# AQUATIC ECOLOGY AT THE APPLIED PHYSICS LABORATORY

The Aquatic Ecology Section of APL's Environmental Assessment Group deals with both basic and applied environmental problems in fisheries ecology and aquatic toxicology in the Chesapeake Bay, its tributaries, and other aquatic ecosystems throughout the United States. Ongoing research is described on fish behavior in chlorinated effluents, on migratory history of fish, and on the toxicity of arsenic and selenium to fish.

# BACKGROUND

The Aquatic Ecology Section, established in 1980, is part of The Johns Hopkins University Applied Physics Laboratory's Environmental Assessment Group, which has had the lead responsibility for integrating the impact analyses for all proposed power plants in the State of Maryland since the beginning of Maryland's Power Plant Siting Program in 1972. The primary objectives of the group are to provide the technical basis for policy decisions by the state government regarding environmental issues and to present technical analyses in public hearings before the Public Service Commission and other regulatory bodies. The group has research interests in the areas of chemical processes, dispersion modeling and analysis, electromagnetic fields, acoustics, energy resource recovery, groundwater hydrology, power plant technology, socioeconomic analysis, systems analyses, waste disposal, and, more recently, aquatic ecology.

## **FUNCTION**

The Aquatic Ecology Section, which is currently composed of eight professional staff members, conducts research in aquatic toxicology, behavioral toxicology, aquatic ecology, and aquatic chemistry, and assesses the impact of energy production on aquatic organisms. It deals with environmental problems associated primarily with the Chesapeake Bay and its tributaries; however, results can generally be used to address problems in other geographic areas as well. Research projects include cooperative studies with personnel from other educational and research institutions, e.g., Rutgers University and the National Bureau of Standards.

### **UNIQUE FEATURES**

The Aquatic Ecology Section is located at The Johns Hopkins University's Shady Side campus on Parrish Creek at the mouth of West River about 15 miles south of Annapolis, Md. (see Fig. 1 and the inside back cover). The laboratories are designed and equipped to evaluate the acute, chronic, and sublethal effects of environmental factors and toxic substances on both freshwater and saltwater aquatic organisms under continuous-flow conditions. Bioassay facilities and test organism maintenance areas (15,000-liter tank space) are equipped with estuarine water systems that can deliver 375 liters of ambient estuarine water per minute, a freshwater system, an oil-free air supply, and programmed systems for simulation of day-night photoperiods. Associated equipment includes serial diluters for delivering toxicants, behavioral test units, and constant-temperature units for control of temperatures in water.

The analytical laboratories are well equipped with a wide variety of instrumentation for analysis of environmental samples. In addition to APL's equipment and facilities, the Aquatic Ecology Section has available on a cost-per-unit basis additional analytical capabilities and research vessels at the Chesapeake Bay Institute, also located at Shady Side.

# **CURRENT RESEARCH PROJECTS**

In addition to having a support role in power plant siting evaluation for the Environmental Assessment Group, staff members of the Aquatic Ecology Section are conducting a number of research projects for various sponsors. Recent investigations have included a joint study with scientists from the University of Maryland that deals with the stress effects of low chlorine concentrations and how they influence the susceptibility of striped bass (Morone saxatilis) exposed to bacterial disease organisms, and a hazard assessment of the effects of treated pulp and paper mill effluent on the early life stages of striped bass. which was conducted in a field laboratory at a large pulp and paper mill in Franklin, Va. Current investigations include the bioconcentration, metabolism, and elimination of certain water-soluble munitionsderived compounds (picric and picramic acids) in fish and oysters; behavioral avoidance responses of



**Figure 1** — The Johns Hopkins University Shady Side campus (center of figure) located on Parrish Creek at the mouth of West River in Maryland.

schooling fish exposed to heated chlorinated and dechlorinated power plant discharges; the use of ultrasound as an alternative to toxic biocides (such as chlorine) for prevention of biofouling in power plant cooling water systems; chemical speciation and biological effects on fish of arsenic and selenium from runoff and leachate from the storage of coal wastes; development of technology that can be used to evaluate the behavioral responses of fish exposed to very low concentrations of organotin antifouling paints or other toxic materials that may be used in naval shipping operations; and analysis of the elemental composition of striped bass scales and otoliths (ear bones) to determine the migratory patterns of the fish in and out of the Chesapeake Bay and Atlantic Ocean.

