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NOAA Technical Memorandum NMFS-F/NEC-61

Fish as Sentinels

of Environmental Health

U.S. DEPARTMENT OF COMMERCE National Oceanic and Atmospheric Administration National Marine Fisheries Service Northeast Fisheries Center Woods Hole, Massachusetts

September 1988

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<u>Abstract</u>: Boston Harbor, Massachusetts, is polluted by storm water runoff, municipal wastewater, and sewage sludge, as well as riverborne domestic and industrial pollutants, and introductions characteristic of and related to the sustained heavy commercial ship traffic in the port. Shallow areas of the inner harbor provide suitable spawning and nursery areas for winter flounder (Pseudopleuronectes americanus). The harbor contains large numbers of winter flounder during most of the year and is virtually a sanctuary for the species since commercial fishing is prohibited. Recent disease surveys have shown that harbor winter flounder have a high prevalence of gross and microscopic hepatic lesions, including carcinomas. Although harbor sediments contain high concentrations of polycyclic aromatic hydrocarbons (PAH) and polychlorinated biphenyls (PCB), presently the evidence ascribing either one of these organic chemicals as a direct cause of the neoplastic lesions observed is circumstantial.

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Fish as Sentinels of Environmental Health

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Boston Harbor, Massachusetts, is polluted by storm water runoff, municipal wastewater, and sewage sludge, as well as riverborne and industrial pollutants, and introductions domestic characteristic of and related to the sustained heavy commercial ship traffic in the port. Shallow areas of the inner harbor provide suitable spawning and nursery areas for winter flounder (Pseudopleuronectes americanus). The harbor contains large numbers of winter flounder during most of the year and is virtually a sanctuary for the species since commercial fishing is prohibited. Recent disease surveys have shown that harbor winter flounder have a high prevalence of gross and microscopic hepatic lesions, including carcinomas. Although harbor sediments contain high concentrations of polycyclic aromatic hydrocarbons (PAH) and polychlorinated biphenyls (PCB), presently the evidence ascribing either one of these organic chemicals as a direct cause of the neoplastic lesions observed is circumstantial. As a case study, however, investigations as to the cause and prevalence of hepatic carcinoma in Boston Harbor winter flounder will attest to the utility of fish as sentinels of aquatic environmental health.

INTRODUCTION

It is well known that anthropogenic activity has contributed to deterioration in marine water quality, especially in coastal areas adjacent to centers of large populations. A variety of effects, including diseases, have been studied in marine fishes, crustaceans, and mollusks from polluted marine environments (12, 13, 19, 20). Neoplastic diseases of the integument (papilloma) and liver (hepatoma), particularly of bottom-dwelling fishes, are more prevalent in coastal areas than areas which are relatively pristine (6, 10, 11, 17, 21). Metabolic especially those mediated by cytochrome Metabolic transformations, P-450 dependent mixed-function oxygenases, may predispose certain fish species to the development of hepatic neoplasms (22). Boston Harbor winter flounder (Pseudopleuronectes americanus), which are important commercial and recreational fish, have a high prevalence of hepatic carcinoma which may be caused by environmental carcinogens (14).

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The Environment

Boston Harbor is an estuary whose exit to Massachusetts Bay is bounded on the northeast by Deer Island and on the southeast by Hull (Fig. 1). The harbor contains about a dozen islands of varying size; the larger ones are clustered in the central part and, together with a seaward extension of the harbor's western shore, divide the harbor into northern and southern parts. The northern harbor consists of Dorchester Bay and the inner harbor. A deep shipping channel, President Roads, traverses the northern harbor from the inner harbor to Massachusetts Bay. President Roads is 1.6 km (1 mi) wide and up to 22 m (73 ft) deep. The southern part of the harbor consists of Hingham and Quincy Bays. A second deep shipping channel, Nantasket Roads, proceeds from Massachusetts Bay to Quincy Bay; the channel has several smaller branches terminating in Hull Bay.

Except for dredged channels, Boston Harbor is relatively shallow with mean depths (at mean high tide) of 5.4 m (17.7 ft) in Dorchester Bay, 6.8 m (22.4 ft) in Quincy Bay, and 5.1 m (16.7 ft) in Hull Bay. Freshwater inputs to the harbor from north to south include the Mystic, Charles, Neponset, Weymouth Fore, Weymouth Back, and Weir Rivers. The harbor is a saline estuary (salinity = 25 0/00) which is well mixed during periods of greatest freshwater flow (late winter, spring) and stratified in late summer (August) when freshwater input is minimum. Water temperature and salinity vary seasonally.

