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Pollutants and fish predator/prey behavior: A review of laboratory and field approaches

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Abstract Fish behavior can be altered by contaminants. There is an extensive literature on laboratory behavioral assays, with many chemicals impairing feeding or predator avoidance. However, there is not extensive work on fishes that live in contaminated environments. Therefore, we then review our recent research on feeding and trophic relations of populations from contaminated estuaries compared with relatively unpolluted sites. The mummichog *Fundulus heteroclitus*, is a non-migratory fish; those from more contaminated areas are poor predators and slower to capture active prey (grass shrimp, *Palaemonetes pugio*). In the field, they consume much detritus and sediment, which is not nutritious. They are less active than fish from cleaner sites and more vulnerable to predation. They have altered thyroid glands and neurotransmitter levels, which may underlie altered behaviors. Fish from the reference site kept in tanks with sediment and food from the polluted site showed bioaccumulation and reduced prey capture after two months, although fish from the polluted site did not show significant improvement when maintained in a clean environment. Poor nutrition and predator avoidance may be responsible for their being smaller and having a shorter life span than reference fish. Bluefish *Pomatomus saltatrix*, are a marine species in which the young-of-the-year spend their first summer in estuaries. We found bioaccumulation of contaminants and reduced activity, schooling, and feeding in young-of-the-year bluefish from a relatively unpolluted site that were fed prey fish from a contaminated site. They also had altered thyroid glands and neurotransmitter levels. Many field-caught specimens had empty stomachs, which is rare in this species. In the fall, when they migrate back out to the ocean, they are smaller, slower, and more likely to starve or to be eaten than those that spent their summer in cleaner estuaries [*Current Zoology* 58 (1): 9–20, 2012].

Keywords Predator, Feeding, Prey, Pollution, Activity, Trophic

Behavior is a particularly sensitive measure of an organism's response to stresses, including environmental contaminants. Noticeable changes in behavior can be found at concentrations of chemicals that are orders of magnitude below those that can cause mortality (LC_{50}), bioassays that are still used in toxicity testing (Little and Finger, 1990; Gerhardt, 2007). Chronic toxicity tests may include growth effects but generally use non-living foods during the tests. Even when live food is used, the food density is high so the fish are not challenged to locate, pursue, and capture prey as they would be under natural conditions. In addition to being more sensitive, sublethal responses like behavioral changes are far more likely to occur in nature (Sandheinrich and Atchison, 1990; Scott and Sloman, 2004) and behavioral responses can have ecological effects at the population and community level.

1 Links to Higher and Lower Levels of Biological Organization

Fish have been used frequently in behavioral toxicity testing, as many of their behaviors can be quantified in a laboratory setting. The kinds of behaviors that have been studied have included attraction/avoidance behavior, swimming activity, reproductive behavior, schooling, feeding/prey capture, and predator avoidance, and reproductive behavior. The ecological consequences of changes to these behaviors can be great. For example, reduced feeding can affect growth rate and thereby affect the population. Reduced predator avoidance ability can increase mortality rates and also affect the population size and size-structure. Prey exposed to contaminants may fail to detect predators, take more risks in leaving refugia to find food, have a poor fast start performance, reduced stamina, inability to school, altered activity patterns and increased conspicuousness (hyperactivity), all leading to increased predator vulnerability (Mesa et al., 1994; Scott and Sloman, 2004). If individuals with higher concentrations of contaminants are easier for predators to capture, this can also facilitate the transfer of the contaminants to higher trophic levels.

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Altered predator/prey interactions involve organisms from two trophic levels and can cause changes in populations of predators, prey, or both and thus affect the community. In contaminated environments, both predator and prey are exposed, and one may be more susceptible than the other.