Highlights of three of the current research projects will be discussed to give an indication of the types of aquatic research being conducted. The use of ultrasonics as an alternative to chlorine for inhibiting biofouling was recently described.<sup>1</sup>

### Fish Behavior in Chlorinated Effluents

Chlorine is the most widely used biocide in the United States for control of biological growth in both once-through and recirculating cooling water systems of electric power plants.<sup>2</sup> In recent years, however, the use of chlorine for biofouling control has come under intense scrutiny and regulation because of possible detrimental effects on aquatic ecosystems. Numerous investigations have shown that chlorine and its by-products are toxic to aquatic organisms; however, little work has been performed on the sublethal physiological and behavioral responses of fish ex-

posed to chlorinated waters.<sup>3</sup> The present study addresses the behavioral responses of schooling fish to chlorinated discharges from power plants.

The study has two objectives. The first is to determine the avoidance behavior of Atlantic menhaden (Brevoortia tyrannus) and striped bass exposed simultaneously to chlorine and elevated temperatures. These represent conditions during the spring, summer, and fall, when power plants discharge chlorinated water into the Chesapeake Bay. Avoidance behavior is being investigated because it is known that fish can detect various pollutants in the environment and, in many cases, avoid the pollutants before they are adversely affected. Atlantic menhaden are being studied because of their commercial importance in the Chesapeake Bay and the fact that they are frequently found in large schools in the discharge areas of power plants (Fig. 2). Striped bass are commercially and recreationally important in the Bay, and they too use habitats in the area of power plants.

The second objective of the study is to evaluate the avoidance responses of the above species when they are subjected simultaneously to dechlorinated water and elevated temperatures, conditions that simulate those occurring in power plant effluents. The sequence of reactions involved in dechlorination of water by sulfur dioxide is shown in Fig. 3. Chlorination is a proven technology for control of biofouling in power plants; therefore, feasible as well as economical methods for reducing its toxic effect need to be evaluated. Sulfur dioxide treatment has been shown to reduce the toxicity of chlorine to aquatic organisms. It is particularly promising for large-scale use in power plants because the handling and measurement of sulfur dioxide are very similar to those





Figure 2 — School of Atlantic menhaden in the lower Chesapeake Bay.

of chlorine.<sup>4</sup> The evaluation of the avoidance responses of fish exposed to conditions of dechlorinated water and elevated temperatures will provide useful data for recommending or rejecting dechlorination practices for power plants in Chesapeake Bay and other estuarine and marine environments where chlorine is being used.

The results of the Atlantic menhaden studies conducted at summer temperatures (30°C) will be used as an example to describe the behavior of schooling fish in a power plant discharge.<sup>5</sup> Atlantic menhaden strongly avoid both increased temperatures ( $\Delta T$ )

Chloring reactions

and chlorine (TRC, total residual chlorine) in chlorinated discharges during the summer.<sup>6</sup> Figure 4 shows that Atlantic menhaden schools that are acclimated to summer temperatures ( $\Delta T = 0^{\circ}$ C) with no chlorine present (0.00 TRC) randomly spend approximately 50% of their time in the control portion of a behavioral test chamber. As soon as temperatures are increased as little as 2°C above ambient (30°C) with no chlorine present, the menhaden actively avoid the increased temperatures; they spend about 85% of their time in the control area at ambient temperature (30°C). Greater than 95% avoidance occurs at  $\Delta T$  of 6°C above ambient temperature. The avoidance response of Atlantic menhaden is more pronounced when the school encounters chlorine. All fish avoid chlorine at concentrations as low as 0.05 milligram per liter TRC when there is no increase in ambient temperature ( $\Delta T = 0^{\circ}$ C). (Federal law normally allows power plants to chlorinate up to 0.2 milligram per liter TRC.<sup>7</sup>) Atlantic menhaden avoid all combinations of TRC above 0.05 milligram per liter and  $\Delta T$  above 2°C. These data show that Atlantic menhaden can avoid adverse chlorine and temperature conditions that may occur in the discharge area of a power plant.