As with most estuaries located adjacent to large population centers, Boston Harbor is the recipient of a large variety of point and non-point source pollutants, including substantial quantities of domestic sewage and industrial wastes. In the northern harbor, there are numerous storm sewer outlets and one major treatment plant discharge (Deer Island). There also is a major treatment plant discharge in the southern part of the harbor (Nut Island). The Deer Island plant services the northern municipalities and provides primary treatment for an average daily flow of 343 million The plant began operating in 1895 and gallons per day (mgd). serves a population of approximately 1,340,000 people in a 273 km² (105.4 mi⁻) area. The Nut Island plant services the southern municipalities and provides primary treatment for an average daily flow of 112 mgd. The plant began operating in 1904 and serves a population of approximately 630,000 people in a 258 km² (99.6 mi²) area. The Deer Island plant discharges into President Roads and the Nut Island plant into Nantasket Roads.

Because of its shallow bays and numerous creeks and marshes, Boston Harbor is a habitat for several economically important species of fish and shellfish. Other than for Atlantic menhaden (<u>Brevoortia</u> <u>tyrannus</u>), there are no commercial finfisheries in the harbor. Recreational fisheries for Atlantic cod (<u>Gadus morhua</u>), bluefish (<u>Pomatomous saltatrix</u>), American eel (<u>Anguilla rostrata</u>), pollock (<u>Pollachius virens</u>), striped bass (<u>Morone saxatilis</u>), and winter flounder are seasonal (summer primarily) and vary considerably in

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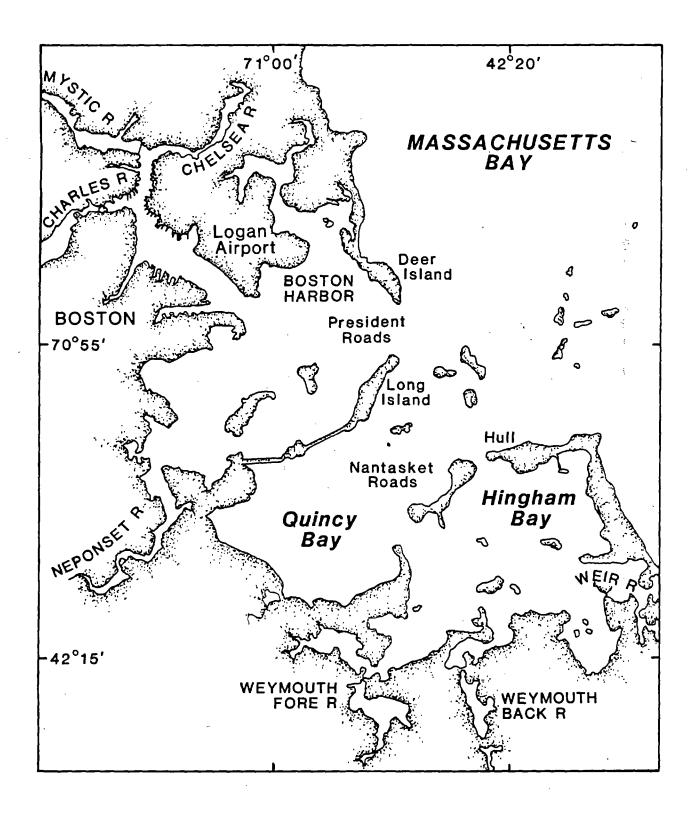


Figure 1. Chart of Boston Harbor. Major sources of freshwater are shown.

importance and size. The winter flounder fishery is by far the largest in the harbor and is of substantial economic importance to the area. Although winter flounder are caught by recreational fishermen in the harbor, when the fish migrate to nearby Massachusetts Bay they are caught in trawls by commercial fishermen.

The primary economically valuable crustacean resource in Boston Harbor is the American lobster (<u>Homarus americanus</u>). Lobsters are trapped throughout the harbor between April and October.

The soft-shell clam (<u>Mya arenaria</u>) is the most economically important molluscan resource in the harbor. Harvesting of clams from areas with coliform most probable numbers (MPN) of 71-700/100 ml is restricted and clams must be depurated before sale; areas where coliform MPN exceed 700/100 ml are closed to clamming.