Links can also be established between behavioral changes and underlying mechanisms. Developmental exposures to endocrine disrupting chemicals like xenoestrogens can alter neurodevelopment in fish (Kishida et al., 2001). Physiological changes such as altered neurotransmitter levels or hormones can affect behavior (Scott and Sloman, 2004; Rademacher et al., 2003). Altered behavior may result from nervous system damage, for example neurotransmitters are sensitive to contaminant exposure and can affect behavior. Hg decreased levels of serotonin (5- HT) in brains of striped mullet *Mugil cephalus* and tilapia *Oreochromis mossambicus*, which was associated with loss of motor control (Thomas et al., 1981; Tsai et al. 1995). Neurotoxicants that alter dopaminergic systems can affect swimming behavior and memory in fishes (Panula et al., 2006). Some pesticides target the enzyme cholinesterase that breaks down the neurotransmitter cholinesterase, and mosquito fish *Gambusia affinis* exposed to sublethal concentrations of the pesticide chlorpyrifos for 20 days reduced their locomotor behavior and swimming speed and accumulated acetylcholine at synaptic junctions (Rao et al., 2005). There is also an abundant literature linking behavioral changes in fishes to alterations in the thyroid gland (Clotfelter et al., 2004; Cooley et al., 2001; Morin et al., 1989). Thyroid hormones are essential for normal brain development, and altered thyroid hormones can alter neurobehavioral development including circadian rhythms of neurotransmitters (Spieler et al., 1995). Altered thyroid function can affect activity and feeding in fish (Castonguay and Cyr, 1998), and toxicant exposure can alter levels of thyroid hormones (Bleau et al., 1996). When stimulated by TSH from the pituitary gland, the epithelial cells surrounding the follicle increase in height, and overall the follicles enlarge. Abnormally enlarged follicles, the fish equivalent of a goiter, reflect deficiencies in thyroid hormones, which trigger the pituitary to secrete excess TSH, which stimulates the thyroid follicles. Such follicles have been seen in fish from the Great Lakes (Leatherland, 1994).

2 Laboratory Exposures to a Variety of Chemical Stressors

Chemicals can differ in the types of behaviors that they affect, depending on their mode of action (Robinson, 2009). Some of the chemicals that have been investigated include the pesticides DDT, carbaryl (Weis and Weis, 1974a and b), carbofuran (Bretaud et al., 2002), sumithion (Bull and McInerney, 1974), esfenvalerate (Little et al., 1993), methyl parathion (Henry and Atchison, 1984) chlorpyrifos and others (Rice et al., 1997) the herbicides diquat and simazine (Drummond and Carlson, 1977[1]), pentachlorophenol (Brown et al., 1987), polychlorinated biphenyls and tributyltin (Schmidt et al., 2004), hydrocarbons like 4-tert-octylphenol (Gray et al., 1999). Endocrine-disrupting chemicals frequently have deleterious effects on courtship and reproductive behaviors (Clotfelter et al., 2004; Saaristo et al., 2010). Some toxins are produced naturally by organisms like some species of dinoflagellates. The dinoflagellate *Alexandrium fundyense* produces saxitoxin, which causes paralytic shellfish poisoning in humans. Exposure to very low numbers of cells of this species (either directly in the water or via copepod vectors) can also produce behavioral effects in larval and juvenile fishes (Samson et al., 2008).

Focusing particularly on predator/prey behavior, Bull and McInerney (1974) found a decreased feeding rate in coho salmon *Oncorhynchus kisutch* after exposure to 0, 1 ppm of the pesticide sumithion. Brown et al. (1987) found that exposure to as little as 67 μg/l pentachlorophenol reduced feeding in largemouth bass *Micropterus salmoides*. Little et al. (1990) examined spontaneous swimming activity, swimming capacity, feeding behavior, and vulnerability to predation in rainbow trout *Oncorhynchus mykiss* after 96-hr exposures to sublethal concentrations of six agricultural chemicals: carbaryl, chlordane, dimethylamine salt of 2,4-dichlorophenoxyacetic acid (2,4-DMA), tributyl phosphorotrithioate (DEF1), methyl parathion, and pentachlorophenol. Effects on specific behaviors varied with the particular contaminant and its concentration. Feeding behavior was inhibited most by DEF1, 2, 4-DMA, and methyl parathion. In contrast, vulnerability to predation was increased most by carbaryl and pentachlorophenol. Gregg et al. (1997) investigated effects of

[1] Drummond RA, Carlson RW, 1977. Procedures for measuring cough (gill purge) rates of fish. US Environmental Protection Agency Report No. EPA 600/3–77–133.

diesel-oil contaminated sediments on feeding of the darter goby, a bottom feeder. Diesel fuel reduced feeding rate by 50%–100% in all but the lowest dosage examined; feeding rate after exposure to suspended sediments above 200 mg PAH kg dry sediment^{-1} was significantly reduced relative to uncontaminated controls.

Metal pollutants have also been studied in depth, including lead (Weber, 1996; Weber et al. 1997) and mercury, which are both well-known neurotoxicants. Other metals have also been studied. For example, Sullivan et al. (1978) reported increased vulnerability to largemouth bass predation in fathead minnows *Pimephales promelas* after exposure to cadmium.

