ADVANCES in FISHERIES SCIENCE

PATHOBIOLOGY of MARINE and ESTUARINE ORGANISMS

Edited by
John A. Couch and John W. Fournie

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CRC Press
Boca Raton Ann Arbor London Tokyo

Library of Congress Cataloging-in-Publication Data

Pathobiology of marine and estuarine organisms / edited by John A. Couch, John W. Fournie.
p. cm. -- (Advances in fisheries science)
Based on presentations at the Gulf Breeze Symposium on Marine and Estuarine Disease Research held in October 1990 and sponsored by the Center for Marine and Estuarine Disease Research, Gulf Breeze, Fla.
Includes bibliographical references and index.
ISBN 0-8493-8662-4
1. Marine fauna--Diseases--Congresses. 2. Estuarine fauna--Diseases--Congresses. 3. Marine fishes--Diseases--Congresses. I. Couch, John A. II. Fournie, John W. III. Gulf Breeze Symposium on Murine and Estuarine Disease Research (1990) IV. Center for Marine and Estuarine Disease Research (Gulf Breeze, Fla.) V. Series.
SF997.5.M33P37 1993
591.5'2636--dc20 91-37370
CIP

Direct all inquiries to CRC Press, Inc., 2000 Corporate Blvd., N.W., Boca Raton, Florida, 33431.

International Standard Book Number 0-8493-8662-4

Library of Congress Card Number 91-37370

Printed in the United States 1 2 3 4 5 6 7 8 9 0

Printed on acid-free paper

CRC Series in ADVANCES in FISHERIES SCIENCE

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SERIES PREFACE

ADVANCES IN FISHERIES SCIENCE

Advances in Fisheries Science is a book series published with the objective of providing in-depth treatment of the diverse subject matter that, taken together, forms the scope of fisheries science. Areas of emphasis within the series will include, but not be limited to, aquaculture, fishery management, fishing methods, descriptions of vertebrate and invertebrate fisheries, taxonomy and evolution of commercially important aquatic organisms, policy making with respect to fisheries, and relationships between fisheries and both natural and perturbed environments. Many additional topics of sufficient scope for a book in the series are encompassed within those broad headings. Some of the additional topics are genetics, molecular biology, nutrition, pathology, reproduction, behavior, and the general ecology of aquatic animals and, in some cases, plants.

Books in the series are designed to meet the needs of fisheries professionals and, in most instances, will be appropriate for use in upper division undergraduate and graduate courses in fisheries science.

ACKNOWLEDGMENTS

Many people contributed to the development of this book. We gratefully acknowledge authors of individual chapters. Informal comments on technical aspects of each chapter were provided by experts in relevant fields, whom we thank for their time and thoughtful critiques. Betty Jackson, editor for the U.S. EPA Laboratory, Gulf Breeze, Florida, was responsible for copy editing and organization of manuscripts. We thank Valerie Coseo, of Computer Science Corporation, for production of camera-ready text and Tom Poe, of Computer Science Corporation, for graphics. Susan Martin, of Technical Resources Incorporated, Gulf Breeze, assisted in reading page proofs. Cover illustration was designed by Lee A. Courtney of the EPA Laboratory, Gulf Breeze.

PREFACE

With the support of the United States Environmental Protection Agency's Environmental Monitoring and Assessment Program (EMAP), the Gulf Breeze Environmental Research Laboratory has created a Center for Marine and Estuarine Disease Research. As it develops the Center will focus on research questions dealing with the impact of xenobiotics and other anthropogenic stress factors on disease processes in marine and estuarine organisms. The Center will bring together research activities in this area of science within the Environmental Protection Agency, other federal agencies, universities, and independent research institutes. In addition to conventional research activities, the Center might organize response teams to investigate and identify the causes of major incidents involving mortality of marine or estuarine organisms with the objective of establishing the cause of the incident, either directly or through research which would ensue under the auspices of the Center.

The Center's first activity was to convene the *Gulf Breeze Symposium on Marine and Estuarine Disease Research* in October, 1990. The purpose of the symposium was to assess the state of the science in marine and estuarine disease research. Experts in the various aspects of diseases of aquatic mammals, fishes, and invertebrates comprehensively and critically reviewed the specific areas of their expertise. These presentations have been converted into the chapters of this book. The book is, however, an up-to-date compendium of what we know about diseases of marine and estuarine organisms at this time, not simply a compilation of the papers presented at the symposium. We believe it will serve students of marine pathobiology well as a resource for years to come.