The Fish

Winter flounder can be found from Newfoundland to the Chesapeake Bay; however, individual fish have been captured as far north as Labrador and as far south as Georgia (1).

Winter flounder are estuarine dependent and spend a major part of each year in shallow nearshore waters. Their seaward migrations are mediated by changes in water temperature and determine the abundance and distribution of the fish in all coastal areas, including Boston Harbor (8). Winter flounder north of Cape Cod seek lower temperatures in deeper waters when inshore temperatures are unusually high in summer, and higher temperatures in deeper waters when inshore temperatures are unusually low in winter. Sexually mature fish migrate to shallow breeding areas, 2-6 m (7-20 ft) deep, in the fall and winter and spawn in late winter through early spring. Winter flounder from Massachusetts are sexually mature when they are 3-4 yr old. Young fish remain in shallow inshore waters (nursery grounds) and do not migrate.

The winter flounder commercial fishery is conducted almost exclusively with otter trawls in the spring and fall. In 1983, the total U.S. catch of winter flounder was 14,967.7 metric tons (mt) valued at 15,795,000 dollars (Thompson; pers. comm.¹). The New England catch (Connecticut, Maine, Massachusetts, New Hampshire, Rhode Island) was 14,147.9 mt, the Middle Atlantic catch (Delaware, New Jersey, New York) was 794.4 mt, and the Chesapeake catch (Maryland, Virginia) was 24.5 mt. The winter flounder catch in Massachusetts was 10,642.2 mt valued at 11,557,000 dollars. The Massachusetts landings constituted 64.6% of the U.S. total in weight and 73.1% of the U.S. total in dollars; 90.2% (8,729.5 mt)

¹Thompson, B.G.; U.S. Dept. of Commerce, NOAA, National Marine Fisheries Service, Office of Data and Information Management, National Fishery Statistics Program, Washington, DC 20235.

of the Massachusetts catch is made within 3 to 200 miles of the coastline. The bulk of the U.S. catch, especially in the New England States, comes from Georges Bank. As in most bottom fish fisheries, the most trawled areas are those with abundant fish and favorable bottom characteristics.

Although winter flounder support a substantial otter trawl commercial fishery, the size and value of the recreational catch also are very significant. The fish can be caught along the entire U.S. Middle Atlantic and Northeast coasts by recreational fishermen using hook and line. In 1982 (the latest year with complete statistics), the New England recreational catch of winter flounder was 5,575.9 mt (Thompson; pers. comm.²). The Middle Atlantic recreational catch was 3,163.9 mt. Together, the 1982 New England and Middle Atlantic recreational catch of winter flounder totaled 8,739.8 mt; the commercial catch in the same year was 16,163.6 mt. By percentage, the commercial catch was 63% and the recreational catch 37% of the total catch. Although economically important to the Commonwealth of Massachusetts, the dollar value of the Boston Harbor recreational winter flounder fishery is unknown.

The Lesions

Histology is that discipline of biology which is concerned with the normal microscopic structure of tissues. Histopathology, however, is concerned with the structure of abnormal tissue. Histopathology, with some exceptions, is not used extensively in the diagnosis of diseases of warm-blooded animals. Modern diagnostic medicine employs biochemical, hematologic, radiographic, and other innumerable sophisticated technologies. In human medicine, histopathology is used primarily to identify presumptive neoplastic lesions and in surgical pathology to substantiate the need for operative procedures. The use of histopathology to diagnose diseases of fish has considerably more appeal than it does with warm-blooded animals, partly because of the limited value of other procedures and partly because the fish can be sacrificed and all organs examined for the presence of structural changes.

In recent years, several species of marine fishes have been found to have high prevalences of hepatic neoplastic lesions, including Atlantic hagfish (<u>Myxine glutinosa</u>) (7), English sole (<u>Parophrys</u> <u>vetulus</u>) (11), and Atlantic tomcod (<u>Microgadus tomcod</u>) (21). Winter flounder from Boston Harbor display a variety of hepatic lesions. The most numerically prevalent ones are hypertrophy and hyperplasia of macrophage aggregates and a modified form of hepatocyte apoptosis. Other significant lesions include pericholangitis and focal parenchymal cell necrosis. Lesions which are presumed to be preneoplastic include basophilic, eosinophilic, and vacuolated cell foci. Non-neoplastic, proliferative lesions include cholangiolar (bile duct) hyperplasia and cholangiofibrosis.

