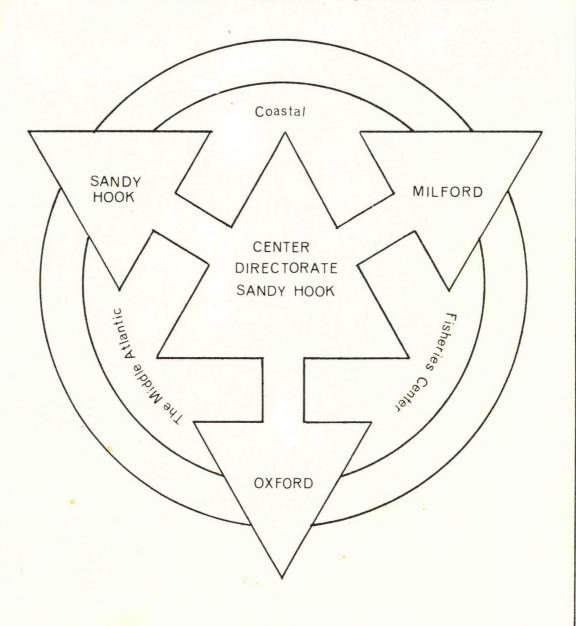
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INVESTIGATION SUMMARIES

U.S. DEPARTMENT OF COMMERCE
National Oceanic and Atmospheric Administration
National Marine Fisheries Service
Northeast Region

MIDDLE ATLANTIC COASTAL FISHERIES CENTER



Informal Report No. 4

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MIDDLE ATLANTIC COASTAL FISHERIES CENTER

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INVESTIGATION SUMMARIES

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INFORMAL REPORT NO. 4

September, 1972

MIDDLE ATLANTIC COASTAL FISHERIES CENTER

INVESTIGATION SUMMARIES

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I. INTRODUCTION: THE MIDDLE ATLANTIC COASTAL FISHERIES CENTER

ORGANIZATION AND MISSION OF THE CENTER

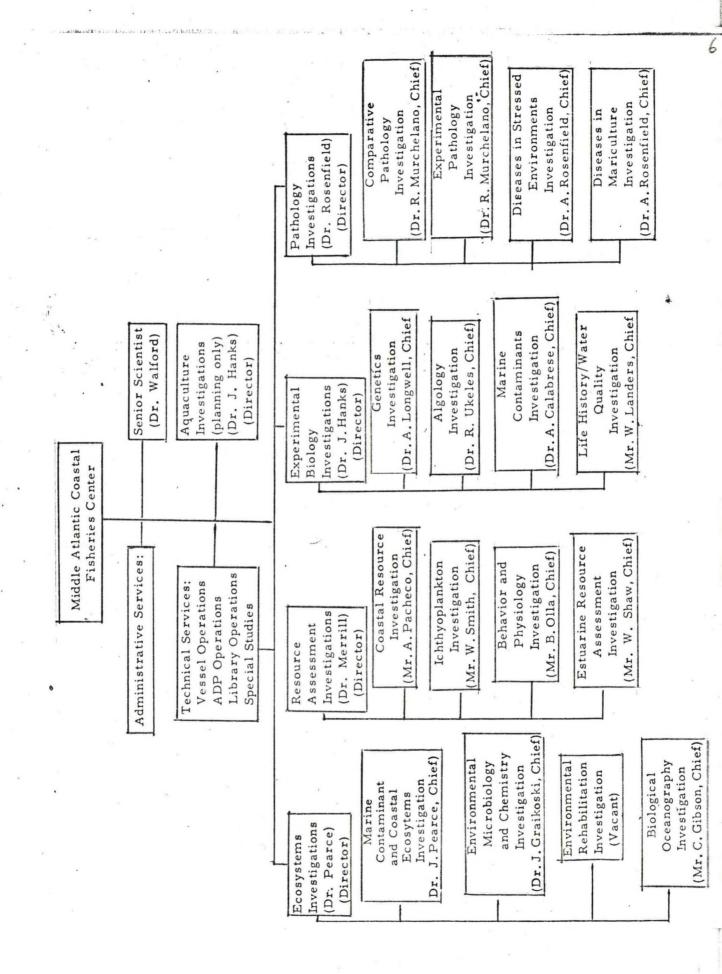
The Middle Atlantic Coastal Fisheries Center is one of a series of seven centers established recently by the National Marine Fisheries Service, National Oceanic and Atmospheric Administration, U. S. Department of Commerce. Each of these centers represents a consolidation and grouping of several laboratories, often in different geographical locations.

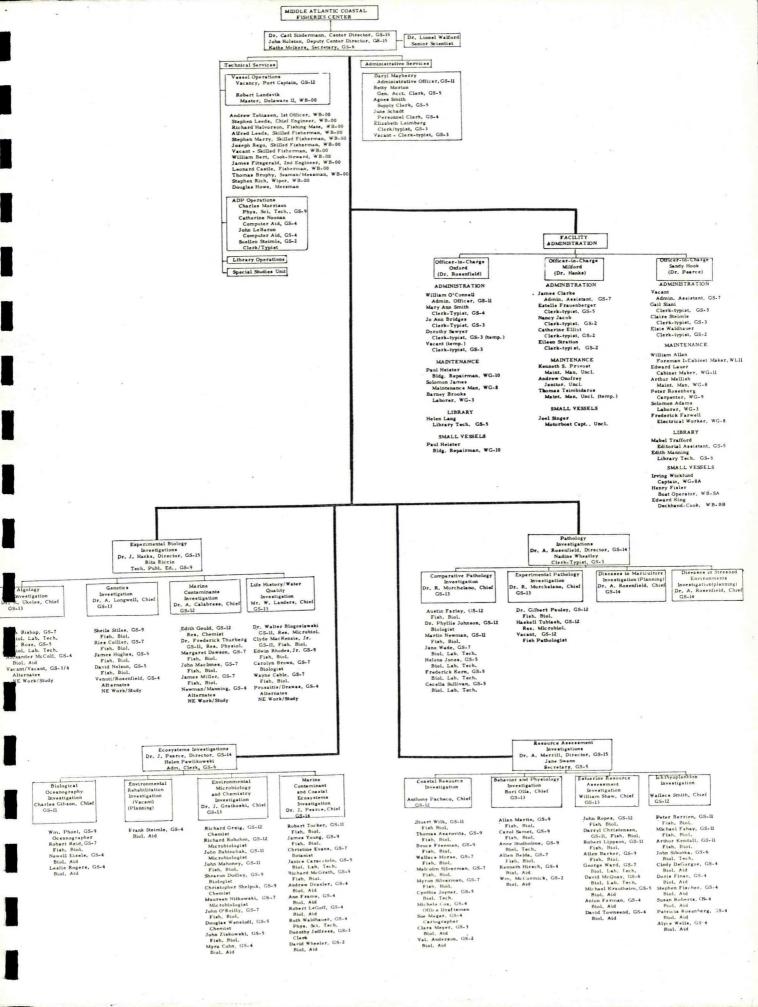
The Middle Atlantic Coastal Fisheries Center is a component of the Northeast Region, National Marine Fisheries Service, National Oceanic and Atmospheric Administration, U. S. Department of Commerce. The Center is a consolidation and integration of the Sandy Hook Marine Laboratory, the Oxford Biological Laboratory, the Milford Biological Laboratory, and the former Ann Arbor Technological Laboratory (now based at Milford). These units (regrouped and subdivided by disciplines) function under direct line authority of the Center Director, Dr. Carl Sindermann, who is responsible to the Director, NMFS North Atlantic Region, for broad integrated programs of research on living coastal resources of the Northeast Region. The Center is composed of a Directorate, an associated centralized Administrative Unit, and five discipline-oriented research units. Research facilities of the Center are located at Sandy Hook, N. J., Milford, Conn., and Oxford, Md.

Major research units may encompass activities at several facilities. Major units are:

Ecosystems Investigations
Resource Assessment Investigations
Experimental Biology Investigations
Pathobiology Investigations
Aquaculture Investigations (planning phase only)

The functional organization of the Center and its major research units is summarized in the attached organizational diagrams.





The mission of the Center is to develop, establish, and to prosecute aggressively an integrated, cooperative multi-disciplinary research program on the biology and ecology of the living marine coastal organisms of the North Atlantic Ocean, especially in the zoogeographic area known as the Middle Atlantic Bight. This program is to be prosecuted in full cooperation with other interested Federal, State, and municipal agencies and with local academic and other research institutions. The research responsibilities of the major research units are as follows:

Ecosystems Investigations (Dr. J. B. Pearce, Director of Investigations) are primarily concerned with benthic-zooplankton food chain studies, physiological responses to toxins and organic wastes, zoogeographic distribution of benthic populations, evolution and succession of reef structures, and with surveys and analyses of the effects of man-made environmental changes on abundance and distribution of marine organisms. A major immediate responsibility is the New York Bight area, where such man-made changes are most profound.

Important aspects of ecosystem investigations concern environmental chemistry and microbiology. This work, located principally at the Milford facility, is primarily concerned with the determination of the level of chemical contaminants in marine resources, food chain organisms and in the environment of the marine animals. It is also concerned with the distribution of microorganisms (bacteria, viruses, fungi and algae) in the marine animals, as well as in the estuarine, inshore and marine environment with particular attention to the effects of man-made changes on the flora and the introduction and survival of potential human pathogens in the marine environment.

The Resource Assessment Investigations (Dr. A. S. Merrill, Director of Investigations) are primarily concerned with assessment of abundance, distribution, and surplus yields of important coastal fish and shellfish species of the Middle Atlantic Bight and adjacent waters. Fisheries biological studies constitute an important aspect of the investigations. Included are: studies of life histories, distributions, migrations, physiology, and behavior of coastal species of the Middle Atlantic Bight and adjacent waters; subpopulation studies using immunogenetic and biochemical techniques, statistical and geographical inventories and atlases of marine sportfish; and larval fish distribution, abundance, and physiology.

Experimental Biology Investigations (Dr. J. E. Hanks, Director of Investigations) are primarily concerned with genetic manipulation of selected mariculture species and with mutagenesis related to environmental factors; as well as with experimental studies of the effects of environmental factors, natural and man-made, on growth, development, behavior, and survival of marine fish and shellfish.

Pathobiology Investigations (Dr. A. Rosenfield, Director of Investigations) are primarily concerned with pathogens which affect living marine resources, with environmental influences on marine diseases, with assessment of the impact of diseases on such resources, and with effective methods of control of diseases in mariculture.

Aquaculture Investigations (Dr. J. E. Hanks, Director of Investigations) are concerned with planning research and development important to aquaculture systems, with special emphasis on molluscan and crustacean species.

The Middle Atlantic Coastal Fisheries Center, because of the nature of existing staff competence and location of facilities, is admirably suited for the development and execution of coherent programs of coastal fisheries research. Among the critical problem areas to be confronted are: Effects of increasing levels of pollution and other manmade environmental changes on the survival and abundance of fishes; biology, ecology, and behavior of species whose distribution or migration extend beyond State boundaries; study and surveillance of living resources of the contiguous zone; study of trends in coastal fisheries production -- both recreational and commercial, and determination of causes for certain obvious declines in abundance and shifts in centers of abundance; need for specialized studies of the impact of disease on marine animals.

Critical immediate problems which need to be confronted by the Middle Atlantic Coastal Fisheries Center include: coordination and integration of research efforts of component laboratories oriented toward a few broad programs (e.g., New York Bight Study; effects of pollutants on fish); increased visibility for NMFS research in coastal resource problems; development of effective cooperative working relations with other research groups (State, Sea Grant, EPA, universities, Corps of Engineers, other NMFS research centers, etc.); development of effective communication with NMFS water resources staff, State conservation agencies, industry segments and sportsmen.

Coastal research areas in which it seems possible to develop high visibility and significant coherent programs include:

- 1. Full assessment of the effects of pollutants and other manmade changes on marine life. The New York Bight Study would be a focus of field and experimental work.
- 2. Analysis of trends in coastal fishery resources -- both recreational and commercial -- with an evaluation of the impact of various factors, man-made and other, on abundance.
- 3. Development of nationally recognized programs in disease and marine genetics (including immunogenetics) -- oriented especially but not exclusively toward mariculture.

The following sections of this volume will outline in detail the objectives and research plans of each of the major investigational areas of the Center. It should be clearly understood that this is an <u>interim</u> document -- one which will require updating and modification, probably on an annual basis, as the research efforts of the Center develop.

RESOURCE ASSESSMENT INVESTIGATIONS

SUMMARY STATEMENT

BACKGROUND The National Marine Fisheries Service (NMFS) has historically been engaged in offshore surveillance of fish stocks along the Atlantic coast. Early cruises were conducted periodically on the highly productive Georges Bank and Gulf of Maine grounds. More recently, studies have been extended into the mid-Atlantic and Chesapeake Bay region.

In the past, this program was deemed sufficient to assess the North Atlantic commercial stocks. However, there has been an increase in fishing pressures by foreign fleets which has decreased the stocks in the traditional North Atlantic grounds and have brought about an increase in fishing intensity in the Middle Atlantic areas.

Concurrently with dwindling stocks of some important commercial species, i. e., yellowtail flounder, haddock and sea scallops, in the apparent degradation of critical estuarine and coastal spawning and nursery grounds. The Connecticut and Hudson Rivers and Delaware Bay are excellent examples of important areas which have been environmentally degraded.

Another significant factors is our technological gains in the field of freezing, holding and processing of fishery products. This has been coupled with swift transportation which has enabled the industry to expand its markets into the heartlands of countries where previously seafood products were primarily consumed immediately adjacent to fishery landing ports. Because of increased fishing pressure by foreign fleets, apparent diminution of suitable spawning and nursery grounds, and an expanded seafood market, the fishery resources along the Atlantic coast of the United States are in jeopardy of being depleted by overfishing. It, therefore, becomes mandatory that the assessment of fishery stocks along the Atlantic coast and adjacent estuaries be augmented in order that proper management policies can be established to save these important fishery stocks. To properly assess the resources along the entire Atlantic coast, a comprehensive and intensive program of integrated research is required. Although Resource Assessment Investigations have ongoing projects to answer some of these needs, further relevant projects are being designed to extend the studies in this large area.