Dechlorination of power plant effluent with sulfur dioxide radically changes the avoidance behavior of schooling Atlantic menhaden (Fig. 5); the behavior was studied under the same test conditions as those described above except that the chlorine was removed by addition of sulfur dioxide. A comparison of Figs. 4 and 5 shows that the avoidance responses elicited by chlorine do not occur when the effluent is dechlor-

Sulfur dioxide hydrolysis

Cl <sub>2</sub> + HOH ← HOCI + H <sup>+</sup> + Cl <sup>-</sup> chlorine gas water hypochlorous hydrogen chloride acid ion	$\begin{array}{rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$
HOCI ₹ H <sup>+</sup> + OCI <sup>-</sup> hypochlorite	$H_2SO_3 \stackrel{\rightarrow}{\leftarrow} H^+ + HSO_3^-$ hydrogen ion bisulfite
NH <sub>3</sub> <sup>+</sup> HOCI → NH <sub>2</sub> CI <sup>+</sup> HOH ammonia monochloramine	$HSO_3$ $\stackrel{-}{\leftarrow}$ $H^+$ + $SO_3^{2^-}$ sulfite
RNH <sub>2</sub> + HOCI → RNHCI + HOH organic amine organic chloramines	

# Dechlorination reactions



Figure 3 — Sequence of reactions involved in dechlorination of water by sulfur dioxide.

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Figure 4 — Three-dimensional response surface of percent avoidance for Atlantic menhaden exposed to various chlorine concentrations and increased temperatures (ambient temperature is 30°C).

inated. The fish do, however, avoid heated waters as the temperatures increase above ambient temperature. The fact that Atlantic menhaden do not avoid dechlorinated effluent is important. For example, a chlorinated effluent that extends across a river could block the migration or movement of fish during the summer; however, when the effluent is dechlorinated and the changes in temperature are within acceptable limits, normal movement of the fish school will not be affected. This has important implications for mitigating or reducing adverse environmental effects of large power plants on fish migration and behavior.

# Migratory Histories of Fish

The elemental composition of striped bass scales and otoliths (ear bones) is being studied to reconstruct age- and sex-specific migratory histories of Chesapeake Bay striped bass. Information on migratory patterns in such anadromous fishes as the striped bass is essential to the development and implementation of fisheries management strategies. Based on analyses of tagging, mortality, and fall sex composition data, it has been concluded that the migratory behavior of Chesapeake Bay striped bass is age and sex specific.8 For example, it has been suggested that young adult males rarely emigrate from the Bay to coastal waters and, because they are subjected to heavy fishing pressure in the Bay from age 2 to 6 years, that few survive to an age when emigration might occur. It has also been suggested that more than 50% of the adult females between ages 2 and 4 years emigrate to coastal waters and do not return to the Bay until they are age 5 or 6 and ready to spawn.



**Figure 5** — Three-dimensional response surface of percent avoidance for Atlantic menhaden exposed to dechlorinated water that had contained various concentrations of chlorine and to increased temperatures (ambient temperature is 30°C).

These age- and sex-specific migratory patterns have important implications for fisheries management for Maryland, Virginia, and other Atlantic states. Ideally, one would like to know where an individual male or female striped bass spent each year of its life so that a composite of migratory histories for each age group of males and females in the stock can be constructed. Conventional mark and recapture studies reveal only where each individual was released and where it was recaptured; nothing can be learned about its movements during its time at large. Tracking large numbers of individuals tagged with radio, sonar, or ultrasonic transmitters over several years throughout the Bay and along the Atlantic coast is clearly impractical.

Detection of chemical markers in various fish tissues offers a more promising approach to the problem.<sup>9</sup> The tissues of geographically distinct populations or stocks of fish tend to reflect the chemical constituents of the water in which they reside. Scales and otoliths are ideal tissues for deposition of chemical markers since they are calcareous structures laid down concentrically as fish grow (Fig. 6) and are relatively inert metabolically during most of the fish's life. Since the elemental composition of the scales and otoliths deposited during each growth period is preserved through time, these tissues should provide a continuous chemical record of the environment in which the fish spent each year of its life.