²Thompson, B.G., <u>op.</u> <u>cit.</u>, p.4.

Neoplastic lesions include adenoma, cholangiocarcinoma, and hepatocarcinoma (Figs. 2, 3, 4). The neoplastic lesions observed resemble those experimentally induced with carcinogens in rodents. Although the hepatic carcinomas frequently were large and involved a substantial portion of the liver, no grossly visible metastatic lesions were observed in gill, heart, intestine, kidney, or spleen.

For the non-specialist, the abnormalities seen in the livers of the Boston Harbor winter flounder include inflammation, necrosis (cell death), and tumor formation. The liver tumors (neoplasms) are comparable microscopically to several types of liver cancers of warm-blooded animals, including man. Presently, there is no evidence that the winter flounder liver cancers spread to other organs.

The Cause

Discussions of the cause(s) of the hepatic neoplastic lesions observed in winter flounder from Boston Harbor are necessarily speculative. For marine fishes, no conclusive evidence (based on experimentally derived data) presently exists substantiating a specific chemical agent as the inducer of a particular neoplastic Any of innumerable environmental chemical contaminants lesion. could be the cause of the neoplasms. Experimentally induced tumorigenesis has been studied in few fishes, and with relatively few chemicals, at least in proportion to the number of chemicals likely to be present in anthropogenically degraded aquatic environments (5). All the fish used in studies of experimentally induced carcinogenicity have been freshwater species and include, in descending order of use, rainbow trout (Salmo gairdneri), guppy (Poecilia reticulata), medaka (Oryzias latipes), and zebra danio The chemical agents used in laboratory studies of (Danio rerio). tumor induction include primarily aflatoxins and nitrosamines; the liver was the principal organ affected by neoplastic transformation.

Relatively few studies have focused on the cause of epizotic neoplasia in marine fishes. Polychlorinated biphenyls (PCB) have been implicated as the inducers of hepatic neoplasms in Atlantic hagfish from Swedish fjords (7) and Atlantic tomcod from the Hudson River (21). Although PCB's initially were implicated as possible causes of hepatic neoplasms in English sole from Puget Sound (11), more recent studies suggest that polycyclic aromatic hydrocarbons (PAH) may be responsible (10). PCB concentrations (wet weight) were approximately 5 ppm in livers of Atlantic hagfish with hepatomas, and were 10.9-98.2 ppm (mean 37.5 ppm) in livers of Atlantic tomcod with hepatomas. Concentrations (dry weight) of PCB's in livers of English sole from two urban areas in Puget Sound, Commencement and Elliot Bays, were 39 ppm and 47 ppm, respectively (for liver, dry weight/2 approximates wet weight). PCB concentrations in livers of English sole from two non-urban areas, Case Inlet and Port Madison, averaged 2.3 ppm.

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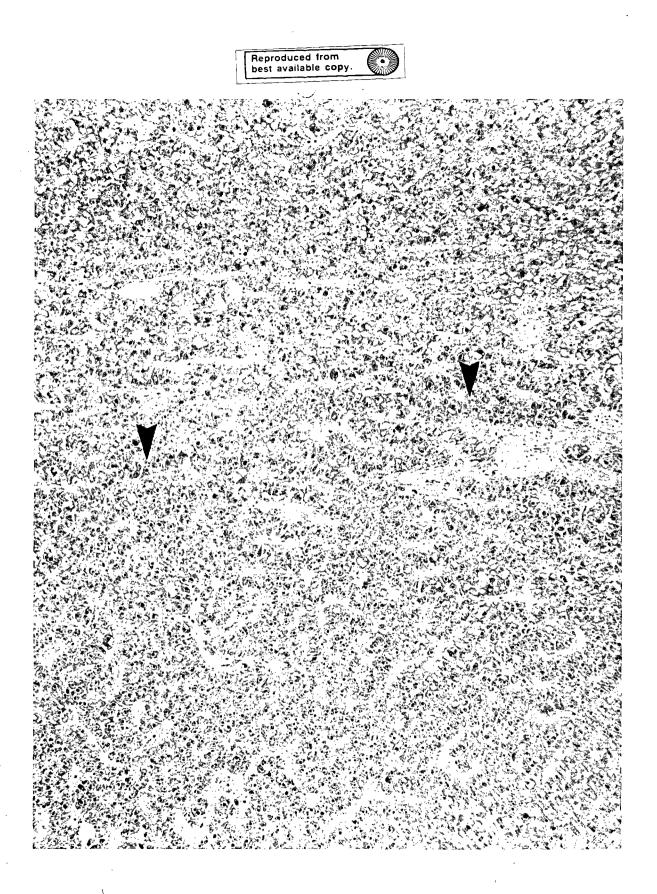


Figure 2. Hepatocellular adenoma in liver of winter flounder from Boston Harbor. Hepatocytes of adenoma are darker and less vacuolated than those of adjacent liver parenchyma. Arrows define the edge of the lesion.