OBJECTIVES Some important objectives which must be achieved in order to develop the skills for future optimal management of marine resources are:

- 1. Assess the abundance and distribution of offshore, coastal, and estuarine finfish and shellfish stocks and determine, from recent historical information, changes in abundance and distribution of important species in the Middle Atlantic Bight.
- 2. Determine the migrational patterns and seasonal changes in geographic distribution of finfish and shellfish stocks and estimate fishing effort -- including segments attributable to both commercial and recreational fishing.
- 3. Determine spawning locations and times by means of ichthyoplankton surveys and compare success of spawning with stock abundance in subsequent years.
- 4. Survey and estimate the total biomass associated within the study areas and describe the environmental conditions under which it exists and collate this information with stomach analyses as an indication of the level of food chain preference...
- 5. Perform descriptive and quantitative behavioral activities related to light, temperature, and other factors in selected species.
- 6. Determine the role of our major estuaries, e.g., Chesapeake and Delaware Bays, play as spawning and nursery grounds for finfish and shellfish.

RECENT AND ONGOING RESEARCH

During the past several years a series of primarily inshore cruises were made by the Coastal Resource Investigations

from Long Island to Florida to obtain information on the distribution, abundance and migration of bluefish and weakfish. Recent cruises have included attention to all fish species and relevant hydrography. These survey cruises are continuing and have been developed to include expanded transect patterns so as to be compatible with the Woods Hole groundfish surveys. The surf clam (Spisula solidissima) program is an ongoing study and includes two broad areas of research -- population dynamics and biology.

Base line studies of important estuarine rivers of Chesapeake Bay are providing data for the determination of the relative condition of the rivers and their utilization as spawning, nursery and feeding grounds by commercial species of finfish and shellfish. Estuarine Resource Investigation biologists are participating in the Chesapeake Bay Study (Corps of Engineers) and are contributing information towards developing a capability for predicting future demands for commercial and sport fisheries.

The Ichthyoplankton Investigation is continuing work on assessing the degree of estuarine dependence of coastal fishes. Spawning seasons and the dispersion of eggs and larvae away from the spawning grounds are being determined. The diurnal habits of larval fishes are being studied, especially the vertical distribution of fish eggs and larvae. Ichthyoplankton specialists are participating in extended surveys of the middle and southern Atlantic as part of a massive ongoing study of ichthyoplankton from the North Atlantic to the Tropical Atlantic Ocean. Intensive work will also be done in accordance with MARMAP, Phase I guidelines. Biologists in the Behavioral Investigation are studying the behavior of tautog and their relation to bottom habitats. Other work in progress includes schooling behavior of mullet and mackerel in reference to light, temperature and habitat.

PROPOSED STUDIES

We propose to expand our efforts toward a more complete assessment of offshore, coastal and estuarine marine resources of the Middle Atlantic area, including

simultaneous larval fish surveys, as well as finfish and shellfish surveys. Extensive offshore cruises with the Delaware II on an expanded strata configuration will be integrated as closely as possible with the ongoing Woods Hole groundfish and ichthyoplankton surveys. Cruises will also include periodic benthic sampling to obtain information relative to the biomass. The area of coverage will extend from Block Island southward and include the ranges of commercial and recreational species which are important in the Middle Atlantic Bight. Additional assessment studies of coastal and estuarine resources will be implemented to complement the ongoing offshore investigation. Coastal areas, inaccessible to larger research vessels, will be sampled on standardized strata similar to offshore strata, utilizing smaller boats. Estuaries, with their shallow water and innumerable tributaries, will be sampled along stations established in specific regions. Finfish and shellfish of general concern within the study area include:

Finfish

Bluefish
Yellowtail flounder
Striped bass
Spot
Shad
Sea bass
Yellow perch
Weakfish
Winter flounder
Spotted hake
Red drum
Menhaden

Eel pout
White catfish
Atlantic mackerel
Silver hake
Tautog
Scup
Sea herring
White perch
Brown bullhead
Spanish mackerel
Red hake

Butterfish
Eel
Fluke
King whiting
Croaker
Pollock
Mullet
Northern puffer
Cunner
Alewife
Blueback herring

Crustacea

Cod

Lobster Cancer crab Blue crab

Mollusca

Oyster Sea scallop Hard clam Bay scallop Soft clam Squid Surf clam Mahogany clam Rangia clam

Of major significance in the attempt to assess fishing stocks is the knowledge of the percentage of resources taken commercially and recreationally. Historical landings data must be reviewed and developed into an appropriate format so that previous abundances and distributions of stocks can be coupled with current information from commercial and recreational fisheries. Current data will be obtained through analyses of port landings and interviews of representatives of commercial and sport fishing interests. In addition to approaches which are clearly assessment and statistically oriented, it is important to maintain a broad spectrum of supporting studies in fishery biology, including studies of life histories, stock identification, physiology, population dynamics, migration, and behavior. Some of these have been

ongoing at facilities of the Sandy Hook Center for a number of years, and these will be integrated into the prime mission of resource assessment. Such studies will provide the necessary scientific and quantitative bases for determining the status of Middle Atlantic stocks. This information, along with improved assessment and statistical capabilities, will allow development of increasingly more accurate predictive models for managerial purposes.

RELATION TO OTHER PROGRAMS Other Investigations are concerned with the current and future status of commercial stocks and to ultimately developing predictive

capabilities. To achieve these objectives the investigations must evaluate water quality and bottom conditions, general health of populations and intensity of commercial fishing. Cooperation and coordination with other NMFS components as well as the various state and university laboratories is, therefore, essential.

RESEARCH PLANNED FY 1973. Acquire necessary equipment and establish sampling design for assessment of finfish and shellfish in coastal and estuarine waters.

Initiate age, growth and fecundity studies. Intensify acquisition of port landings. Continue ichthyoplankton surveys. Continue laboratory studies on the effect of light and temperature on selected marine species and continue field behavior studies on social feeding behavior of benthic fish species.

FY 1974. Continue semi-quantitative sampling of coastal and estuarine species. Examine results to develop stock strengths. Continue ichthyoplankton survey of selected species. Expand behavioral laboratory work to include species not already under study. Expand field program.

FY 1975. Continue coastal and estuarine assessment studies and implement a cooperative state-federal program of ichthyoplankton research in Chesapeake and Delaware Bays. Continue to extend behavioral laboratory work to include species not previously investigated.

RESOURCE ASSESSMENT INVESTIGATIONS

COASTAL RESOURCE INVESTIGATION

BACKGROUND There is an increasing concern over notable decreases of traditional stocks. The expansion of foreign fleet effort, changes in economic motivation which have shifted some fleets to new target species, and an increasing number of sport anglers have pointed up the need for constant assessment efforts. The demands on finfish and shellfish resources require safeguards against overexploitation and environmental degradation which could reduce the biomass. The allocation of our fishery resources is not being implemented in the mid-Atlantic or southern coastal waters. To achieve this end, an ongoing analysis of coastal fisheries stocks is required. This analysis should be based on biological information integrated with economic factors.

OBJECTIVES The objective of the Coastal Resource Investigation is to determine the population dynamics of biological stocks based on such information as growth and mortality rates, and reproductive cycles. A data base is sufficient when resultant information allows reliable estimates of relative abundance. During a regular series of cruises using trawls and dredges, we will collect samples to determine stock ranges and the age, growth and fecundity of target species. Because of seasonal variations in stock abundances, we must be totally aware of the complexity of our fishery stocks and carefully select target species on a priority basis. Assessment studies must include not only samples from our research cruises, but also those from commercial and sport landings as well.

A predictive estimate of future commercial and sport fishery stocks will be derived by analyzing the young-of-year. These studies will be related to surveys in selected estuaries and to samples obtained from MARMAP cruises.

Also required in the assessment of mid-Atlantic stocks is the expansion of our catch and effort information now being collected at selected ports. This is necessary to estimate fishing mortality.

RECENT AND ONGOING RESEARCH

An age analysis of bluefish is being completed and a five-year collection of sciaenid scales from samples taken from mid- and

South Atlantic sites is being read. The species composition in trawl catches from R/V Dolphin cruises is being coded for ADP data bank. The coastal distribution of marine animals, based on surface sightings, with respect to water temperature is being analyzed. An "Angler's Guide to Atlantic Coast Fishing" is near completion. Search on the distribution of juvenile fishes in estuaries is being conducted. Studies on the fecundity, growth, food habits and distribution of important coastal species are being implemented or expanded. Since 1965 the surf clam program has been studying the population dynamics and biology of this important commercial species. Data from seven cruises (1965-70) are being analyzed. Presently, under the Federal-State Partnership Program, an extensive survey is being conducted on the abundance of surf clams along the New Jersey coast. Work in the future will include other shellfish species.

PROPOSED STUDIES

- 1. Cruise data will include recording of all species brought aboard by size, weight and numbers. These data will be associated with hydrographic parameters and included in a continuing series of summary reports.
- 2. Biological studies will concentrate on selected species groups, e.g., sciaenids, sparids and bothids will receive early and special attention.
- 3. Recruitment estimates will be derived from the distribution and relative abundance of young-of-year fish and shellfish. The data will be obtained from cover-bag and dredge samples during the R/V Delaware II cruises. The data will be correlated with independent studies in selected estuaries.
- 4. An information bank will be developed on the age and size composition of species landed by commercial and sport fishermen and compared with samples taken aboard the research cruises.

RELATION TO OTHER PROGRAMS

All investigations concerned with egg and larvae occurrence and density should be related to the young-of-year collections and studies of adult fish. Similar studies carried on at other centers, state and university laboratories, should be coordinated and exchange of data should be given top priority.

RESEARCH PLANNED

FY 1973. Determine sampling design, and acquire shipboard and laboratory equipment and technical support. Compare effectiveness of sampling gear. Begin age, growth and fecundity studies of bottom species. Develop port sampling technique.

FY 1974. Continue semi-quantitative sampling and analyses. Examine results to develop stock hypotheses. Refine port sampling techniques.

FY 1975. Continue to analyze results for consistencies of differences. Begin forecasts. As ongoing priority species analyses become routine, expand the base groups under direct study, including work on pelagic species.

RESOURCE ASSESSMENT INVESTIGATIONS

ESTUARINE RESOURCE INVESTIGATION

BACKGROUND Estuaries have been described as semi-enclosed coastal bodies of water which have free connection with the open sea and within which sea water is measurably diluted with fresh-water derived from land drainage. Estuaries and lagoons with their extensive marsh or wetland shore lines constitute a high percentage of the world's coasts. Approximately 80 to 90 per cent of the Atlantic and Gulf of Mexico coast line consists of estuaries and tidal lagoons.

Estuaries are typically highly fertile and support large phytoplankton populations. The high primary production often sustains abundant benthic organisms and marine fishes which breed in the estuary, since a plentiful food supply exists for their larvae. Chesapeake Bay, for example, annually produces about 80 pounds of seafood per surface acre. However, man-made changes are gradually reducing its carrying capacity.

Greater and greater demands are being placed on the resources of estuaries. As the cities increase in size, e.g., Washington, D. C., Baltimore, and Wilmington, their pollutants degrade the water quality and may cause a reduction in the stocks.

Intensive assessment of estuarine finfish and shellfish populations is necessary to evaluate the effects of lower water quality and commercial and sport fishing on the stocks.

OBJECTIVES The objectives of this Investigation are to delineate the spawning and nursery grounds of finfish and shell-fish; to thoroughly understand their life cycles; to determine the population dynamics of biological stocks from growth, mortality, and reproductive rates; and to develop techniques that will allow reliable prediction of relative abundance.

RECENT AND ONGOING RESEARCH

Biweekly water quality collections and monthly trawl samplings of two Chesapeake river systems (the Choptank and Wicomico) have

provided important base line data in determining the relative conditions of the rivers and their utilization by finfish and shellfish. Recently, a third river, the Miles, has been selected for similar analyses. In addition, quarterly benthic samples are being collected along a 40-mile reach of the Choptank River. Sediment samples are also being collected and analyzed from the Choptank preparatory to the development of a distributional map of the sediments.

Resource Assessment biologists are participating in the Chesapeake Bay Study (Corps of Engineers) and are engaged in a study designed to define the future demand for commercial and sport fisheries.

PROPOSED STUDIES

be attained.

Future research will focus on the assessment of relative stock strengths of important commercial species in Chesapeake and Delaware Bays and their tributaries. Studies will also be developed to determine and delineate spawning and nursery grounds. Cooperative programs with State and other agencies will be expanded so that these important objectives can

RELATION TO OTHER PROGRAMS Information developed within this program will be useful in establishing guidelines by State management agencies. Data has and

will be provided to research institutions around Chesapeake and Delaware Bays and to the seafood industry. Estuaries, such as the Chesapeake, provide important spawning, nursery, and feeding areas for many commercial coastal and offshore species. Estuarine assessment data will be relevant and made available to NMFS investigations concerned with ocean stocks.

RESEARCH PLANNED

FY 1973. A fourth river along the eastern shore of Maryland will be selected for detail assessment. Necessary equipment plus a suitable research vessel will be acquired in order that we can assess the major

fish and shellfish stocks in Chesapeake and Delaware Bays. The acquisition of pertinent fish and shellfish landings will be intensified. FY 1974. We will continue estuarine assessment studies in the two bays and expand activities to inshore coastal waters to ascertain the recruitment impact in estuaries. A fifth river will be assessed along the eastern shore.

FY 1975. We will continue estuarine assessment studies in the bays and implement a co-coperative State-Federal program of ichthyoplankton research.

RESOURCE ASSESSMENT INVESTIGATIONS

ICHTHYOPLANKTON INVESTIGATION

BACKGROUND Fishery scientists generally agree that shallow protected waters of bays, rivers and sounds along our eastern seaboard provide essential habitat for many marine fishes. The occurrence of juvenile fishes in estuaries has led biologists to conclude that a major role of the estuarine zone is its function as a sanctuary or nursery ground for young fishes, many of which are spawned at sea. In 1965, the Sandy Hook Laboratory began a study of the distribution of fish eggs, larvae and juveniles from the inner reaches of the littoral zone to the edge of the continental shelf to elucidate the degree of estuarine dependence of coastal fishes. Impetus for the study resulted from the accelerated degradation of the estuarine zone and a concurrent decline in some of our most valuable coastal fishery yields.