The editors and authors hope that this book will serve as a stimulus for research in this important area of environmental science. In addition to the obvious economic importance of fish and shellfish, the organisms living in the marine and estuarine environment serve as useful indicators of the health of the nation's coastal waters. Their disease state is of critical importance.

Robert E. Menzer, Director
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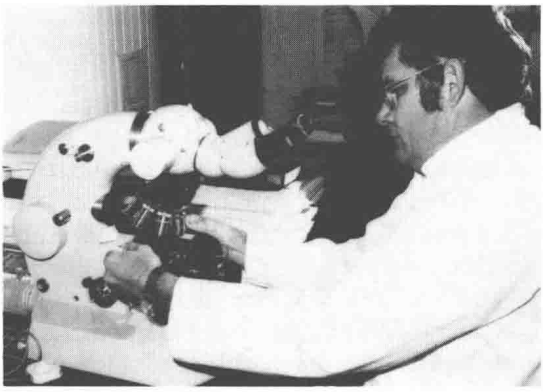
THE EDITORS



JOHN A. COUCH, Senior Research Scientist, U.S. Environmental Protection Agency (EPA), Environmental Research Laboratory, Gulf Breeze, FL, received a M.S. in Parasitology in 1964 and a Ph.D. in Cell Biology and Parasitology in 1971 from Florida State University. Dr. Couch has completed graduate and special course work in comparative pathology at Johns Hopkins School of Medicine, U.S. Armed Forces Institute of Pathology, and Harvard Medical School. His research interests are in toxicological pathology, experimental oncology with aquatic animals, and pathology of microbial disease agents in aquatic species. He has published extensively on infectious and noninfectious diseases of aquatic animals, and planned and coordinated major research programs on pathology and on lower animals as cancer research models.

Dr. Couch is a leading authority on diseases of molluscs and crustacea, and a pioneer in the pathology and experimental study of cancer in poikilothermic animals. He helped establish the emerging discipline of toxicological pathology of fishes. He originated and managed a four-year, multimillion dollar research program for the EPA and National Cancer Institute on the use of aquatic species in cancer research. He organized and managed the Pathobiology Branch at the EPA Research Laboratory in Gulf Breeze, FL, and originated the concept and original plan for implementation of the center for Marine and Estuarine Disease Research to resolve cause-and-effect problems in coastal and marine organisms' mass mortalities and diseases.

JOHN W. FOURNIE is a Fish Pathologist in the Pathobiology Branch at the EPA Center for Marine and Estuarine Disease Research in Gulf Breeze, Florida. He obtained B.A. and M.S. degrees in Biology from St. Louis University in 1974 and 1979. Dr. Fournie obtained a Ph.D. in Biology from the University of Mississippi and the Gulf Coast Research Laboratory in 1985 where his research emphasized fish parasitology and tumor pathology. He was a post doctoral fellow at the laboratory from 1985-1986, conducting research in small fish carcinogenesis.

Dr. Fournie has attained international recognition as a fish parasitologist for his work on coccidian parasites of fishes. He discovered that a true intermediate host is necessary in the life cycle of an extraintestinal fish coccidian, *Calyptospora funduli*, and reported on its infectivity, life stages, ultrastructure, and pathogenicity to a number of fish species. His work led to the redescription of this group, the creation of a new genus (*Calyptospora*) and a description of a new species. Dr. Fournie's more recent work has expanded into tumor pathology of fish and the use of small fish as carcinogenesis models. He has published numerous papers on various aspects of neoplastic diseases of fish, including detailed descriptions of ocular and exocrine pancreatic neoplasms. Research with carcinogen-induced pancreatic neoplasms in guppies led to an invitation to participate in the International Pancreatic Cancer Study Group meeting in Verona, Italy in 1988. Dr. Fournie has authored chapters on neoplasms of the exocrine pancreas and cardiovascular system, and co-authored a chapter on neoplasms of bone, cartilage, and the soft tissues.

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Importance of Marine Fish Diseases--An Overview

Michael L. Kent
John W. Fournie

INTRODUCTION

Diseases and pathogens may impact commercially important fish species, which include food, sport, and ornamental fishes. In addition to causing morbidity and mortality, diseases may cause poor flesh quality and undesirable aesthetic changes. Furthermore, some fish pathogens may infect humans or domestic animals. Fish diseases may also be useful as bioindicators of anthropogenic contamination (AC) or to elucidate the pathologic effects of specific toxicants. Fish disease research has generally evolved in two directions. One area of research has concentrated on diseases in economically important species, especially fishes reared in government or private aquaculture facilities. The other area involves diseases of fishes as indicators of anthropogenic contamination, either in natural populations from potentially polluted waters or in experimental populations exposed to toxicants. However, as with most areas of research, boundaries are arbitrary and overlap exists.