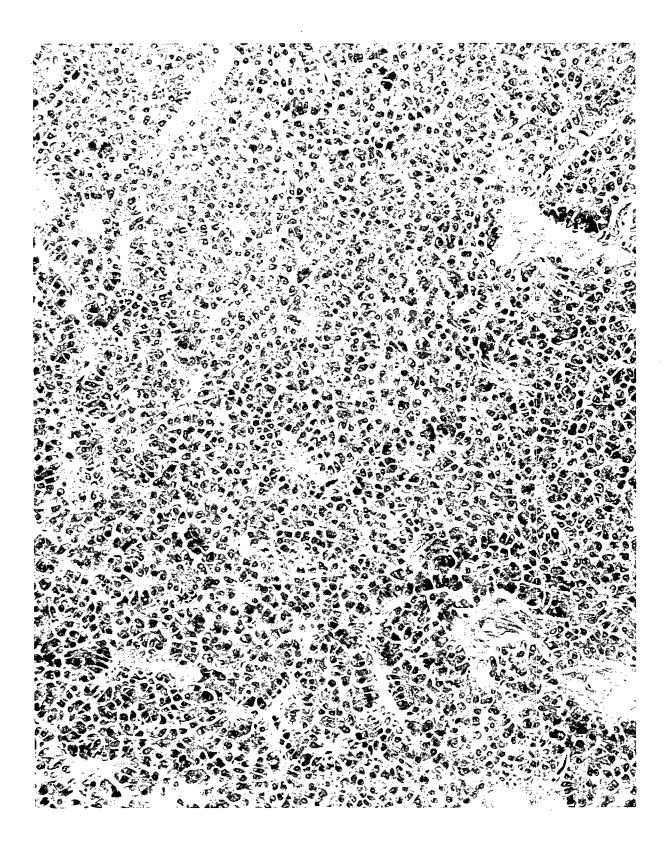


Figure 3. Hepatocellular carcinoma in liver of winter flounder from Boston Harbor. The hepatocytes are poorly formed and pleomorphic and the tumor has a solid appearance.

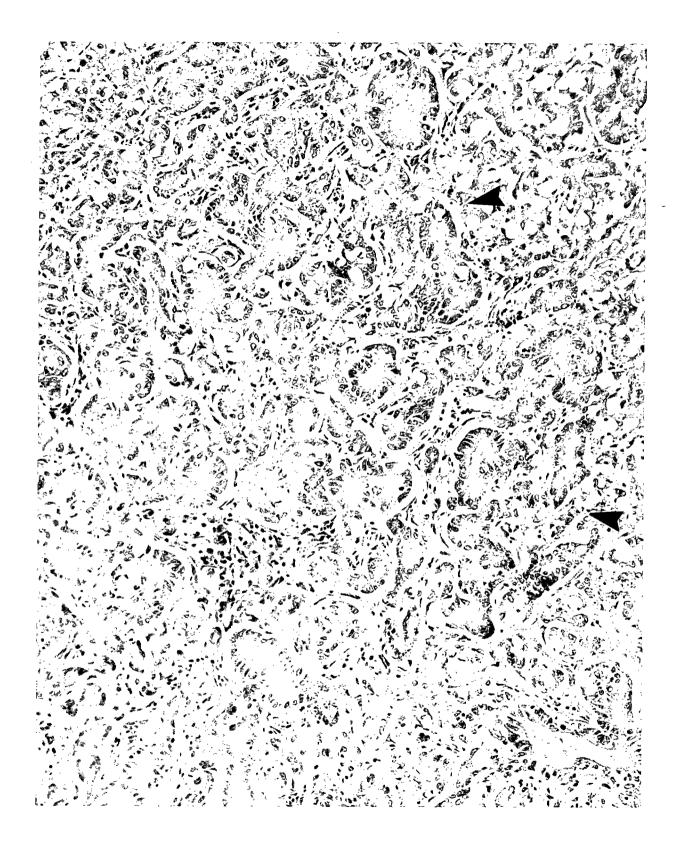


Figure 4. Cholangiocellular carcinoma. The proliferating cells form structures which resemble bile ducts. Arrows define the advancing edge of the lesion.