OBJECTIVES To study the early life histories of coastal fishes by: investigating the diel movements of larvae and their effects on both active and passive transport; gathering information on the vital statistics of larvae that will allow prediction of year class indices; maintaining surveillance of spawning areas of selected species to determine yearly fluctuations in abundance; relating offshore occurrence of larvae to distribution of juveniles.

RECENT AND ONGOING RESEARCH

The initial phase of the sampling program involved a survey of the continental shelf from Cape Cod, Mass., to Palm Beach, Fla.,

to locate areas of spawning, to estimate the seasonal duration of spawning and to trace the dispersal of young fishes spawned on the shelf. From the 1500 samples collected, we have information on the young stages of 60 species. With this base line data at hand, we are planning to initiate the second and more quantitative phase of sampling.

PROPOSED STUDIES

Field research will be coordinated to satisfy several objectives. We will study initially the vertical distribution of fish eggs and larvae and investigate diurnal

movements of larvae to better understand the mechanisms influencing transport between spawning areas and nursery grounds. Cruises will be conducted during different seasons of the year and research will concentrate on selected species that spawn in the Middle Atlantic Bight, an area thought to delimit the range of geographically distinct stocks of some fishes and one thought to include the major spawning and nursery grounds of still others. Diel distribution studies will be followed by a series of closely spaced (in time) cruises to study: 1) incubation periods, 2) larval movements between spawning and nursery grounds; 3) food items and feeding periods, and larval growth rates, and 4) mortality. An atlas and other papers dealing with the distribution of larvae from our original surveys are being prepared.

RELATION TO OTHER PROGRAMS

As an integral part of the resource assessment investigation, the ichthyoplankton program will provide useful data for estimating the existing

abundance and distribution of both pelagic and demersal fish stocks. We will depend initially on the estuarine survey and the Long Island Sound studies to provide us with data on estuarine nursery grounds. The program will provide supplemental biological and environmental data to the ongoing MARMAP program. When possible, we will standardize gears for collecting ichthyoplankton and hydrographic data. We will supplement our data with collections from within and outside the Middle Atlantic Bight by utilizing data collected on MARMAP surveys and on groundfish surveys conducted for resource assessment investigations.

RESEARCH PLANNED FY 1973. Study vertical distribution and diel movement of larvae spawned on shelf waters of the Middle Atlantic Bight, using accoustical

link discrete plankton samplers (ALDIPS). Participate in two MARMAP I.O.C. cruises. Continue work on collections from previous cruises.

FY 1974. Begin field work on distribution and transport of larvae from spawning to nursery grounds. Cooperate with teams conducting groundfish surveys along Middle Atlantic Bight to monitor egg and larval concentrations on known spawning grounds.

FY 1975. Continue 1974 field work on larval transport during different season to collect similar information on additional species.

RESOURCE ASSESSMENT INVESTIGATIONS

FISH BEHAVIOR INVESTIGATION

Until recent years, knowledge about the behavior of BACKGROUND marine species has come mainly from the analysis of commercial and sport fish catches, tagging experiments, and from collections of various life stages, from eggs to adults, gathered during scientific expeditions. Over the years, these kinds of studies have proven to be of value in giving us the times and pathways of migrations, the time and place of spawning, food habits and the relation of these findings to various physical and biological parameters. However, there are many questions that have been left unanswered, especially those concerning details of normal daily patterns of behavior and the effect of seasonal changes on them. These behavioral patterns can often have a direct bearing on the life habits of a species. By studying the behavior of selected marine species under controlled laboratory conditions in conjunction with carefully monitored direct visual observations in the sea, it will be possible to define normal patterns precisely and measure quantitatively how they are affected by selected environmental parameters.

OBJECTIVES The objectives of the program are to establish normal patterns of behavior in selected marine fish species in relation to: a) various physical environmental factors such as light and temperature; b) predator and prey organisms; c) bottom habitat, and d) inter-and intra-specific interactions other than predator-prey relations.

These findings may be used as base line indices of normality enabling us to predict with some confidence the effects of environmental modifications, either natural or man-made, and the occurrence and availability of marine species in relation to naturally fluctuating environmental factors. The studies will also provide essential base line measures for future mariculture studies and reef management.

RECENT AND ONGOING RESEARCH

As a primary part of our studies to define normal patterns of behavior in marine fishes, we are currently investigating the roles that

light and temperature play on the activity, feeding and schooling of adult Atlantic mackerel held in a 32,000 gallon experimental aquarium. The results of these laboratory studies will be used to give us further insight on the influence of particular environmental parameters on the distribution and movements of natural populations as well as aiding us in predicting the effects of both man-made and natural stresses on populations.

Another area of ongoing research deals with observing patterns of visually-mediated behaviors such as feeding and schooling in juvenile striped mullet. The results of these studies, besides providing essential information about the habits of this important species, will also serve as base line measures in assessing the subtle effects of various environmental factors, such as temperature, on behavior.

We are currently conducting field studies on daily activities, such as feeding, movement, habitat selection and social interaction of adult tautog in their natural habitat, utilizing ultrasonic tracking methods in conjunction with direct underwater observations. We are also studying their behavior in the laboratory. A primary goal in this study is to define the role that both artificial and natural reefs play in the life habits of this and similar species of northern reef fish.

PROPOSED We will study by experimental methods in both field and STUDIES laboratory the influence of light and temperature on the activity and feeding behavior of selected marine fish species. These studies will involve measuring, statistically analyzing and publishing data on the following:

- 1. The effect of light and temperature on activity cycles.
- 2. Feeding responses and the possible influence of various physical parameters on feeding motivation.
- 3. Relation of various fish species to underwater habitats, including territoriality, feeding range and responses to current.
- 4. Schooling responses and other types of social behavior in both adults and juveniles.

RELATION TO OTHER PROGRAMS

The results of behavioral studies on various fish species relate closely to other assessment activities as well as programs concerned

with environmental contaminants, natural and artificial fish habitats, and mariculture studies. Research performed at other centers and at state and university laboratories will necessitate close cooperation.

RESEARCH PLANNED FY 1973. Continue laboratory studies on the effects of light and temperature on selected marine fish species. Develop statistical

techniques to describe mathematical relations of the various behavioral responses, continuing to publish results. Continue field behavior studies on the social and feeding behavior of benthic fish species, employing both SCUBA and sonic tracking.

FY 1974. Expand laboratory work to include species not already under study and to use a comparative study approach to gain further insight into behavioral mechanisms in a variety of species. Expand field programs to allow study of related species residing in different habitats.

FY 1975. Continue to extend laboratory work to include species not previously investigated. Begin to analyze results from different species studied on a comparative basis allowing the formulation of principles governing the behavior of different categories of fishes such as benthic and pelagics and various subgroups of each. To organize and analyze these results so that they will be of value in predicting the effect of varying environmental factors influenced by both man and nature on the different species categories.

ECOSYSTEMS INVESTIGATIONS

SUMMARY STATEMENT

BACKGROUND To an ever greater extent the yield of marine fisheries is dependent upon water quality in coastal marine environments. Anadromous species of fish and shell-fish have traditionally been the first species affected by deteriorated coastal and estuarine waters. There seems little doubt, however, that the deterioration of coastal environments is having or will have an effect on coastal and offshore marine species which reproduce in or migrate through coastal and estuarine ecosystems.

In many instances the effects of deteriorated environments do not impinge directly upon finfish or shellfish which are important in the commercial and game fisheries; rather, polluted waters or physically disrupted environments may result in an elimination of or diminution in the standing crops of invertebrates important as forage species in marine food chains. Deteriorated environments may also disrupt the flora and fauna which play an important role in stabilizing marine sediments. Finally, invertebrate species, which are often attached or relatively immotile forms which cannot avoid polluted waters, are often excellent indicator organisms which can be used to detect or assess change in environmental quality.

OBJECTIVES

The Ecosystems Investigations program was developed to provide data for a comprehensive overview of the coastal and estuarine environments of the New York Bight and Long Island Sound. This program includes four investigations which are closely integrated. The Biological Oceanography Investigation was designed to provide data on base line distributions and life histories of benthic, natatory and planktonic invertebrates and their relationships to marine and estuarine finfish. The Marine Contaminants and Coastal Ecosystems Investigation is concerned principally with the effects of pollution and environmental deterioration on the living resources of coastal and estuarine ecosystems. It includes both field and laboratory studies of plants and animals as they are affected by Man's activities.

The Environmental Chemistry and Microbiology Investigation consists of several projects designed to quantitate the distribution and abundance of marine microorganisms and their effects on higher plants and animals. This investigation also has a principal role in providing data on the distribution and abundance of heavy metals and other known toxins within the physical environment and the tissues of living organisms.

The fourth investigation in Ecosystems Investigations is the Environmental Rehabilitation Investigation. Earlier research indicated that artificial reefs or habitats increase finfish standing crops and provide better sportfishing, at least at the site of the reefs.

It has been hypothesized that artificial habitats might be used to rehabilitate polluted or physically impoverished environments. We propose to study the effects of artificial habitats in Raritan Bay, a heavily polluted embayment adjunct to the New York Bight and once extremely productive of gamefish and commercial finfish and shellfish. We are using energy flow and biomass measurements as well as diversity indices as measures of increased standing crops, productivity and stability of reef communities when compared to unrehabilitated areas.

In the following paragraphs each investigation is described in greater detail and milestones are given. It is important to note that there are numerous interrelationships between each of these investigations. In addition there are data resulting from these investigations which are of immediate relevance to Resource Assessment, Pathology and other major components of the Middle Atlantic Coastal Fisheries Center.

ECOSYSTEMS INVESTIGATIONS

BIOLOGICAL OCEANOGRAPHY INVESTIGATION

BACKGROUND A rational management of marine fishes is dependent upon several factors which include a knowledge of the population dynamics of the fish in question, catch, quality of the aqueous environment and organisms which comprise the food chains which the fishes use for sustenance.

To measure water quality and develop an understanding of plants and invertebrate animals which form the food chains of marine fishes, it is necessary to make in-depth investigations of estuarine and coastal ecosystems. Despite the fact that the New York Bight and ancillary embayments constitute one of the most intensively used coastal environments in the world, little is known about the present status of living marine resources in these waters. The Biological Oceanography Investigation is designed to provide information concerning the more important forage and commercially valuable invertebrates and their environment within the Bight.

OBJECTIVES 1) Establish base lines in regard to the areas and temporal distribution of benthic, natatory and planktonic invertebrates. We are particularly interested in correlating areas and temporal (either seasonal or long-term fluctuations) distributions with observed physical conditions and man-induced changes. Are the successes and failures of finfish year-classes directly or indirectly related to the success and failures of invertebrate larvae and juveniles? We would like to answer this basic question through this research.

Geographically we are particularly interested in establishing these base lines within the Hudson Shelf Valley and Hudson Canyon. We are, however, interested in establishing base lines for invertebrate distributions at stations on the continental shelf, especially in areas designated or proposed for alternating dumping sites. Such sites have been proposed by EPA and U. S. Corps of Engineers. In addition, we wish to assess the invertebrate stocks present in embayments and estuaries such as Raritan Bay, Jamaica Bay, and Great South Bay. We also plan to continue our studies in the East River and Western Long Island Sound, waters directly connected to and affected by conditions in the New York Bight.

- 2) Determine the life histories and ecology of invertebrate species found to be of unusual importance in particular marine and estuarine food chains. Studies of most individual species or ecological groups, i.e., the meiofauna, would be done through contract or cooperative studies with academic organizations.
- 3) Establish the relationship between invertebrate species found in gut contents of finfish and invertebrate species diversity and standing crops in areas habituated by the finfish from which the gut contents were removed. This information will enable us to determine those species used as forage by finfishes and the action-reaction phenomena within particular marine ecosystems.
- 4) Obtain sufficient samples to furnish tissues and living animals to the Marine Contaminants Investigation for analyses for heavy metal and microbial contamination as well as experimental manipulation to determine the effects of lethal and sublethal doses of toxins and pathogens.
- 5) Develop, based on data collected from the New York Bight, hypotheses and predictive models which will be generally applicable to other temperate water ecosystems and which can be used in the management of living resources.

RECENT AND ONGOING RESEARCH

We have intensively sampled the area near the sites which are designated for receiving dredging spoils and sewage sludge wastes.

Stations were initially located 2.5 miles apart but once the approximate boundaries of the disposal grounds were determined sampling grids were established with stations placed as close as one-quarter nautical mile apart. Standard chemical, geological and physical measurements were obtained at selected stations on a regular schedule.

Standing crops and species distributions were determined for the benthic and zooplankton populations. Recently we have extended our program of intensive sampling to alternative dumping sites and the Hudson Shelf Valley and Canyon.

PROPOSED STUDY

We are presently developing a grid of sampling stations which will encompass the New York Bight and western Long Island Sound, including adjunct estuaries. Initially, grab, dredge and epibenthic

embayments and estuaries. Initially, grab, dredge and epibenthic samples will be taken at 2.5 mile intervals, except within the Hudson Shelf Valley, alternative dumping sites and embayments. At these sites samples will be taken at those intervals necessary to provide statistically or descriptively valid data. Subsequently, depending on continuity of sediment types and physical environment, samples will be taken at closer intervals throughout the grid system. When sufficient samples have been collected to allow us to describe the community structure of discrete benthic and zooplankton communities, correlations will be made with those chemical, geological and physical parameters which obtain within the confines of the communities.

At the same time, through contract or cooperative studies, individual taxa or ecological groups within the communities will be investigated by students or authorities on the particular taxa or group. These independent autecological studies will concentrate on explaining the observed fluctuations in abundance or variations in distribution of particular species.

All data will be entered into a standardized ADP system which will facilitate data retrieval and synoptic comparisons of biological communities and associated physical conditions. This system will also be developed so that it is compatible with the input from the other Center and Service investigations.

RELATION TO OTHER PROGRAMS All data collected by the Biological Oceanography Investigation will be done in a manner which facilitates comparison with data derived

by the other Investigations within MACFC.

In many cases data collected by the Biological Oceanography Investigation will be immediately furnished to the Marine Contaminants or Resource Assessment Investigations. Certain situations will call for cooperative cruises with personnel from other NMFS or NOAA facilities or academic institutions or organizations.