The numbers of researchers and programs in fish pathology have increased significantly in recent years. Fish farming is growing rapidly on a worldwide basis. As a result, research on diseases of these commercial stocks is increasing in importance. Research on diseases of wild fishes from potentially polluted waters has also increased due to greater concern about xenobiotics in the aquatic environment and an elevated awareness that fish may be useful sentinels of xenobiotic effects. In addition, the use of certain fish diseases as models in biomedical research is a new and growing field because fish have some advantages over mammalian models for toxicity and carcinogenicity testing (Dawe and Couch 1984).

Contribution No. 751, Environmental Research Laboratory, Gulf Breeze, FL

0-8493-8662-4/93/$0.00 + $.50

Other chapters in this book assess fish diseases according to etiologic agents (e.g., viruses, bacteria, and parasites) or specific pathological changes (e.g., neoplasms). This chapter reviews the major diseases of marine and estuarine fishes in three categories: 1) those affecting wild fishes, 2) those affecting captive fishes (e.g., fishes reared in fish farms or aquaria), and 3) those used as models for biomedical research. The three categories reflect the two major research areas of fish pathology. Research on infectious diseases is performed primarily on captive fishes, whereas research on fish as sentinels of xenobiotic effects primarily utilizes wild fish but may use small aquarium species for biomedical models.

Diseases of Wild Fishes

It is often difficult to assess the importance of diseases in wild fishes because moribund fish may be quickly eaten by predators or otherwise lost from the population. This is particularly true for chronic infectious diseases. The best documentation of infectious diseases in the wild are those which have resulted in massive, acute mortalities, yet there are undoubtedly many infectious diseases that occur in wild fish that have gone unrecorded. Capture methods may also be biased. Bait and lures tend to catch healthy fish, whereas trawls and nets may be biased toward catching moribund fish. Determining the effects of disease is complicated by the lack of knowledge on normal histological features and hematological parameters for most wild species. As a result, it may be difficult to determine the overall effect of a disease or pathogen on an individual or population. For example, it is often difficult to assess the significance of heavy parasite infections or the presence of *Mycobacterium* tubercles in an individual fish that appears otherwise normal, let alone the impact of such infections on the population.

Diseases Caused by Physical and Chemical Changes

Disease and mortality in wild fishes from coastal marine environments may be caused by changes in physical or chemical conditions of the water, including AC. Dramatic changes in water temperature have caused fish mortalities in various coastal areas throughout the world (Möller and Anders 1986). A few examples of massive fish kills associated with excessively cold water include kills in Mississippi (Overstreet 1974), Georgia (Dahlberg and Smith 1970), and the Gulf of St. Lawrence (Templeman 1965). Mortalities associated with a rapid decrease in temperature may be dramatic, and an estimated 30,000 tons of fish died due to cold shock in Texas during the spring of 1935 (Gunter 1952). In

most cases, cold shock does not induce any rapidly recognizable pathological changes, but some affected fishes have exhibited skin lesions (Möller and Anders 1986). Cold water temperatures may also cause increased susceptibility to infectious disease by inducing immunosuppression (Avtalion *et al.* 1976; Ellis 1982), or by altering natural host-parasite relationships. For example, Gulf killifish, *Fundulus grandis,* that are infected with the coccidian, *Calyptospora funduli,* can be severely impacted by exposure to low temperatures. Solangi *et al.* (1982) showed that temperatures of 7-10°C inhibited development and caused abnormalities in developmental stages of *C. funduli* in the Gulf killifish. Fournie (1985) showed that exposure of experimentally infected killifish to 3-4°C resulted in mortalities in infected fish, whereas uninfected fish survived. In addition, this cold temperature killed the parasite in the few surviving infected fish. The parasite is highly prevalent in wild killifish, and this observation could explain the low abundance of this fish reported by Solangi and Overstreet (1980) in years that followed cold winters.

There are examples of direct mortality in fishes due to increases in water temperature, particularly in planktonic stages of deep sea species (Möller and Anders 1986). These mortalities usually occur in fish trapped in abnormal water conditions. Fish trapped in the thermal effluent of power plants may be exposed to excessively high water temperatures. Young (1974) reported four separate incidents of high mortality in Atlantic menhaden, *Brevoortia tyrannus,* exposed to warm water from power plant outlets.