In the Puget Sound studies, it was noted that the concentrations of persistent organic chemicals, e.g., PCB's, were higher in fish than in sediments; however, concentrations of labile organic chemicals, e.g., PAH's, were higher in sediments than in fish. For this reason, tissue contaminant levels were not used as evidence of exposure to chemicals in sediments. The prevalence of liver neoplasms in Puget Sound English sole was positively correlated with the concentrations of PAH's in bottom sediments (10).

Recent studies of organic pollutants from Boston Harbor (4, 16) have revealed that levels are high. In one study (4) total PCB concentrations ranged from 70 to 330 ppb (mean 140 ppb). These values are comparable to those found in other polluted harbor sediments, but are lower than those found in the Arthur Kill and Newark Bay, New York, or New Bedford Harbor. PAH concentrations in Boston Harbor ranged from 2.4 to 880 ppm (mean 180 ppm). The high mean value is the result of a single high value at a sediment station sampled in the northern part of the harbor (near Deer Island). The range in harbor sediment PAH concentrations excluding this single value is 2.4-6.5 ppm (mean 4.4 ppm), approximately twice as high as PAH concentrations in the New York Bight apex. Computation of a Fossil Fuel Pollution Index (FFPI) (2, 3) which qualified the PAH values as either due to fossil PAH's (petroleum, coal) or combustion PAH's (fossil fuel combustion products, airborne particulates) indicates that there are multiple sources of sediment PAH's in the harbor. At the station near Deer Island, the sources of PAH's are approximately equal, i.e., both petroleum and combustion hydrocarbons.

Concentrations of coprostanol, a steroid whose presence in the harbor signifies contamination by sewage effluents, are high (1-16 ppm) and comparable to those found in the sewage sludge disposal site in the New York Bight. The highest coprostanol value (15.9 ppm) was obtained at the sediment station near Deer Island. Coprostanol/total steroid ratios also are highest at this station, signifying that the sterol composition of the sediment sample is approximating that of pure sewage sludge.

In the second study (16) substantially higher concentrations of PCB's and PAH's were found in Boston Harbor sediments. The mean concentration (1984, 1985) of total PCB's was approximately 8000 ppb. In sediments from western Long Island Sound, Narragansett Bay (RI), and eastern Long Island Sound mean total PCB's were approximately 170 ppb, 220 ppb and 5 ppb, respectively. The mean concentration of total PAH's in Boston Harbor sediments for the same period was approximately 17,000 ppb. In sediments from western Long Island Sound, Narragansett Bay (RI), and eastern Long Island Sound mean total PAH's were approximately 4400 ppb, 2200 ppb, and 100 ppb, respectively.

Few organic chemical analyses of Boston Harbor winter flounder hepatic tissues have been made. Recent determinations (dry weight) disclosed substantially elevated contaminant levels (total chlorinated pesticides = 1078 ppb; DDT = 827 ppb; PCB = 10,487 ppb)

(15). Total hepatic chlorinated pesticides in winter founder from western Long Island Sound, Narragansett Bay (RI), and eastern Long Island Sound were 282 ppb, 119 ppb and 101 ppb, respectively. Hepatic DDT levels were 105 ppb, 105 ppb and 49 ppb and hepatic PCB levels were 3222 ppb, 221 ppb and 1577 ppb, respectively in livers of winter flounder from the aforementioned areas. Livers of winter flounder from Great Bay, New Jersey, a relatively clean area, had a median PCB concentration (wet weight) of 0.89 ppm (18); however, median PCB concentrations in livers of winter flounder with fin rot from Sandy Hook/Raritan Bays, New Jersey, and the New York Bight apex, all degraded areas, were 4.2 ppm and 2.8 ppm, respectively. No analyses of hepatic PAH content of Boston Harbor winter flounder Hepatic tissue of winter flounder contains have been made. cytochrome P-450 dependent mixed-function oxydases (MFO) (22, 23); therefore, the PAH's themselves may not be detectable, but rather their metabolic derivatives. Concentrations of winter flounder hepatic microsomal MFO and other enzymes involved in election transport were generally less than in mammals (mice), but were capable of transforming PAH's to mutagenic metabolites. Biochemical studies conducted by the above authors (22) led them to conclude that, "the results indicate that coastal marine fishes may be at a risk to carcinogenic aromatic hydrocarbons in marine sediments".