Inshore base line assessments will form a background for future environmental rehabilitation investigations.

RESEARCH PLANNED FY 1973. Continue sampling within Hudson Shelf Valley and Canyon, alternative dumping sites and embayments. Develop New

York Bight Shelf sampling grid and convert existing station nomenclature to standard system based on half degree squares. Initiate sampling at 2.5 mile intervals throughout the Bight. Process samples within laboratory and through contract.

FY 1974. Initiate sampling on grid at closer intervals as warranted by analyses of samples collected at 2.5-mile intervals.

FY 1975. Continue close interval sampling as indicated above. Begin computer analyses and construction of predictive models based on results to date; determine stress relationship of communities to sediment types and pollutants or man-induced changes.

ECOSYSTEMS INVESTIGATIONS

MARINE CONTAMINANTS AND COASTAL ECOSYSTEMS INVESTIGATION

BACKGROUND The New York Bight and ancillary embayments constitutes one of the most intensively used coastal environments in the world. Its uses include shipping, pleasure boating, waste disposal, and recreational and commercial fishing. In addition to these current uses, additional waste disposal activities, nuclear-electric power generation, and offshore mineral recovery are proposed for this coastal area.

Are all of these uses compatible in a single, multiple-use scheme? We cannot presently answer this question, particularly as long-term management of the living resources is concerned. To provide the data necessary to answer the foregoing question, we propose a continued program to:

1) investigate those invertebrate and algal species which have a commercial or recreational importance or play a significant role in coastal food chains, and 2) assess the impact of a variety of wastes on living marine resources in coastal and estuarine ecosystems.

Objectives of the investigation are to: establish the OBJECTIVES patterns of distribution of heavy metals, pesticides, petrochemicals and other toxic materials in sediments collected in the New York Bight and ancillary waters and embayments; determine patterns of distribution of bottom-dwelling macrofauna and meiofauna as these patterns are related to heavy metals and other toxins and deteriorated environments; determine the effects of a variety of contaminants on the physiology, behavior and reproduction of selected species of marine organisms indigenous to the New York Bight; continue our present investigation concerned with the sublethal and lethal effects of thermal additions and oil pollution on marine and estuarine species; and develop, based on laboratory and field data collected from the New York Bight, hypotheses and models which can be used to predict the effects of waste disposal in coastal and estuarine waters on the life history of selected species and which would be generally applicable to other temperate water ecosystems.

RECENT AND ONGOING RESEARCH

Field and laboratory studies concerned with the effects of ocean disposal of sewage sludges, dredging spoils, industrial

wastes and thermal additions have been carried on at the Sandy Hook Laboratory. At present this facility is continuing research on the effects of ocean dumping on the Hudson Shelf Valley; conducting base line studies on alternative dumping sites; and completing a comparative study of the effects of thermal additions on epibenthic animals and algal populations in Barnegat Bay and Long Island Sound.

PROPOSED

We are presently developing a grid of sampling stations which will encompass the New York Bight STUDY and western Long Island Sound, including adjunct embayments and estuaries. Initially, grab, dredge, trawl and epibenthic samples will be taken at five-mile intervals, except within the Hudson Shelf Valley, alternative dumping sites, and embayments. At these sites collections will be taken at intervals necessary to obtain samples which will provide statistically or descriptively valid data. Subsequently, depending on continuity of sediment and community types and physical environment, samples will be taken at closer intervals throughout the grid system. Data will be immediately processed through the Center ADP program to facilitate: real time reporting; the calculation of diversity indices, standing crops, and distribution; and immediate changes in experimental or sampling design as dictated by field observations.

RELATION TO OTHER PROGRAMS

All data collected by the Marine Contaminant Investigation will be processed and distributed in a manner which facilitates comparison with

data derived by other investigations. Samples will be processed so that biological materials are available for the Biological Oceanography Investigation as well as for the Marine Contaminants Investigation. Invertebrate and finfish tissues will be made available to analytical chemistry groups for analyses for heavy metals, pesticides and other wastes. Tissues removed from diseased organisms will be chemically fixed or otherwise preserved for examination by the Pathology Investigation.

Certain situations will call for cooperative cruises with personnel from other NMFS or NOAA facilities or academic institutions or organizations.

Inshore base line assessments and laboratory experimentation will form a background for future environmental rehabilitation investigations.

RESEARCH PLANNED FY 1973. Continue sampling in Bight to obtain data necessary to the development of overall offshore sampling program.

Initiate intensive sampling program within Raritan Bay and other embayments. Process field collection samples. Continue physiological experiments to determine effects of sludges, dredge spoils and other real wastes on enzymes systems and membrane transport; compare results of experiments with effects of known amounts of heavy metals, chlorinated hydrocarbons, and other contaminants.

FY 1974. Continue field collections. Begin preparation of: distribution maps, abundance and diversity catalogs, and correlations between distribution of organisms and contaminants.

FY 1975. Based on initial results of field observations and laboratory experimentation begin to develop predictive models.

RESOURCE ASSESSMENT INVESTIGATIONS

ICHTHYOPLANKTON INVESTIGATION

BACKGROUND Fishery scientists generally agree that shallow protected waters of bays, rivers and sounds along our eastern seaboard provide essential habitat for many marine fishes. The occurrence of juvenile fishes in estuaries has led biologists to conclude that a major role of the estuarine zone is its function as a sanctuary or nursery ground for young fishes, many of which are spawned at sea. In 1965, the Sandy Hook Laboratory began a study of the distribution of fish eggs, larvae and juveniles from the inner reaches of the littoral zone to the edge of the continental shelf to elucidate the degree of estuarine dependence of coastal fishes. Impetus for the study resulted from the accelerated degradation of the estuarine zone and a concurrent decline in some of our most valuable coastal fishery yields.

OBJECTIVES To study the early life histories of coastal fishes by: investigating the diel movements of larvae and their effects on both active and passive transport; gathering information on the vital statistics of larvae that will allow prediction of year class indices; maintaining surveillance of spawning areas of selected species to determine yearly fluctuations in abundance; relating offshore occurrence of larvae to distribution of juveniles.

RECENT AND ONGOING RESEARCH

The initial phase of the sampling program involved a survey of the continental shelf from Cape Cod, Mass., to Palm Beach, Fla.,

to locate areas of spawning, to estimate the seasonal duration of spawning and to trace the dispersal of young fishes spawned on the shelf. From the 1500 samples collected, we have information on the young stages of 60 species. With this base line data at hand, we are planning to initiate the second and more quantitative phase of sampling.

PROPOSED STUDY

We are presently developing a grid of sampling stations which will encompass the New York Bight and western Long Island Sound, including adjunct

embayments and estuaries. Initially, grab, dredge and epibenthic samples will be taken at 2.5 mile intervals, except within the Hudson Shelf Valley, alternative dumping sites and embayments. At these sites samples will be taken at those intervals necessary to provide statistically or descriptively valid data. Subsequently, depending on continuity of sediment types and physical environment, samples will be taken at closer intervals throughout the grid system. When sufficient samples have been collected to allow us to describe the community structure of discrete benthic and zooplankton communities, correlations will be made with those chemical, geological and physical parameters which obtain within the confines of the communities.

At the same time, through contract or cooperative studies, individual taxa or ecological groups within the communities will be investigated by students or authorities on the particular taxa or group. These independent autecological studies will concentrate on explaining the observed fluctuations in abundance or variations in distribution of particular species.

All data will be entered into a standardized ADP system which will facilitate data retrieval and synoptic comparisons of biological communities and associated physical conditions. This system will also be developed so that it is compatible with the input from the other Center and Service investigations.

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ECOSYSTEMS INVESTIGATIONS

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FY 1975. Based on initial results of field observations and laboratory experimentation begin to develop predictive models.

ECOSYSTEMS INVESTIGATIONS

ENVIRONMENTAL CHEMISTRY AND MICROBIOLOGY INVESTIGATION

Man's activities can change the coastal waters BACKGROUND and thereby have a diverse effect on living marine resources. Increased discharges of organic nutrients and chemicals into the aquatic environment as well as elevation of water temperature, can modify the marine biota both quantitatively and qualitatively. Of immediate concern is to determine the degree to which microbial populations and chemical pollutants can cause diseases in living marine resources as will the utilization of the resource by man. Contamination by fecal pollution as well as by certain species of bacteria belonging to the genus Clostridium, Vibrio, Pasteurella and Gaffkya, as examples, have recently been identified in regards to diseases of the fishery resources and in man by the utilization of the contaminated fisheries. In Japan, mercury poisoning in man has occurred through the fisheries as a direct consequence of chemical contamination of the fisheries environment. Recently, in the United States, elevated levels of mercury and pesticides have been demonstrated to occur in certain fish species with resulting regulatory restrictions on their utilization. Whether additional heretofore unknown and undetected microbial species and chemicals (compounds) exist in the aquatic environments warrants immediate attention.

OBJECTIVES

The objectives of the microbial aspect of this investigation is to determine, both qualitatively and quantitatively, the microbial flora (both aerobic and anaerobic) of inshore waters, sediments and organisms. The objective of the chemistry aspect of this investigation is to determine the level of chemical contamination, heavy metals, pesticides, etc., in the same elements of the ecosystems. The prime objectives of these two studies are to determine whether changes in the indigenous microbial flora and increased levels of chemical pollutants adversely affect the food chain organisms, the fishery resource and the utilization of the resource. Basic to the attainment of the objectives is the development of sensitive and specific methodology for the identification of marine microbes and chemical pollutants.

RECENT AND ONGOING RESEARCH

Both the microbial and chemical investigation are outgrowths of programs initiated by the former Ann Arbor Technology

Laboratory several years ago. With the recent organization of the Middle Atlantic Coastal Fisheries Center, the microbial aspects of fin rot disease and the red tide study are included in the current investigation.

PROPOSED STUDY

The initial phase of the study will be a quantitative, or at least qualitative determination of the more prominent bacterial flora (aerobic and anaerobic) of

water, sediments and organisms from Middle Atlantic Coastal areas utilizing established enumeration and enrichment methods. Recognizing the limitation on methodology in regards to the cultural growth of marine bacteria, various cultural techniques including in situ methods, specialized anaerobic methods, serological and immunological techniques will be evaluated and applied for isolation and demonstration of fastidious organisms. Because of current interest relative to pollution problems in the marine environment, the initial phases of these studies will be directed towards organisms which are indicators and consequence of fecal pollution, as well as those belonging to the genus Clostridia and Vibrio. As studies progress, additional procedures for additional bacterial groups will be included. The isolation of the predominant bacteria and characterization studies relative to their active role in the environment will follow. These involve physiological studies on the isolates' metabolic responses to physical and chemical factors present in the environment and pathogenicity towards fish, shellfish, and food chain organisms. The classification of the bacterial isolates will be by the determination of specific physiological characteristics, DNA base ratios and homology, cellular fine structure, serological identification and phage typing. The utilization of in situ methods to compare experimental results directly with bacterial activity in the environment such as growth rates, metabolic activity, ectosymbiosis, cellular morphology and population cycling will be a phase of the proposed study. Experimental laboratory systems that measure the direct effect of pollutants on a bacterial population and bacterial degradation and cycling of metals, chemicals, pesticides, and hydrocarbons will be developed for a better understanding of the role of bacteria in this regard. In addition, other laboratory systems will be utilized to study the effects of pollutants on the resistance and immune response of certain fish species to select fish pathogens. The primary effort of the environmental chemistry group will be to determine base line levels of chemical contaminants in sediments, plankton, fish and invertebrates from the marine environment. Continuation of past work on the establishment of base line data on heavy metals will be the prime effort for the near

future. The main geographical areas of sampling on established grids for this work will continue to be the New York Bight, Long Island Sound and selected North Atlantic areas for comparison to the other two areas. In addition, other Atlantic Coastal areas, i.e., Chesapeake Bay, will be programmed in for additional comparisons. Establishment of capabilities to conduct analyses of pesticide type compounds will be initiated in order to begin accumulation of base line data in the marine environment. Investigation of other chemical contaminants will be undertaken as information is accumulated by other agencies relative to the types of chemicals that are being (or have been) dumped into the marine environment. Investigations on the effects which result from the exposure of marine organisms to metals will be started in the near future. The determination of the amount of uptake and cycling of specific metals by laboratory experimental animals and microorganisms is an important role for chemical analysis and will become a significant part of the chemistry effort when fully implemented.

RELATION TO OTHER PROGRAMS

All data obtained by the Environmental Chemistry and Microbiology Investigation will be made available on a timely basis to State and Federal agencies who have the responsibilities in regards to resource contamination and public health hazards. Many situations in environmental chemistry and microbiology are related to other Center investigations, i.e., water quality, experimental biology (bioassay) and pathology and resource assessment. Some situations call for cooperative research. Examples of proposed cooperative research are the synergetic relationships between chemical contamination and pathogenic bacteria which cause fish disease and toxicity of chemicals to fish, shellfish and molluscs.

FY 1973. Evaluation of chemical (element and

RESEARCH PLANNED

chemical form) and microbial methodology. Preparation of analytic reagents, i.e., antisera, chemical standards. Surveys for the enumeration, isolation of bacterial types and the distribution of heavy metals in elements of the ecosystem in the North Atlantic and mid-Atlantic coastal areas. Initiate characterization of bacterial isolates by chemical serological and animal assay methods, with emphasis on target species (predominant types isolated and Clostridia (toxic) and Vibrio species). Emphasis on heavy metal contamination in target marine animals, plankton and invertebrates. Continuation of bioassay on marine animals in reference to uptake, distribution of the specific metals, and the immune response as affected by the metals in the animals. Initiate in-house capability of analysis for pesticides. Initiate systematic surveillance for fin rot disease and red tide blooms in affected areas, and study microbial etiology and water quality parameters related to these phenomena.

FY 1974. Continuation of above, with more concentrated effort and extension to other microbial groups on taxonomy and serological relationships. Continuation of base line data for heavy metals and intensification of base line data for pesticides. Begin investigations on other chemical contaminants such as PCB's and petroleum waste products. Examine the immune response in marine animals during chronic exposure to specific heavy elements and PCB's. Extend antigenic studies to other bacterial species, and develop radioimmunoassay techniques for antibody detection in marine animals. Initiate in situ and laboratory setups for studies on bacterial population growth and conversion and transforming mechanisms.