Low dissolved oxygen levels have caused fish kills in fjords and bays in Europe (Möller and Anders 1986), in estuaries of the southeastern United States (May 1973), and in Namibia (Brongersma-Sanders 1957). Reduced dissolved oxygen levels are often caused by decomposing organic material following plankton blooms or by upwelling of nutrient-enriched deep waters.

In addition to their importance as food and recreational sources, wild fishes may be useful as indicators of environmental contamination. There are numerous reports associating diseases and acute fish kills with AC of coastal waters, and there are several reviews on this subject (Overstreet and Howse 1977; Hodgins *et al.* 1977; Sindermann 1979; Sindermann 1980; Möller 1985; Mix 1986; Harshbarger and Clark 1990; Dawe 1990). In addition to acute mortalities, AC can cause non-neoplastic and neoplastic lesions, immunosuppression, increased susceptibility to infectious agents, and may affect reproduction. Contaminants may also affect fish by changing the composition of available food sources and may cause behavioral changes, such as avoidance of polluted waters. Möller and Anders (1986) suggested that changes in fish behavior caused by exposure to toxic wastes (e.g., emigration of sensitive species) may be a more important factor than disease induction in changing the composition of resident fish populations.

A few examples of acute mortality in fishes caused by AC in coastal waters include phosphorus pollution in Placentia Bay, eastern Canada (Iangaard 1970), copper sulfate pollution in the Netherlands (Roskam 1965), and parathion in waters along the eastern coast of Denmark (Boetius 1968).

Chronic pollution can cause histopathological changes, including neoplasms. Epizootics of neoplastic diseases in estuarine fishes in several locations have been associated with xenobiotics (Smith at al. 1979; Couch and Harshbarger 1985; Murchelano and Wolke 1985; Mix 1986; Baumann *et al.*, 1987; Harshbarger and Clark 1990; Myers *et al.* 1990; Vogelbein *et al.* 1990). In addition, AC has been cited as the cause of fin lesions, skin ulcers (Balouet and Laurencin 1983), skeletal anomalies (Couch *et al.* 1977), gill lesions (Couch 1975), hepatic megalocytosis (Myers *et al.* 1987; Peters *et al.* 1987), and preneoplastic liver lesions (Malins *et al.* 1987; Myers *et al.* 1987; Baumann *et al.* 1990; Hayes *et al.* 1990; Vogelbein *et al.* 1990).

However, the occurrence of these lesions, as well as many frank neoplasms, are not necessarily pathognomonic indicators of exposure to AC. Some of the diseases and lesions that have been attributed to AC are poorly substantiated (Mix 1986). Fin and skin lesions have been associated with AC, but they can also be induced by physical trauma, often with concurrent *Cytophaga-Flexibacter* bacterial infections (Anderson and Conroy 1969; Hikida *et al.* 1979; Kent *et al.* 1988). Epithelial separation of the gills has often been associated with exposure to contaminants, but this change can also be induced within minutes of death if the gills are not preserved immediately in appropriate fixatives (Speare and Ferguson 1989). Although hepatic megalocytosis is commonly observed following exposure to anthropogenic hepatotoxicants, this change is also observed in captive salmon (Kent 1990) and in certain species of wild fish from apparently pristine waters. Lastly, epizootics of hepatic and epidermal neoplasms have been shown to be strongly influenced by AC; however, others such as hemic, neural, and connective tissue neoplasms seem unrelated to environmental pollution exposure. In fact, there is evidence that some hemic and neural neoplasms of fishes are caused by infectious agents (Sonstegard 1976; Schmale and Hensly 1988; Kent and Dawe 1990).

Less obvious changes caused by pollution include impairment of defense mechanisms. Weeks and Warinner (1984) and Weeks *et al.* (1986) reported reduction in chemotactic efficiency and phagocytic activity of macrophages in fishes from the heavily polluted Elizabeth River, Virginia. These studies are substantiated by immunosuppression observed in laboratory exposure of fishes to specific chemicals (Anderson *et al.* 1984). Immunosuppressive chemicals include cadmium (Schreck and Lorz 1978; O'Neil 1981), copper (Donaldson and Dye 1975; Anderson *et al.* 1989), tributyltin (Wishkovsky *et al.* 1989), dioxin (Spitsbergen *et al.* 1986), and aflatoxin (Arkoosh and Kaattari 1987).