Although relatively little research has been conducted to determine the cause of Boston Harbor winter flounder neoplasms, several years of research on hepatic carcinoma and other hepatic lesions of English sole from Puget Sound revealed positive correlations (p \leq 0.003) between the numbers of total hepatic lesions and both PAH's and metals in sediments. The number of neoplasms, however, was positively correlated only to PAH concentrations in sediments (10). Positive correlations were not found with chlorinated hydrocarbons. studies disclosed that the relative amounts of Additional benzo(a)pyrene (BAP) 7,8 diol (the penultimate carcinogen of BAP) metabolized by English sole from BAP are higher than in rat liver. Sole from polluted areas had concentrations of bile metabolites (fluorescence spectra compatible with naphthalene, phenanthrene, and BAP) that were substantially higher than in bile of sole from reference sites (9). Fish with liver lesions from polluted areas had significantly higher concentrations of metabolites with BAP-like fluorescence than fish without liver lesions. Although these studies seem to implicate PAH's as the cause of the hepatic neoplasms observed, the large number of chemicals in polluted sediments with unknown effects, and the complexity of the themselves, dictate biological processes caution in the interpretation of the data.

The Significance

The consequences of hepatic carcinoma for winter flounder populations presently are unknown. The majority of fish with hepatic neoplasms appear as robust as those without such neoplasms; however, objective estimates of "condition" of healthy and diseased fish have not been made. It also is not known if the ages of the two groups (diseased and healthy) being compared are the same since comparable length does not necessarily signify comparable age. The survival of individual diseased fish depends on many factors, including the extent of liver damage, the degree of toxicity resulting from decreased hepatic function, the impairment of other essential organs, whether metastasis occurs, and the degree of behavioral modification (the ability to capture prey and avoid predators may be compromised). Population effects are possible if large numbers of fish with hepatic carcinoma die and/or if the fecundity of individual fish is reduced. If gonadal tissues contain organic contaminants such as PAH's and PCB's, the viability of eggs and sperm, and vigor of zygotes may be reduced. Analyses for these chemicals in sexually mature, prespawning fish are needed. If elevated levels are found, laboratory experiments with eggs and sperm from Boston Harbor winter flounder should be conducted to evaluate the viability and normality of the larvae. It is possible that gonadal organic contaminants decrease the number of live larvae and/or increase the percentage of abnormal larvae in the progeny. If these phenomena occur, then the abundance of winter flounder, at least locally, may be affected.

High prevalence of hepatic carcinoma in Boston Harbor winter flounder may affect the acceptability of the fish as food, although no evidence presently exists substantiating that edible tissues (muscle) contain unacceptable levels of either PAH's or PCB's.

The most significant aspect of the high prevalence of hepatic carcinoma observed in Boston Harbor winter flounder is its relationship to poor environmental quality. Data on the prevalence of hepatic lesions, including carcinoma, in winter flounder from East Coast environments which have not been substantially degraded by anthropogenic activity (south shore of eastern Long Island, New York; Georges Bank; western Gulf of Maine) substantiate that the overall prevalence of microscopically detectable hepatic lesions is low and that no spontaneous carcinomas are present (Murchelano, unpubl.). As discussed above, both PAH's and PCB's are chemicals which are potential carcinogens. Although elevated levels of PCB's have been found in livers of Boston Harbor winter flounder, no experimental studies have implicated the specific role of PCB's in tumor induction in these fish. Planned analyses to evaluate the content of PAH metabolites in bile of fish with and without carcinomas should be informative. Although circumstantial evidence exists linking these (and other) hepatic lesions to environmental contaminants, determining the specific cause of neoplastic disease in any animal species is a complex, time-intensive process which is not likely to be resolved quickly. In spite of the lack of adequate experimental data implicating a specific environmental contaminant as the cause of the hepatic carcinomas observed in Boston Harbor winter flounder, there appears to be a relationship between environmental quality and fish health which suggests that fish are good sentinels of poor environmental quality.

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