FY 1975. Continue survey work both chemical and microbial, in areas which have been determined by previous studies to be a primary interest. Continue characterization of microorganisms of particular interest by techniques developed by previous studies. Initiate sampling of the biota for other chemicals which have shown from other studies to be an environmental hazard. Initiate field work on the control of algae blooms. Evaluation of results obtained in relationship to other investigations and for future research.

ECOSYSTEMS INVESTIGATIONS

ENVIRONMENTAL REHABILITATION INVESTIGATION

BACKGROUND Raritan and Sandy Hook Bays represent two heavily polluted marine water bodies in the New York-New Jersey region. Although shipping, waste disposal, and limited recreation constitute the primary use of these waters, no effort has been made to improve them for fish habitat. In order to "rehabilitate" these embayments for sport and commercial fishermen the following base line study is proposed in which miniature artificial reefs, constructed of tires, will be evaluated as a potential tool for increasing finfish productivity. Experimental reef sites are planned for Raritan Bay in the vicinity of Great Kills Harbor. According to EPA officials, over 2 million gallons of primary treated sewage is discharged into the waters north of the test area daily. Although finfish reside in Raritan Bay the effect of reefs on the finfish abundance and distribution is not known.

1) To determine the spatial effect of reef distribution OBJECTIVES on finfish abundance. Information on fish inhabiting reef sites throughout the months of April-November is of special interest since it will provide answers to questions concerning seasonal fish mobility and reef interaction during the prime fishing months. 2) To compare secondary productivity in reef and non-reef areas of Raritan Bay. Invertebrates and algae utilized as food for finfish is of particular interest since their seasonal settlement and abundance influence the resident finfish population. Using a bioenergetic approach, details on the rate of utilization by fish will be obtained through laboratory experiments and the effect of predation on the reef communities will be examined in the field. 3) To examine the effect of an experimental reef on the behavior of finfish. Diving observations indicate that finfish utilize a reef for "cover" in addition to obtaining food. Coordination of field and laboratory experiments together with a record of the physical parameters will elucidate the importance of this phenomena relative to reef productivity.

RECENT AND ONGOING RESEARCH

In 1967, the Sandy Hook Laboratory began investigating the development of artificial habitats in relatively unpolluted waters from

Gloucester, Massachusetts to St. Thomas, Virgin Islands. The studies have been principally concerned with measuring increases in the standing crops of epibenthic forage organisms and finfish predators associated with artificial habitats.

Recently a grid of sampling stations have been developed in the highly deteriorated Raritan-Sandy Hook Bay complex. Core and dredge samples are being collected to provide base line data on sediments and benthic standing crops.

PROPOSED STUDY

Small artificial reefs, made of tires bound together into units and anchored to wooden pallets, will be randomly placed on the sampling stations, and monitored for finfish activity at monthly intervals. In addition, invertebrate and algal food species will be collected and measured, either directly from the tire units or rubber test panels. The sampling design will include comparisons within and between habitat types.

The bioenergetic approach will be adopted in examining reef productivity. In addition to their growth and settling rates, the caloric value of the important food species will be determined.

Certain aspects of the study, such as identification and sorting of epibenthic samples and morphometric analysis of finfish, will, in part, be completed through contract or cooperative studies.

RELATION TO OTHER PROGRAMS

The data collected will be used by the Marine Contaminants and Resource Assessment Investigations. The information on the settling,

colonization and survival of epibenthic organisms will be used in ongoing research concerned with epibenthic communities along the Atlantic seaboard.

RESEARCH PLANNED

FY 1973. Develop Raritan-Sandy Hook Bay sampling stations. Initiate sampling at these stations. Process samples

within the laboratory and through contract.

FY 1974. Experiment with new artificial reef designs, increasing vertical displacement.

FY 1975. Develop energy flow systems on artificial reefs.

EXPERIMENTAL BIOLOGY INVESTIGATIONS

SUMMARY STATEMENT

BACKGROUND There exists, at all levels of both the public and the private sectors, considerable alarm that the living marine resources of the estuarine, coastal and offshore waters of the Middle Atlantic Bight are being adversely affected by extensive offshore dumping of untreated wastes and by run-offs of highly polluted waters. The mode and intensity of such adverse physiological effects is largely unknown. Base line findings of marine environmental quality cannot be interpreted without such knowledge nor can rational water quality standards be established or enforced when such knowledge is lacking. Quantitative, controlled exposure experiments on living organisms, both static and chronic, and involving all stages in their life histories, followed by a battery of tests are necessary to permit evaluation, standards-development, successful enforcement, and resource conservation. The nucleus of such a research team, expert in hatching, rearing and algology and in the physiology, genetics, pathology and chemistry of living marine organisms became available through the termination of a molluscan aquaculture program at Milford Laboratory. Should the aquaculture program be reactivated, these groups could, with a relative small increment in personnel, service both programs.

OBJECTIVES

- 1. Determine lethal effects of a large variety of known pollutants on the larval stages of molluscs, crustaceans, and finfish.
- 2. Determine lethal effects of a large variety of known pollutants on the juvenile stages of molluscs, crustaceans, and finfish.
- 3. Determine the long-term sub-acute effects of exposure to a large variety of known pollutants on the larval, juvenile and adult stages of molluscs, crustaceans and shellfish.

- 4. Define the physiological and biochemical pathways affected and relate them to the metabolic disorders, tissue abnormalities, etc., which result in death or permanent damage to the living marine organisms.
- 5. Determine effects of marine pollutants on the chromosomes, and genetic development, of the American oyster, C. virginica, initially, and on other fish and shellfish. Evaluate findings in terms of specific pollutants and of population genetics.
- Determine effects of pollutants on marine phytoplankton, on benthic fauna, etc. - as these form the base of food chain for all other living organisms.

RECENT AND ONGOING RESEARCH

This program was initiated in fiscal 1972. No formal organized research in this area had previously been conducted. During the

year, standard rearing procedures for oysters, surf clams, sea scallops, and several species of crustaceans and finfish have been initiated with considerable success. Standardized procedures for acute (static) testing of larvae have been developed, tested and, in the case of molluscan larvae, yielded information on the LD50's of some 8 to 10 heavy metals.

PROPOSED STUDIES

evaluation.

Standardized procedures for subacute, long-term testing of larvae, juvenile and adult molluscs, crustaceans, and finfish will be undertaken in fiscal '73 upon completion of pollution abatement systems now in planning stages. Concurrent studies on pollutant effects on physiology, enzymology, osmotic regulations, genetics, algology and pathology will continue. Indicator organisms, those deemed most responsive to pollutants, most exposed to pollutants in the natural environment and those most representative of Middle Atlantic Bight resources, will be selected for intensive testing and

RESEARCH FY 1973. Continue static testing of heavy metals; PLANNED extend testing to include crustaceans and finfish. Design and install pollution abatement system and initiate long-term subacute bioassay studies on larvae of oysters, clams and sea scallops. Continue induced spawning studies on crustaceans and finfish. Develop techniques for delivery of live juvenile and adult specimens to the laboratory.

FY 1974. Initiate static testing of other pollutants, DDT, BCB's, etc.) and of mixtures of pollutants (heavy metals and pesticides, etc.). Initiate long-term subacute bioassay studies on larvae of crustaceans. Initiate longterm subacute bioassay studies on juvenile and adults of molluscs, finfish, and crustaceans. Assemble and interpret fiscal '73 findings. Consolidate with interpretations of cooperating research groups (physiology, chemistry, etc.).

FY 1975. Complete static test studies. Continue long-term subacute studies on larval. juvenile and adult forms on selected indicator organisms (molluscs, crustaceans and finfish). Expand studies to include mixtures of pollutants. Assemble and interpret fiscal '73 and '74 findings. Consolidate with interpretations of cooperating research groups (physiology, chemistry, genetics, etc.). Prepare monograph on consolidated findings.

EXPERIMENTAL BIOLOGY INVESTIGATIONS

GENETICS INVESTIGATION

BACKGROUND Part of the process of bringing the oyster - and any other candidates for intensive mariculture under artificial conditions - under some greater measure of control necessitates information on the species' genetic and breeding system. This is particularly so for profitable hatchery production and for the best pond culture. Knowledge of oyster genetics would also be of value for stocking of decimated beds or for the introduction of stock to previously uncultivated beds in the wild.

The alternative to research is trial and error breeding, which has its place and can achieve results, as evidenced by man's domestication of animals starting in pre-historic times. It is hard to suppose, however, in this age of science and technology, that a sound aquacultural industry will develop in competition with other foods and the natural fisheries and with other uses of water resources without employing some of the genetic know-how now being resorted to as a matter of course in breeding just about all our farm animals and crop species.

The idea of profitable and continued application of scientific information to any industry which will become increasingly dependent on mancontrolled artificial, as opposed to natural cultivation without an expansion of the general base of knowledge about the species, is wishful thinking. This is especially so for fish which, in terms of our knowledge of their genetics, lag far behind our other food species.

OBJECTIVES This genetic program is so geared to provide information on the basic genetic system of the oyster - and on other marine species as the opportunity and need arise - as a basis for development of increasingly strong, sound applied programs. It is to set up and conduct experimental breeding programs devised to provide the kind of information a commercial, large-scale breeder needs to make nowledgeable decisions about conducting his own operations. Small amounts of starter seed should be provided industrywhen obtaining this requires experimental expertise beyond that of commercial breeders now operating or likely to operate in the near future - stocks unobtainable via the common methods of mass, individual or family selection.

Commercial breeders should be stimulated and guided into sensible applications of the scientific information provided them, and helped to avoid the genetic pitfalls any breeder can lapse into. Areas of genetic information should be developed that will make possible an increase in the natural range of the oyster or enable it to survive and reproduce in conditions under which the natural wild type could not survive. Genetic information gained through research should be used to advise on restocking of old and decimated natural populations and on the introduction to new areas of special genetic stocks already tested under pilot field conditions. Scientific and semi-popular publications should make clear such possibilities, as well as the natural limitations of introductions. Another objective is to monitor any probable future necessity for active protection of the valuable gene pools which still exist in different wild marine populations. Wild populations tend to be neglected and then lost as a species becomes intensively farmed (for example, the United Nations-recognized crisis which now exists in respect to cultivated grain crops as genes of great value to breeders are lost to man for good as the last traces of the wild ancestral species of these crops disappear). Protection of such populations would consist of traditional conservation - but now for genetic purposes - the establishment of special stock collections and reserves; and in research on the cryopreservation of the gametes of marine forms. A final objective is to appraise the genetic and reproductive damage resulting from the types of marine contamination which can be mutagenic and carcinogenic to wild populations.

RECENT AND ONGOING RESEARCH

Inbreeding Crassostrea virginica, the American Oyster. Inbreeding to some extent will accompany mass selection in

commercial hatcheries. More extreme forms will eventually be used to develop lines for subsequent hybridization in hopes of so obtaining hybrid vigor.

Commercial shellfish hatcheries seem prone to start selection programs with too few oysters. Because the spawn of a single, excellent cross might fill even a good-sized commercial hatchery, the problem is accentuated.

Inbreeding lines of oysters were initiated at Milford to determine what form inbreeding depression would take in the oyster, and to determine the extent of this depression per coefficient of inbreeding.

Selective Breeding of C. virginica. Quantitative, commercially important traits are controlled predominantly by exceedingly large numbers of either of two types of genes. It is the additive type of gene that responds to molding and change by selective breeding. The heritability of a trait is a measure of this additive genetic variance.

A large-scale selection experiment for growth rate of <u>C</u>. <u>virginica</u> in the adult form was set up from the spawn of 835 oysters (1) to estimate heritability over a number of generations for both faster and slower growth, (2) for a particular coefficient of inbreeding, and (3) to determine the ultimate extent of response to selection for growth.

Hybridization of <u>C. virginica</u>. Plants and animals are artificially crossbred or hybridized in order to combine in the offspring some of the desirable characteristics displayed by either set of parents. Another purpose of crossbreeding or hybridizing is to utilize the effects of hybrid vigor. Hybridization for the sake of combining the desirable genes of two different types sometimes takes the form of upgrading practiced when the over-all performance of the import is better than the local, but when the local is superior in some particularly important traits related to local environmental conditions. Another way of combining the characteristics of two types is the introgression of special desirable foreign genes into the local stocks.

Old, well-established subspecies usually have gene combinations so well adapted to the environments they occupy that any new gene combinations created by hybridization will nearly always be less favorable. If, however, hybridization takes place in an unstable environment, some of the vast array of segregates appearing in later generations will very likely be better adapted to the new field environment than any individual of the parental group. They could also be better adapted to the artificial environment of a commercial hatchery or experimental shellfish laboratory.

More specifically, hybridization of <u>C. virginica</u> might supply a less sensitive, more vigorous larval form better able to hold up to the vicissitudes of the hatchery. Hybrids could furnish a diversification of the U.S. East Coast oyster crop. Diversification can break the wild-fire spread of a disease by the inter-dispersal of resistant types. Were more known about the genetics of different geographic populations of the American oyster, some of the fear might be diminished of experimental transplantation of oyster set to commercial beds badly in need of seed from areas where it occurs in great abundance and goes to waste.

The American oyster occurs extensively over an unusually wide range of temperature, from cold to subtropical. Wild <u>C. virginica</u> sampled from approximately twenty-five different sites from Prince Edward Island, Canada to Greenwich, Connecticut, are being test-hybridized.

Environmental Mutagenesis Using the Oyster as an Assay Species. Not to be overlooked in studies on the effects of pollutants on aquatic organisms is the genetic damage which can be caused by sublethal levels of a pollutant. By now very large numbers of reports have shown terrestrial plants and animals to be genetically affected - damaged in terms of the future recruitment of normal healthy fecund specimens - by any number of environmental pollutants at sublethal concentrations. Since most mutagens are also carcinogens, there is the additional hazard that genetically damaging levels of a pollutant will lead to an increased serious or at least nuisance-level incidence of tumors in natural or cultivated populations. This could have serious marketing implications for commercial species.