If we apply this concept to the reconstruction of migratory histories of Chesapeake Bay striped bass, it is logical to expect that some distinguishable chemical record of the years spent in the Bay and in the Ocean is made on each scale and otolith. Therefore,



Figure 6 — Scale taken from a 4year-old striped bass.

measurements of those elements whose concentrations in estuarine and marine waters are known to be quite different in areas of the scales or otoliths that correspond to known growth periods should indicate whether the individual spent a particular growth period in the Chesapeake Bay or in the Atlantic Ocean. Several alkaline earth elements appear to be potentially useful chemical markers deposited in the scales and otoliths. Strontium, magnesium, and calcium concentrations are much higher in seawater than in waters of low salinity or in fresh water.

Recent experiments, based on whole-scale analyses using atomic absorption spectrophotometric methods, show that striped bass collected from salt water and from fresh-water land-locked reservoir populations can be distinguished on the basis of strontium concentrations in their scales as well as on the basis of ratios of calcium to strontium and of magnesium to strontium. Preliminary microdissection studies, which were used to separate individual growth periods of the scales, have also demonstrated differences in the concentrations of the above alkaline earth elements for various populations. However, microdissection is rather tedious and time consuming and, in general, a crude method for isolating individual growth periods. The feasibility of using Xray microprobe scanning electron microscopy to measure the concentrations of selected elements within a specific growth period on the scales and otoliths is being explored. It is hoped that this technique will provide the technological breakthrough needed for analyzing large numbers of samples. This, in turn, should enable us to begin reconstructing the age- and sex-specific migratory histories of Chesapeake Bay striped bass.

### Arsenic and Selenium Toxicity to Fish

The use of coal for generating electric power will likely increase in future years so that we might achieve our national goal of energy independence.

The increased use of coal will result in the production of large quantities of fly ash, scrubber sludge, and other coal waste disposal piles that contain a complex mixture of salts, metals, and organic material. Several investigators have reported that arsenic and selenium are important components of coal pile and coal waste effluents that could cause potential biological problems in the aquatic environment near coal-fired power plants.<sup>10</sup> Increased concentrations of both metalloids above ambient levels have been recently found in some juvenile striped bass collected from various rivers in Maryland and other East Coast rivers.<sup>11</sup> The study discussed here is addressing the chemical speciation and partitioning of both metalloids between the water column and estuarine sediment and the toxicity of both metalloids to the early life stages of striped bass that may encounter coal-related effluents in certain spawning and nursery areas.

The chemical state of arsenic and selenium is a major determinant of the geochemical cycling and toxicity of the metalloids to aquatic organisms.<sup>12</sup> Both elements are found in at least two inorganic oxidation states and several organic forms. Each of these states has different geochemical and biological properties. The biological cycle for arsenic is shown in Fig. 7 (the selenium cycle is similar). Current chemical studies in our laboratory indicate that arsenate and selenate may be the most prevalent forms of the metalloids in the estuarine water column; therefore, these chemical species are being used in the biological studies.

The acute (less than 4 days of exposure) and chronic (exposures up to 3 months) toxic effects of arsenic and selenium on striped bass are being determined in controlled laboratory experiments. The life stages being tested are eggs, prolarvae, larvae, and juveniles because early life stages of fish, especially prolarvae and larvae, are generally more sensitive to pollutants than older life stages. Eggs, prolarvae, larvae and juveniles are being exposed to arsenate and selenate



Figure 7 — Biological cycle of arsenic (modified from Ref. 12).

separately, followed by additional acute tests in which a mixture of both metalloids is being used to test for combined toxicity. Studies of prey consumption and growth with larvae and studies of growth with juveniles are being conducted to determine possible chronic sublethal effects of the metalloids (as isolates and mixtures) on the critical life stages.

Empirical models for predicting the acute combined toxicity of arsenate and selenate to all the life stages are being developed. These types of models are also being constructed to determine the effects of the metalloid mixtures on the feeding and growth responses of striped bass larvae and early juveniles. The results of the chemical and biological studies will provide much needed information about the potential effect of coal-related effluents on the reproductive success of this important anadromous fish species. The studies will also be useful for determining the treatment that may be necessary to control discharges of coal-related effluents into important spawning and nursery habitats.

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