH, and salinity), choice of test species, measurement of population density, enumeration of living and dead cells, numerical expression of toxic effects, and bioaccumulation.

Walsh, Gerald E., Mark J. Yoder, Leslie L. McLaughlin, and Emile M. Lores. In review. Responses of Marine Unicellular Algae to Brominated Organic Compounds in Six Growth Media. Mar. Environ. Res. (ERL,GB 597*).

Marine unicellular algae, Skeletonema costatum, Thalassiosira pseudonana, and Chlorella sp., were exposed to the industrial brominated compounds, tetrabromobisphenol A (TBBP), decabromobiphenyloxide (DBBO), hexabromocyclododecane (HBCD), pentabromomethylbenzene (PBMB), pentabromoethylbenzene (PBEB), and the herbicide, bromoxynil (BROM), in six algal growth media. Saturation concentrations of DBBO (1 mg/l), PBMB (1 mg/l), and PBEB (0.5 mg/l) reduced growth by less than 50%. EC50s of the other compounds varied with growth medium, with high EC50/low EC50 ratios between 1.3 and 9.9. Lowest EC50s, 9.3 to 12.0 ug/l, were obtained with S. costatum and HBCD. It is concluded that responses to toxicants in different media are the results of interactions between algae, growth medium, toxicant, and solvent carrier.

Walsh, Gerald E. 1986. Techniques for Study of Effects and Uptake of Sediment-Associated Chemicals. EPA/600/X-86/134, U.S. Environmental Protection Agency, Environmental Research Laboratory, Gulf Breeze, FL. 41p.

Methods for exposure of epibenthic and benthic marine invertebrates to contaminated sediments are described. The following concerns are addressed: choice of test organisms, composition of the sediment, toxicological endpoints, uptake and depuration. Test animals were: Emerita talpoida (mole crab), Uca pugilator and U. minax (fiddler crabs), Palaemonetes pugio (grass shrimp), Penaeus dourarum (pink shrimp) and Branchiostoma caribaeum (amphioxus). The animals were exposed to fenthion, fenvalerate, bis (tri-n-butyltin) oxide, bis (triphenyltin) oxide, cypermethrin, 1,2,4-trichlorobenzene, di-n-butyl phthalate and PCBs. It is concluded that organic matter must be present in sediment for retention of toxic chemicals, chemical partitioning of toxicants between pore water and sediment particles is an important factor that controls toxicity, quantitative partitioning of toxicants can be predicted for substances such as pesticides and organotins and exposed animals accumulate toxicants from sediments, but depurate them when transferred to an uncontaminated environment. A list of recommendations for future studies on contaminated sediments is given.

Walsh, Gerald E. 1986. Use of Plankton in Aquatic Toxicity Testing. In: Proceedings of the Third International Course in Toxicology and Ecotoxicology, Herriot-Watt University, Edinburgh, Scotland, Sept. 6-13, 1985. J.H. Duffus, editor, WHO. pp. 147-177. (ERL,GB 551*).

Aquatic toxicology is the qualitative and quantitative study of adverse or toxic effects of chemicals and other anthropogenic materials on populations of aquatic organisms. It includes laboratory studies and the field of ecotoxicology, which is concerned with effects of human activities on naturally-occurring populations and communities at sites in nature. Important data for evaluation of effects of toxicants on aquatic systems are gained from work with freshwater and marine plankton. The following discussion is designed to give a brief explanation of the principles of aquatic toxicology demonstrated by tests with plankton. I have interpreted the term "plankton" loosely, to include fish eggs and small swimming forms such as daphnids and mysids. Recent published works that exemplify these principles and that contain important literature references are given. This work stresses methods and principles, and may be read with the author's previous contribution (Walsh, 1983) to the WHO toxicity course, which reviews published effects of toxicants on plankton.

Wang, Yei-Shung, Eugene L. Madsen, and Martin Alexander. In press. Biodegradation by Mineralization or Cometabolism Determined by Chemical Concentration and Environment. Appl. Environ. Microbiol. 17p. (ERL,GB X489*).

Monuron [3-(4-chlorophenyl)-1,1-dimethylurea] was mineralized when added to sewage at a concentration of 10 ug/L but not a 10 mg/L. Organic products were formed at both concentrations. Products with the chromatographic characteristics of 4-chlorophenylurea and 4-chloroaniline were generated during the decomposition of the higher herbicide concentration. Diuron [3-(3,4-dichlorophenyl)-1, 1-dimethylurea] and linuron [3-(3,4-dichlorophenyl)-1-methoxy-1-methylurea] were mineralized when added to sewage at a concentration of 500 ng/L but not at 2.0 mg/L. No evidence for cometabolism of the higher levels of these two herbicides was obtained, but significant amounts of an unknown product appeared at the lower diuron levels. Chlorobenzilate (ethyl 4,4'-dichlorobenzilate) was cometabolized in water samples from Beebe Lake and mineralized if the samples also contained freshwater sediments. Mineralization did not occur if glucose and inorganic nutrients were added to sediment-free lake water. Chlorobenzilate was transformed to organic products but not to CO2 by microorganisms in water samples from three other lakes, but the pesticide was mineralized in sediment-containing water from two of those lakes. The results thus show that a pesticide may be cometabolized at one concentration or in samples from one type of environment and mineralized at a lower concentration or in samples from a different type of environment.

Wolf, P.H., J.T. Winstead, and J.A. Couch. In review. Proctoeces sp. (Trematoda: Digenea) in Australian Oysters, Saccostrea commercialis and Crassostrea amasa. Trans. Am. Micros. Soc. (ERL,GB 605*).

The occurrence of Protoeces sp., a cosmopolitan digenetic trematode, is reported from two different species of Australian oysters. The low prevalence of the helminth is attributed to the intertidal environment inhabited by the Australian oysters.

Wortman, A.T., C.C. Somerville, and R.R. Colwell. 1986. Chitinase Determinants of Vibrio vulnificus: Gene Cloning and Applications of a Chitinase Probe. Appl. Environ. Microbiol. 52(1):142-145. (ERL,GB X515). Avail. from NTIS, Springfield, VA: PB87-152625.

To initiate study of the genetic control of chitinolytic activity in vibrios, the chitobiase gene was isolated by cloning chromosomal DNA prepared from Vibrio vulnificus. Chimeric plasmids were constructed from Sau3A I partial digests of chromosomal DNA by ligating 5 to 15-Kilobase fragments into the BamHI site, i.e., in the Tc(r) gene, of pBR322 (Am(r)Tc(r)). The resulting plasmids were transformed into Escherichia coli DH1. Chitobiase activity of the insert-bearing clones was detected by using a chromogenic substrate, p-nitrophenyl-N-acetylB,D-glucosaminide, and confirmed by the appearance of a fluorescent end product from the hydrolysis of 4-methylumbelliferyl-B,D-N-N'-diacetylchitiobiose. Endochitinase activity was demonstrated by liberation of water-soluble products produced by the degradation of [3H]chitin. Transformation of E. coli Y10R (lacY) with plasmids from chitinase-positive clones restored the lactose-positive phenotype, suggesting the presence of a permease associated with chitinase activity. Physical mapping of plasmids containing the chitinase determinants indicate that transcription of these genes in E. coli may be inititated at a V. vulnificus promoter.

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