In contrast to the population, species, and generic constancy of the chromosomes of the oyster, early developmental stages of <u>C. virginica</u> are marked by a high frequency of variation in chromosome number. It appears that the sensitivity of this oyster egg to all kinds of environmental distress results in acute effects at the chromosome level. This makes the oyster an excellent assay species for detecting genetically damaging and zygote-destructive pollutants.

In conjunction with the Marine Contaminants Investigation at Milford, the effects of several heavy metals on the cytogenetics of fertilization, meiosis and cleavage of the oyster are being determined.

The basic cytogenetics of the oyster has already been thoroughly studied, so mutagen-induced deviations should be readily recognized. A large amount of experience has further been accumulated now with oyster cytogenetics in a variety of experimental situations.

In an extensive study already completed the mutagenic effects of ionizing radiation on <u>C. virginica</u> were examined (results presented at the 1972 meeting of the Radiation Research Society, and being written for publication). This work on ionizing radiation in addition provides a background of expertise on which to base studies of chemical marine contaminants which are potential mutagens and carcinogens.

PROPOSED STUDY

Several inbreeding lines must be carried because of the naturally high percentage loss of many lines in the inbreeding process. The test crossing of

inbred lines for superior hybrids - as in maize - requires large numbers of lines. To be able to continue giving breeders - including those abroad - advice on inbreeding necessitates continued work on a larger scale, and on a variety of species. Highly inbred lines should reveal a variety of homozygous recessive mutants which would serve as marker genes in more basic genetic studies. Such work should be continued and expanded to the level of operation possible in an experimental-scale hatchery.

The selection experiment described above under Recent and Ongoing Research should be continued to provide estimates of prevailing heritability under intensive selection over a number of oyster generations, and to determine how far the oyster can be pushed in the direction of rapid growth. If the experiment can be kept going on a large enough scale, it should provide seed stock for other projects.

Additional selection experiments should also be set up to estimate heritability for other characters. From separate estimates, selection indices which weigh the commercial value of selection for different characters in combination can eventually be prepared.

Geographic hybridization studies should be continued utilizing oysters from all the areas from which they have already been collected and hybridizing oysters from a still wider range. Work should be expanded to a scale where enough seed can be produced for field and laboratory testing under a variety of conditions. Seed stock carefully earmarked for its characteristics would then be available for other purposes.

Concomitant with these hybridizations should be experimental studies aimed at means of overcoming gamete incompatibility factors which prevent or make near impossible many potentially useful inter-species crosses of oysters. Also, there should be development of methods for complete inhibition of oyster sperm in mass crosses so that useful hybrids between two stocks could be made commercially by way of large mass spawnings. Now this must be done by individual crosses, a method too costly and too limiting genetically to be of much commercial use under intensive aquacultural systems.

The effects of heavy metals on the cytogenetics of fertilization, meiosis and cleavage of the oyster (and other choice species) ought to be made a regular joint effort of the Contaminants and Genetics Investigations.

RELATION TO OTHER PROGRAMS As stated already in a formal publication, genetic studies as outlined here must go hand-in-hand with nutritional studies,

better control over the animal's life cycle and increasing attention to the care and preservation of wild stocks if aquaculture is to become a really viable industry and contribute in a truly significant way to the world food supply. These investigations have a bearing on, and can be benefited by proposed Immunogenetic Investigations, by the NMFS Shrimp Culture work in Galveston, and by a variety of genetic studies beginning in the universities under support of NOAA's Sea Grant Program. While the universities importantly have readily available a wide cross-section of people from different departments, a laboratory as Milford can offer the program continuity so important in breeding, stock collections, and conservation of wild gene pools, and the laboratory was designed for just such functions in an over-all aquacultural-directed research effort.

RESEARCH PLANNED FY 1973. Initiate new inbreeding lines of oysters.

Start carrying hatchery-size oyster
cultures in pilot hatchery at Milford

Laboratory. Study cross-incompatibility genes in the oyster. Collect spat from large cultures.

Use X-irradiation to induce parthenogenesis for pure absolute oyster inbreds in one generation.

Continue present oyster selection experiment, but expand its size. Start new selection experiments for different characters. Start setting up selection experiments on other commercial species and on species especially favorable to laboratory analyses.

Expand geographic hybridization studies on the American oyster to include more individual crosses from any single population, and to include a wider diversity of wild populations.

Start work on methods for sperm inhibition for large mass hybridizations of the sort necessary for commercial hybridizing of shellfish.

Explore means for and start to make on large scale inter-species of oysters and maybe even intergeneric hybridizations.

Continue work on cryopreservation of live male and female gametes.

Expand studies on genetic and congenital efforts of marine contaminants on commercial and on ecologically important species.

FY 1974. Measure depression in oyster inbreeding lines. Inbreed next generation. Evaluate performance of absolutely inbred spat obtained via X-ray induced parthenogenesis. Use data to advise breeders on inbreeding tolerances. Test-hybridize inbred lines, report results to industry, and advise on commercial use of such a breeding program. Distribute seed if this is advisable and if there is sufficient supply.

Continue selection experiments and report on the durability of the selection response for different characters.

Continue all phases of the hybridization work. Start backcrossing interspecies hybrids to local types to introgress desirable genes from the foreign into the local types. Mass select each backcross generation. Use laboratory stock for seeding new oyster beds where this is desired, or to repopulate old wild beds with a more vigorous dependable stock for wild. (This is, of course, after field replications). Start field evaluations and field studies.

Continue work on genetic effects of marine contaminants expanding it to include radiation, and to encompass marine species other than shellfish.

FY 1975. Expand a little more all phases of above studies. At the end of the year hold a National Workshop on commercial breeding techniques. Expect that industry will begin much of their own breeding, using results obtained via such research on which to base their own programs.

Use results obtained from hybridization experiments to advise on further stocking of new and old beds.

Begin genetic research on new marine species of commercial value. Start work on genetic studies on marine algae from the standpoint of food values of some and on others in consideration of their importance in the food chain.

Use accomplishments on cryopreservation of gametes to establish at Milford or somewhere a stock collection of genetically important germ plasm.

Do a survey on the world's marine fish populations of value for future breeding use; determine the likelihood of such populations being lost; sponsor the preservation of portions of such populations; use gamete preservation as an adjunct when this is suitable and/or possible.

Hold an International Symposium on Marine Genetics at Milford with the cooperation of an organization such as FAO. Use the symposium as a basis for starting a multi-author comprehensive book on fish genetics as of this date.

EXPERIMENTAL BIOLOGY INVESTIGATIONS

ALGOLOGY INVESTIGATION

BACKGROUND Population pressures on land food resources and demands for more varied diets indicate an increasing need for sea food and other products that originate in the marine environment. An expanding technology resulting in widespread pollution threatens naturally productive areas, particularly coastal zones where there is a concentration of population and industry. The availability of information on 'Protist' organisms that are at the base of the marine food chain is a necessary prelude to protecting and managing these environments, and to developing new areas of productivity, both from natural sources and artificial culture. Detrimental effects on edible commercial species result from loss of useful food chain organisms through effects of contaminants or adverse environmental conditions. Harmful effects to the fisheries also occur from concentration by the food organisms of contaminants toxic to the ingesting species, or from the presence of toxic 'Protists' in the food chain.

Information is available on laboratory culture of some species of microalgae but others have not been successfully cultured although this is a necessary procedure for feeding marine metazoans reared or held in the laboratory for experimental purposes.

Objectives of this program are to carry out research that will lead to a fuller understanding of the responses to the natural and unnatural environment of food chain microorganisms by seeking information that will elucidate basic biological principles. Experiments in the laboratory are to be carried out under controlled conditions and wherever possible related to natural situations. In addition to experimental investigations, objectives are to maintain and enlarge upon a culture collection of unicellular marine micro-algae and to maintain a service for the production of algal foods by mass culture techniques while continuing to explore ways of producing them.

RECENT AND ONGOING RESEARCH

A stock culture collection of representative species of unicellular marine algae consisting of 87 strains isolated locally, as

well as in other parts of the world, is being maintained under standardized culture conditions in various types of media. All but about 15 strains are in axenic culture; work is in progress to purify the remainder. The collection serves as a center for the distribution of starter cultures to other laboratories - government, industry and university. Methods have been developed for different types of mass culture to fill particular needs, such as continuous cultures, semi-continuous cultures, deep tank cultures, and outdoor cultures. Recent innovations in technique have improved culture yields, as well as quality, that are being provided as a service to other laboratory investigations. Nutritional investigations of 14 algal species that center about utilization of organic carbon sources in the light and dark are in progress. The aim is to investigate this as an aspect of chemical pollution, and of the biochemical events in the nutrition of photosynthetic species.

PROPOSED STUDY

Research investigations will collect data on the growth requirements of marine phytoplankton, such as minerals, vitamins, growth factors, temperature, light, pH,

salinity, and other allied factors; investigate the effects of marine contaminants and metabolic inhibitors on reproduction, survival and species stability, enzyme systems and growth kinetics. This information will be utilized in the design and maintenance of apparatus for the production of large quantities of phytoplankton cultures.

RELATION TO OTHER PROGRAMS The work of this investigation is one on which all laboratory rearing and developmental work with metazoan species is dependent. The food

production aspect of this investigation is a year-round responsibility for making non-toxic algal foods available to the laboratory in large quantities. Other programs investigating the effects of marine contaminants, both in the field and in the laboratory, must take into consideration effects on food organisms. An understanding of food specificity, utilization, and toxicity is fundamental to all mariculture-oriented programs. Numerous opportunities are available for cooperative investigations both in the laboratory and the field.

RESEARCH PLANNED FY 1973. Continue maintenance of stock culture collection, purify strains, and make preliminary identifications of unclassified

species; continue operation of mass culture unit as a laboratory service; if equipment becomes available, find techniques for harvesting, storing and preserving large quantities of phytoplankton foods; complete studies on methods of studying food utilization in larvae; complete nutritional investigations of carbon utilization and enzymatic capabilities in phytoplankton; continue studies on salinity adaptation in a cryptomonad flagellate.

FY 1974. Continue maintenance of stock culture collection and expand collection by collecting species from local waters; continue operation of mass culture apparatus as a service, enlarging culture capacity if orientation of laboratory to mariculture takes place; develop facilities for studying food preferences and uptake by marine species not previously studied; study comparative effects of nutrition of phytoplankton on cell composition and growth kinetics in the presence and absence of inhibitors.

FY 1975. Continue stock culture and mass culture service; investigate production of extracellular substances by phytoplankton and their effect on other species; study cytological detail and ultrastructure of species significant in the food chain.

EXPERIMENTAL BIOLOGY INVESTIGATIONS

LIFE HISTORY/WATER QUALITY INVESTIGATION

The continual introduction of pollutants into the BACKGROUND marine environment has become a problem of major concern to marine biologists and others who recognize the vulnerability of marine communities to the effects of these contaminants. Within a species, the stages of development most sensitive to their environment are usually the embryonic and larval; consequently, it is here that the most serious and far-reaching damage occurs, not only to the young organisms directly but also to the continuity of the adult populations which they represent.

The objectives of the investigation are (1) to deter-OBJECTIVES mine the reproductive habits and early development of ecologically and/or commercially important species in the Middle Atlantic Bight about which little is known, and (2) to create in the laboratory environments in which these species can be spawned and their embryos, larvae and post-set stages reared in good health. These techniques will then be made available to the Marine Contaminants Investigation for use as bioassay tools.

RECENT AND ONGOING RESEARCH

Some aspects of gametogenesis, spawning and early development in certain coastal bivalves have been investigated in the recent past but no definitive study has been made of any bivalve species from this environment. At present such studies are under way with the sea scallop, the surf clam and the ocean quahog. Methods for manipulating gametogenesis and inducing spawning out of season in the laboratory are being developed.

Some progress has been made in the recent past in identifying and controlling bacterial pathogens that cause disease in bivalve larvae. This study is continuing. Various methods for altering the composition of natural sea water to eliminate pollutants, both natural and man-made, are being investigated. These include filtration, ultraviolet irradiation, ozonation and activated charcoal adsorption.

PROPOSED STUDY

Because of their long-standing commercial value, much is known of the spawning mechanisms of certain estuarine bivalves and the physiological

requirements of their early developmental stages; consequently, the young of these species are already being used as bioassay organisms in the Center's study of marine contamination. It is logical that this investigation start with a study of the more-recently exploited coastal species, such as the sea scallop, the surf clam and the ocean quahog. Initially, the applicability of standard culture methods for estuarine bivalves to the ripening, spawning and culture of the three offshore species will be determined. It is probable that some of the standard methods can be used directly but because of the significantly different environment in which the coastal species live, it is likely that new techniques for manipulating certain aspects of their reproduction and the culture of their young will have to be developed.

The reproductive biology and early development of other classes of marine organisms which have different physiological requirements and which occupy different ecological niches in the Middle Atlantic Bight than do the bivalves will also be studied. Major differences in habitat needs, nutritional requirements and reproductive mechanics that exist between dissimilar groups of organisms will require much original research to develop precise culture methods for all the species that it will be deemed necessary to rear.

Studies of water quality and the effects of pollutants in the water on the well-being of all classes of organisms being reared must accompany the life-history studies if dependable culture methods are to be developed. The susceptibility of all species selected for study to disease in the admittedly unnatural environment of the laboratory and control of diseases that develop will also be investigated.

RELATION TO OTHER PROGRAMS The work of this investigation will be vital to a complete assessment of the effects of marine contaminants on all stages in the life cycle of

important species in the Middle Atlantic Bight by providing dependable methods for culturing the developmental stages in the laboratory. These methods will be available not only to our own Center Investigations but to other NMFS Centers and to the Environmental Protection Agency.

Although not intended as such, the work of this investigation may contribute to the progress of mariculture in this country by providing improved culture methods for the larvae of marine animals and new insights into the life histories of little known species.

For a complete understanding of the commercial stocks which they are studying, population dynamicists must know how to fit the reproductive mechanisms and the early development of the species into plans for managing the fisheries. This investigation may provide such information for certain fisheries, such as that for the ocean quahog.

RESEARCH PLANNED FY 1973. Continue studies into the early life histories of the sea scallop, the surf clam and the ocean quahog.

Continue to develop dependable methods for ripening and spawning the adults of these three species and for rearing their young stages through metamorphosis.

Initiate a study of the habits, reproductive cycle and early development of a representative crustacean genus, such as <u>Cancer</u>, which has species in both estuarine and offshore environments.

Continue the study of water quality in the laboratory, particularly that of the incoming and resident sea water and its effects on the survival and growth of the embryos and larvae of marine bivalves with emphasis on the role of ozone in altering the composition of natural sea water.

Initiate a study of contaminants in the laboratory sea water effluent and their control to eliminate the possibility of re-contaminating the laboratory intake water.

Continue the study of diseases of embryos and larvae of marine bivalves with an emphasis on the control of those microorganisms already known to be pathogenic to the early developmental stages.

FY 1974. Continue to refine culture methods for offshore bivalves and make the methods available to the Contaminants Investigation as they become standardized. Continue the study of the Cancer crabs stressing the development of of controlled spawning and rearing techniques in the laboratory. Compare survival and growth of the crustacean larvae in natural and artificial sea water.

Continue the study of contaminants in the laboratory sea water effluent. Expand water quality studies to include the effects of ozone on the young of representative crustaceans. Expand the study of diseases of bivalve embryos and larvae to include diseases of post-set stages. Initiate a study of potential diseases in early stages of crustaceans.

FY 1975. Continue the study of crustacean reproduction and development and expand to include a crustacean that occupies a different ecological environment than does the crab. Begin a study of spawning and development of a finfish species that is an important member of the marine community in the Middle Atlantic Bight. Expand water quality and disease studies to include mortality problems that arise in rearing these new species in controlled environments.

EXPERIMENTAL BIOLOGY INVESTIGATIONS

ENVIRONMENTAL BIOASSAY INVESTIGATION

BACKGROUND Environmental stress alters the ecosystem and limits recruitment, abundance and distribution of living marine resources. Some marine contaminants or physical pollutants kill marine organisms outright. The widely publicized oil spills, dispersal of certain industrial wastes and thermal loading of the environment by electric power stations are commonly known examples. To provide a basis for environmental management, it is necessary to establish the precise levels of pollutants that can cause mortalities and the differential responses of marine organisms, including various stages in their life history. But perhaps of even more importance are the long-term effects of exposure to sublethal (stress) levels. Such exposure may limit development, growth, reproduction, metabolism, or other physiological processes.

Objectives of the investigation are to examine in the laboratory by bioassay, physiological and biochemical techniques, a selected group of Middle Atlantic Bight coastal animals and the effect of contaminants on their normal life functions. These laboratory experiments, when correlated with contaminant levels in the environment, will indicate that some marine animals are extremely sensitive to minute amounts of pollutants, or that certain animals or communities will flourish where specific contaminants are available at trace levels. Armed with this knowledge, the species identified by these studies will be useful as indicator organisms. Thus, a ubiquitous species may be absent wherever low levels of a specific contaminant are present. Conversely, indicator organisms may be used in laboratory studies where survival has a direct relation to contaminant concentrations.

RECENT AND ONGOING RESEARCH

Assessment of physiological changes in various species of marine bivalves, crustaceans and fish common to the New

York Bight area is now under way. Embryos of oysters and hard clams are being exposed to heavy metal ions through "in vivo" laboratory experiments to determine concentrations which affect normal development. Adult crustaceans and fish are being exposed to contaminants to determine changes in osmoregulation and respiration rates, and protocols are being developed to determine enzymological changes in key enzyme systems of these same species.

PROPOSED STUDY

These experiments will be expanded as part of the New York Bight Study, to include larvae of oysters and hard clams and embryos and larvae of other

species of shellfish. A variety of crustacean and fish species will also be exposed to heavy metals. The above organisms will also be challenged with a greater assortment of contaminant materials. These organisms will not only be subjected to acute exposures of contaminant material but, as facilities are developed, they will be subjected to long-term, chronic exposures of sublethal stress levels of pollutants.

Research will also concentrate on the effects of pollutants on the physiological processes of marine organisms, such as the changes in osmoregulation, respiration and neuromuscular activity, when under stress. In studies of this type, it will also be desirable to trace the movements and retention of contaminants within an organism to evaluate more fully the specific effects. By using radioisotopic tracers it will be possible to determine the pathways by which contaminants are accumulated in marine organisms, thus, the cause of death in marine organisms or the reason for stress may be more fully understood.

Also, as part of a broad-spectrum study of the effects of pollutants upon representative marine organisms, studies will be made to discover significant enzymological changes in the tissues of target animals during their exposure to known amounts of pollutants, and to interpret any such changes from the standpoint of biochemical adaptation or malfunction in response to the pollutant. By determining the degree and rate of such change in the light of concurrent work in related disciplines, it should be possible to monitor and even to predict the probable success or failure of the species to survive under known conditions of pollution.

RELATION TO OTHER PROGRAMS. Marine contaminant studies using bioassay, physiological and biochemical techniques are a part of the Experimental Biology Investigations

of the Center. Results must be examined and evaluated in relation to inputs from other lines of investigation (chemical and ecological base line studies) to achieve a more comprehensive view of the effect of contaminants on marine resources. Other NMFS Centers have or will have ongoing work using these approaches, as have other federal agencies, certain universities and private organizations. The work outputs derived from these studies would be made available to the Environmental Protection Agency for developing water quality criteria which, in turn, will be used for developing water quality standards for our aquatic ecosystems. These studies will also provide an informational base useful in coastal zone management and future planning functions, as well as to provide information necessary to assess the environmental impact of proposed coastal zone developments.

RESEARCH PLANNED FY 1973. Continue studies on the effect of contaminants on embryos and larvae of bivalves and adult rock crabs, green

crabs and the cunner. Studies of mortality and changes in osmoregulation and respiration will be made. FY 1973 will be the requisite exploratory period, during which tissues of rock crabs, green crabs and cunners will be examined and protocols developed for key enzyme systems.

FY 1974. Begin studies of the effect of contaminants on embryos and larvae of species of shellfish present in the New York Bight area, such as the surf clam, mahogany clam and sea scallop. Chronic exposure systems for bioassay work will be acquired and set up for studies of the effect of pollutants on the entire life cycle of marine organisms. Studies will also be made to determine changes in osmoregulation of small marine organisms, such as the grass shrimp and juvenile mollusks. Respiratory studies with stressed animals will be expanded to include polarographic measurements of large animals, as well as an expansion of present micro-respiratory techniques. Respiration rates of crabs, fish and mollusks will be determined "in vivo" during exposure to contaminants. To study more fully the physiological effects of pollutants, studies of neuromuscular (including cardiac) activity of stressed organisms will be initiated. In studies of biochemical-enzymological changes, work initiated in FY 1973 will be continued and research will concentrate upon acute-statis "in vivo" studies, beginning with exposure of target animals to a range of concentrations of pollutants and progressing to a study of synergistic effects. Main objects of scrutiny will be the polymorphic enzymes, whose enzymographic profiles may well alter as the animal adapts to its changing environment by changing the proportional concentrations of isoenzymes catalyzing the same reaction but differing in their optimal requirements.

taminants on embryonic and larval crustaceans and fish using bioassay techniques. Studies of changes in respiration of these embryonic and larval forms under stress conditions will also be conducted. Studies of chronic exposure and changes in physiological processes will also be continued. Studies of neuromuscular activity will also be continued. For studies of enzymological changes it will be possible to perform chronic-exposure studies that would more nearly reflect environmental conditions. By this time, specific enzyme systems will have been selected for study, their protocols (enzymographic and spectrokinetic) worked out and normal and acute-toxic patterns established.

PATHOBIOLOGY INVESTIGATIONS

SUMMARY STATEMENT

BACKGROUND Disease and parasite induced mortalities are paramount factors limiting the abundance of marine fish, crustaceans, and mollusks. Except for a few species, little is known about the causes of mortalities of eggs, larvae, juveniles, and adults of these valuable natural resources. Adequate knowledge of disease prevalence, whether infectious or non-infectious (nutritionally, genetically, or environmentally induced), is fundamental to the significance of resource assessments.

Mass mortalities of aquatic animals grown under intensive culture conditions are often a consequence of disease. In aquaculture, the indiscriminate transfer of animals from one location to another without regard to the disease entities they may harbor is a hazardous practice, the importance of which has not been adequately recognized. No Federal legislation exists to prevent the transplantation or emportation of diseased or potentially diseased aquatic animals, although the need for such legislation has been recognized by Congress for several years. The diagnosis of infectious diseases of animals used in aquaculture is, therefore, vital to prevent disease caused mortalities and to develop a fundamental competence in understanding the role of parasitism and disease as they affect resident and introduced populations of marine animals.

Host susceptibility to disease is directly influenced by environmental stress, and there are no ways of knowing what stresses are significant in limiting populations without studying their effects on the animals themselves.

One must recognize that it is the exception rather than the rule for abnormalities (pathoses) or mortalities (during any stage of the animals life history) to be caused by any single extrinsic or intrinsic factor acting alone. Rather, it is usually a combination of infectious and/or non-infectious factors acting competitively, sequentially, complementary, or synergistically on or in these animals to modify their behavior, physiology, growth, development, reproduction or to render them more susceptible to the same or still other infectious and non-infectious agents or predators.

Thus, in studies of diseases of aquatic animals, one must consider the ecology or the interrelationships both of the animals in question and the disease entity that affects or may affect it.

Armed with real, precise, and sufficient information on how infectious and non-infectious disease processes operate, it may then be possible to devise methods to present, eliminate, alleviate, or reduce infection pressures or stress factors that impinge on these animals.

OBJECTIVES

- 1. Develop competence, acquire information, and conduct field and laboratory research in pathobiology for purposes of determining the causes or factors leading to mortalities, abnormalities, and pathological manifestations, whether infectious, non-infectious, or environmentally induced, in wild or cultured marine and estuarine resources of the United States.
- 2. Develop a National Marine Diseases Diagnostic Laboratory to identify, classify, and describe infectious and non-infectious diseases in wild and cultured marine and estuarine animals and to establish a slide and tissue repository, a living museum of infectious disease entities, and a data bank of information on diseases and pathoses of living marine and estuarine resources for purposes of understanding the role that disease plays and understanding the interrelationships of other factors in mortalities, population fluctuations, availability, distribution of marine and estuarine resources.
- 3. Expand our histological and diagnostic services to more effectively assist those Federal, State, university, and private laboratories in understanding the causes of mortalities of marine resources, thus enabling them to plan and implement programs to prevent or alleviate losses or control production of living marine resources in nature or under conditions of culture.
- 4. Develop and establish, at the Oxford facility, an intensive program of training, study, and research in the areas of disease diagnosis and pathobiology for Federal, State, university, private, and industrial organizations. The program will be similarly patterned after the existing training program at the B.S.F.W. Eastern Fish Disease Laboratory at Leetown, W.Va. However, the courses, research projects, and areas of study will deal with diseases of marine fish, crustaceans, and mollusks.

PROPOSED STUDY

The Pathobiology Investigations at Oxford Laboratory have long recognized the need to study the causes of mortalities of all marine animals, and have been

pioneers in disease studies. Originally with mollusks, recently with crustaceans, and presently with fish, the Investigations will continually expand their research efforts to effect a multispecies approach to the study of disease. A multispecies study of disease has been substantially more productive than limiting the research effort to only a single species, since from the aspect of comparative and experimental pathology, invaluable basic information has been acquired on both disease processes and defense mechanisms.

By employing both comparative observational and experimental studies and team approaches, disease research on marine organisms will be conducted with the present Pathobiology Investigations staffs at the Oxford facility. This does not preclude the possibility that other projects under Pathobiology Investigations will not be undertaken at other Center facilities, or that teams of experts from Oxford will not be permitted to do "fire-fighting" research at other locations. Primary emphasis will continue to be placed on studies of problems related to infectious disease.

However, the Pathobiology Investigations staffs have long and broad experience and the expertise in recognizing pathological conditions in cells, tissues, and organs of marine animals. Therefore, we shall continue, in cooperation with other Federal, State, university, and industry laboratories, to examine, diagnose, and describe pathological (abnormal) manifestations that may be attributable to non-infectious disease agents or combinations of infective and non-infective agents. Furthermore, the Pathobiology Investigations, when deemed advisable and feasible, will award Investigation and Center supervised contracts to carry out the research necessary to assist in accomplishing our stated objectives.

RECENT AND ONGOING RESEARCH

It has become increasingly obvious that the effects of infectious disease continues to have a major, in some cases a devastating,

impact on the abundance and availability of living marine resources. Examples of reported massive die-offs in the U. S. are: herring in Maine and the Canadian Maritime Provinces; salmonids and alewives in the Great Lakes; yellow perch and menhaden in Chesapeake Bay; oysters and clams in several coastal U. S. locations; blue crabs in the Middle and South Atlantic States. Severe problems related to diseases in hatchery and aquaculture operations and the effects of the release of marine contaminants (oil, hydrocarbons, heavy metals) ocean dumping, dredging, and disposal also cause considerable concern to the scientific community, industry, and State and Federal resource management agencies.

The Pathobiology Investigations have directed most of their attention toward ascertaining the causes and developing remedial measures for widespread epizootics of bivalve mollusks as they occur in nature and under conditions of culture. Past and current research deals with multidisciplinary cooperative studies on: epizootiology of oyster diseases; disease resistance, pathology, and host responses to micropathogen challenges; life history studies, ultrastructure, taxonomy, and in vitro culture and nutrition of molluscan micropathogens such as Minchinia nelsoni (MSX), Minchinia costalis (SSO), Dermocystidium sp., and various other bacterial, protozoan, fungal, and viral pathogens.

Research on blue crabs has similarly dealt with studies of epizootiology and pathology of crab diseases, with many aspects done in cooperation with conservation units of the Middle and South Atlantic States. Laboratory studies have concentrated on isolation, characterization, systematics, in vitro culture of microbial pathogens, particularly Vibrio sp. and protozoan pathogens, particularly amoebae and haplosporidans, such as Paramoeba perniciosa, Minchinia sp. and Urosporidium.

Several cooperative studies with other Center units and State, university, industrial, and Federal laboratories are in progress to determine the pathological effects of marine contaminants on fish, crustacean, and molluscan resources.

Histochemical and histopathological diagnostic services rendered to State, industry, and Federal laboratories are also integral parts of the Pathobiology Investigations. Results of these studies have yielded discoveries of neoplasms and previously undescribed species of protozoans, bacteria, and viruses in several species of animals examined and form the basis of much of our ongoing research.

RELATION TO OTHER PROGRAMS

Experimental, descriptive, and diagnostic pathobiological research on normal and abnormal tissues of marine fish, crustaceans,

and mollusks to compare effects of stressed and nonstressed environments and other environmental influences on living marine resources will require full integration with other Center and NMFS programs. Field experiments and epizootiology, parasitology, comparative pathology, and microbiological studies in "dump site" and control areas of the Middle Atlantic Bight and Chesapeake Bay will involve the cooperation of the Marine Contaminants

and Estuarine Resource Assessment Investigations. Hematological and microbiological studies, parasite surveys, studies on various types of tumors and histopathological examination of "samples of opportunity" will also involve the above programs as well as the Ecosystems Investigations, MARMAP and Groundfish Survey Programs. Histopathology and ultrastructure studies on tissues and cells from specimens or organs of animals challenged acutely and chronically in the laboratory to various contaminants (heavy metals, organophosphates, petroleum products, polychlorinated hydrocarbons, pesticides, fertilizers, etc.) will be carried out cooperatively with the Experimental Biology Investigations at Milford, Conn., and with several State laboratories (Maine, Massachusetts, Maryland).

As the princi; al research arm of the National Marine Fisheries Service in disease studies of living marine resources, the Pathobiology Investigations are also obliged to participate in a broad spectrum of cooperative national and international disease projects with other Federal, State, university, fishery industry, and foundation laboratories. Thus, the histological/diagnostic services project cooperates as fully as possible with State and international conservation and industrial organizations in screening animals intended for transport and import into U. S. waters. Research on thermal effluents, virology, microbiology, and neoplastic diseases also are carried out in collaboration with several State organizations and university laboratories.

Informal liaisons have been established with EPA, FDA, NIH, NCI, AFIP, and with several State and university laboratories to deal with disease problems that may have some relation to public health; for example, those problems dealing with infectious hepatitis, neoplasms, Vibrio sp., human enteric organisms, and protozoan and helminth parasites.

RESEARCH PLANNED FY 1973. Continue ongoing projects of research as outlined above. Increase equipment and technical support staff to expand patho-

biology projects, particularly in the areas of crustacean and fish pathology. Implement research contracts as appropriate. Initiate research in histochemistry, hematology, serology, and in vitro culture of selected marine species.

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FY 1974. Expand histological and diagnostic services, particularly as they may relate to aquaculture operations and collaborative research. Using cell culture systems, increase effort in the study of viral diseases as they may affect living marine resources. Initiate computerized system for data correlation and retrieval. Plan and, if possible, initiate teaching-research program in marine animal pathobiology for NMFS units, and other Federal, State, university, and private laboratory personnel engaged in pathobiology studies. Continue contract research. Plan for acquisition of a scanning electron microscope, ultracentrifuge for virological studies.

FY 1975. Continue histological and diagnostic services, research contracts, and other ongoing projects in experimental and comparative pathobiology. Refine computer systems and projects in tissue culture (virology, microbiology). Implement new projects in biochemistry of infectious and non-infectious disease processes, experimental pathobiology, and ultrastructure studies.

PATHOBIOLOGY INVESTIGATIONS

COMPARATIVE PATHOBIOLOGY INVESTIGATION

A first and sometimes more direct approach used BACKGROUND in establishing the probable cause of a diseased condition is based on the histological examination of particular target tissues. Histological techniques have proven invaluable in the diagnosis of vertebrate pathology, and over the years have been successfully applied to studies of invertebrates. The histological examination of Crassostrea virginica from Delaware and Chesapeake Bays has elucidated the identity and probable life cycle of Minchinia nelsoni (MSX), a severe pathogen of this species of bivalve mollusk. More recently histological studies with other bivalves have established the identity of other parasites and aberrant histology consistent with mammalian neoplasia. It is expedient to employ histological techniques in studying the pathology of other marine invertebrates such as fish and crustaceans. Information so obtained is useful not only for its diagnostic value, but because studies in comparative pathology are more meaningful than studies of pathology in a single species.

Objectives of the Investigation are to conduct histolo-OBJECTIVES gical, cytological, and histochemical studies utilizing light and electron microscopy on moribund or stressed marine fish, crustaceans, and mollusks. Animals known to experience unexplained cyclic mortalities and animals intensively cultured (aquaculture) will be given first priority. As much as possible animals will be examined based on a need to know for regulation of the resource and for the information such studies would provide to the scientific community.

RECENT AND ONGOING RESEARCH

For convenience, it is desirable to categorize ongoing studies by animal species:

Mollusks

- 1. Histopathological studies of clams (Astarte, Macoma, Mya, Rangia, Spisula) from stressed environments.
- 2. Histopathological studies of clams (Macoma, Mya), mussels (Mytilus) and oysters (Crassostrea, Ostrea) with "proliferative" lesions.
- 3. Histopathological studies of oysters (Crassostrea) containing Type A nuclear inclusions.

Crustaceans

- 1. Histological studies of healthy and molting crabs.
- 2. Histopathological studies of stressed or diseased crabs.
- 3. Correlation of histopathological and epizootiological data pertinent to crab mortalities.
- 4. Further studies of "gray crab" disease in the blue crab (Callinectes).

Fish

- 1. Histological studies of commercially important marine fish of the Middle Atlantic coast.
 - 2. Histological studies of the cunner (Tautogolabrus).
- 3. Histopathological studies of cunners deliberately exposed to pollution and pollutants.
- 4. Compilation of a compendium of the hematozoan parasites of marine fish.

PROPOSED Research will concentrate on histopathological studies

STUDY of fish, crustaceans, and mollusks 1) from stressed environments, 2) deliberately exposed to specific toxicants or pathogens in the laboratory, and 3) exhibiting specific disease syndromes or other aberrant histology. Light microscopy, histochemistry, and electron microscope examination of ultrastructure (when indicated) will be employed as needed. Further approaches cannot be predicted at this time, since they will depend substantially upon findings.

RELATION TO

Results of studies conducted by the Comparative OTHER PROGRAMS

Pathobiology Investigation are of fundamental significance to all agencies involved in conservation of marine resources (State and Federal). It is conceivable that findings may have particular utility to academia since several Sea Grant contractees have a considerable investment in aquaculture of marine species.

RESEARCH PLANNED FY 1973. Complete histopathological studies begun on mollusks, crabs, and fish from New York Bight dump site areas

and control areas of the Middle Atlantic coast. Continue description of pathoses in marine animals exposed to experimental challenges with various pollutants. Complete epizootiological and histopathological studies on clams and crabs from stressed areas in Chesapeake Bay and South Atlantic. Continue diagnostic services on mollusks, crabs, and fish for Federal, State, university, and private organizations on both coasts. Continue surveys and monitoring projects on mollusks, crustacea, and fish from epizootic or stressed areas that will reveal distribution of pathoses in marine animals and will permit further research into micropathogen life histories, hematological and ultrastructure studies, description of normal and abnormal tissues. As appropriate, implement cooperative university research contracts in comparative pathobiology.

FY 1974. Continue comparative studies on normal and abnormal histology, cytology, parasitology, and hematology of animals from epizootic and stressed areas. Continue screening marine animals used in aquaculture or farming operations for pathologic conditions and presence and identification or infectious disease entities. Enlarge scope of cooperative studies with the Milford facility to determine qualitative and quantitative host responses to experimental challenges of marine pollutants. Continue ultrastructure studies on tissue and cell systems of marine animals from epizootic areas and those exposed to stress situations. Establish a computer system for correlation and recovery of data pertinent to research in pathobiology. Continue contract research begun in 1973.

FY 1975. Begin phase out or complete monitoring, survey, and epizootiological projects for which sufficient data have been accumulated to permit explanation of mortalities or presence of unusual pathological manifestations. Begin phase out of contract research begun in 1973 in comparative pathobiology host cellular responses to disease and stress factors; implement inhouse projects or cooperative studies on same. Continue ultrastructure, diagnostic services, and pathology projects. Continue computer system development. Initiate new contract research on hemopoietic mechanisms and of hematological characteristics of marine animals under normal, epizootic, or stressed conditions.

PATHOBIOLOGY INVESTIGATIONS

EXPERIMENTAL PATHOBIOLOGY INVESTIGATION

Inasmuch as descriptive studies involving BACKGROUND histology and histochemistry provide valuable information on pathological processes, they must be supplemented with studies in several of the basic sciences to be more effective. The disciplines of bacteriology, biochemistry, immunology, parasitology, and virology have contributed substantially to the understanding of pathology in vertebrates. These same disciplines have contributed materially to solving problems in invertebrate pathology; e.g., parasitologists have described the parasitic amoeba causing "gray crab" in the blue crab Callinectes sapidus and bacteriologists have established the identity of Gaffkya homari, a bacterial pathogen of the American lobster. The recent finding of Type A viral inclusions in the oyster Crassostrea virginica should provide an incentive to conduct virological studies on oysters as well as other invertebrates. In the examples cited, the basic sciences have provided information which was not previously attainable; hopefully, new findings in the area of invertebrate pathology will be achieved with the basic science approach.

Objectives of the Experimental Pathobiology
Investigation are to conduct bacteriological and
immunological research to establish the etiology of disease and to elucidate
mechanisms of disease resistance in fish, crustaceans, and mollusks.
Research will be directed wherever its relevance is greatest. Present
limitations on staffing limit the number of basic sciences represented to
bacteriology and immunology; however, it is anticipated that other
disciplines will be added when possible.

RECENT AND ONGOING RESEARCH

Immunological research in progress centers on the elucidation of the immune response in the blue crab <u>Callinectes</u> sapidus

and consists of the following:

- 1. Determination of natural agglutinin titers for vertebrate RBC's.
- 2. Immunization with RBC's and endogenous and exogenous bacteria.
- 3. Physicochemical characterization of immune serum including cellulose acetate and polyacrylamide electrophoresis and immuno-electrophoresis.

Bacteriological research will include the following:

- 1. Pathogenicity of selected bacteria, including <u>Gaffkya homari</u>, for the blue crab (<u>Callinectes</u>).
- 2. Pathogenicity of Vibrio parahaemolyticus for estuarine crustaceans and mollusks.
 - 3. Cytological studies of marine vibrios.
- 4. Antagonistic and synergistic effects of marine bacteria on Escherichia coli.
 - 5. Serological studies of fish exposed to terrestrial pathogens.
 - 6. Bacteriological studies of mass mortalities of menhaden (Brevoortia).

PROPOSED For the present, research will concentrate on immunoloSTUDY gical and bacteriological studies of crabs. However,
research will be directed where it is needed most. Since
research in these two areas is such that sustained investigations are both
necessary and desirable, interruption of work in progress by periodic
changes in priority will be kept to a minimum. Immunological research
will focus on characterizing the immune response in blue crabs, i.e.,
ascertaining whether it humoral or cellular (or both) and bacteriological
research will assess the pathogenic role of marine vibrios.

RELATION TO

Results of studies conducted by the Experimental
OTHER PROGRAMS

Pathobiology Investigation and by the Comparative
Pathobiology Investigation will complement those
conducted by other Pathobiology Investigations staff. As is true for studies
conducted by the Comparative Pathobiology Investigation, findings will be
of fundamental significance to all agencies involved in conservation of
marine resources (State and Federal).

RESEARCH PLANNED FY 1973. Complete studies of the immune responses of the blue crabs to various particulate and nonparticulate antigens.

Continue electrophoretic and physicochemical characterization of blue crab immune serum. Complete studies assessing pathogenicity of Vibrio parahaemolyticus for estuarine mollusks. Begin microbiological studies of menhaden mortalities. Implement cooperative university contracts on pathobiology of viral diseases of crabs, development of diagnostic techniques, and in vitro culture of invertebrate host cells. Initiate in vivo histochemical studies to determine chemical changes that accompany pathological manifestations in host tissues and cells and to follow biochemical changes as they occur during micropathogen development.

FY 1974. Begin studies of the immune responses of fish and mollusks to various particulate and nonparticulate antigens. Begin studies on the electrophoretic and physicochemical characteristics of immune sera from fish and mollusks. Continue bacteriological studies of menhaden mortalities. Begin studies assessing pathogenicity of Vibrio parahaemolyticus for estuarine crustaceans. Begin studies assessing the pathogenicity of marine vibrios for fish, crustaceans, and mollusks. Expand tissue culture and histochemical projects, especially those involving fish and crustacean cell systems for purposes of studying life cycles and replication mechanisms of fastidious micropathogens such as viruses, bacteria, and protozoans and cytopathic effects of infectious and non-infectious disease agents.

FY 1975. Expand studies in biochemistry of disease processes in host tissue and cell systems as they may result from infectious disease or from stress conditions. Phase out contract research projects as outlined for 1973-1974. Implement others in immunology, virology, and microbiology as appropriate. Continue tissue culture and histochemical projects, particularly those involving fish and crustacean cell systems. Utilizing in vitro techniques, implement projects in experimental parasitology from the standpoint of understanding pathogen growth, development, genetics, cytology, nutrition, and biochemistry.

AQUACULTURE INVESTIGATIONS

Because of the nature of the budgetary process, funding for significant in-house aquaculture research must be delayed until FY 1974. It is important, however, to have a clearly-defined plan of research and action developed during the interim period. It is also very important to note that much of the present research of the Center -- genetics, experimental biology, pathology, physiology, nutrition studies, contaminant exposures -- relates directly to aquaculture. What we have now, then, are the fundamental studies related to aquaculture; what we lack now is emphasis on aquaculture systems -- including systems engineering and development of continuous flow production systems.

Some areas which will be of significance in planning (in addition to aquaculture systems) include genetic selection of molluscan, crustacean and fish species; hybridization; development of technology with species other than oysters and hard clams (bay scallop, calico scallop, smaller clam species) -- species which may be reared in closed systems; endocrinology (particularly as it relates to growth and metamorphosis of larvae); algal physiology; and nutrition (of cultured species and of food supply organisms).