Some years ago, Atchison et al. (1987) reviewed effects of metals on fish behavior. We will not review all the studies, but will emphasize mercury studies, as an example. There have been many studies of behavioral effects of mercury on adult fishes, focusing on activity level, prey capture, and predator avoidance, behaviors that are critical to survival and that have ecological relevance. For example, Weis and Khan (1990) found that exposure of adult mummichogs to 10 μg l $^{-1}$ of either $HgCl_2$ or methylmercury (MeHg) for a week caused reductions in feeding rate. Similarly, fathead minnows *Pimephales promelas* were exposed to mercury for ten days then tested for foraging efficiency, capture speed, and the ability to learn information regarding habitat characteristics (Grippo and Heath, 2003). Comparisons between control fish and fish from the two highest exposure groups (6.79 and 13.57 μg/L $HgCl_2$) revealed consistent deficits in foraging efficiency and capture speed. However, no effects on learning were detected. Kania and O'Hara (1974) used a test system with a shallow water refuge that allowed mosquitofish *Gambusia affinis,* to avoid predation by bass, which were restricted to the deeper water. After 24 h of exposure to 10 pg 1^{-1} of mercury, vulnerability of mosquito fish to predation increased significantly. Golden shiners *Notemigonus chrysoleucas* were fed diets with different levels of Hg and then exposed to a model avian predator and videotaped (Webber and Haines, 2003). Fish fed higher levels of Hg had greater dispersal, took longer to return to pre-exposure activity, and schools had greater areas after return to pre-exposure activity, all of which would increase their vulnerability to predation.

3 Developmental Exposures and Delayed Effects

In addition to studies on adults, there have been studies on developing fishes. Behavioral development occurs in association with the development of the nervous system and developing fishes are generally more sensitive to environmental contaminants than adults. Embryonic exposures to various chemicals at concentrations that do not produce anatomical malformations (teratogenic effects) may nevertheless produce functional deficits at later stages in life (Choi, 1983). There has been some study of delayed effects in fishes. Weis and Weis (1995a) examined behavior of mummichog larvae after embryonic exposure to levels of MeHg lower than those that produced anatomical malformations (5 and 10 μg 1 $^{-1}$). After hatching, larvae were maintained in clean water. Prey capture ability of early larvae was impaired, but approximately one week after hatching the prey capture ability was comparable to controls, showing that this was a temporary effect. The exposure may have caused retardation of neurological development that was subsequently compensated for, and therefore no long term effects were produced. Larvae that had been exposed as embryos were also more susceptible to predation by grass shrimp *Palaemonetes pugio* or adult mummichogs (Weis and Weis, 1995b). Larvae that had been exposed to MeHg as embryos generally had increased activity levels, which could serve to attract a predator, resulting in increased capture (Zhou and Weis, 1998). After exposure at both embryonic and larval stages, effects on prey capture were greater than embryonic exposure alone (Zhou et al., 2001). Social behavior was also impaired in larvae that had been exposed as embryos (Ososkov and Weis, 1996). When in groups, these larvae had poorer swimming coordination resulting in a greater frequency of collisions than controls. This behavior may have been caused by effects on the lateral line system, as seen by Koltes (1985) in copper-exposed silversides. This response also disappeared by four weeks after hatching, indicating that the effect was reversible.

In contrast, Samson et al. (2001) exposed zebrafish *Danio rerio* embryos to 0, 5, 10 or 15 μg MeHg l^{-1} for varying periods of time. Larvae were kept in clean water. Continuous embryonic exposure to 15 μg/l resulted in delayed mortality: by day 3, post-hatch activity was reduced; by day 5, post-hatch larvae were moribund with faint heartbeat, edema and vertebral flexures. Most embryos were dead by day 6 post-hatch. Shorter exposures to 15 μg l^{-1} caused reduced activity and impaired prey capture; by four days post-hatch, larvae showed signs of delayed mortality. Continuous exposure or exposure during the last 24 h of development to 10 μg l^{-1}

reduced activity, which did not improve. Prey capture was impaired after continuous embryo exposure to10 μg l^{-1}, even after four days in clean water. Single-day embryonic exposures to 10 μg l^{-1} did not affect activity or prey capture of larvae, however. Thus, in contrast to the mummichog experiments, the delayed effects seen in zebrafish did not appear to be reversible, at least under the conditions of the experiments performed.

Smith et al. (2010) exposed zebrafish embryos from 2 to 24 h post fertilization to MeHg alone (up to 0.30 μM), selenomethionine alone (up to 0.30 μM), which is supposed to protect against mercury, and combinations of the two. Learning (food delivery on alternating sides of the aquarium) was tested in adults, four months after developmental exposure. Low levels of MeHg (< 0.1 μM) exposure delayed learning, and fish exposed to higher concentrations were unable to learn the task. Furthermore, exposure to selenomethionine did not protect against these effects. This is in contrast to effects seen by Weber et al. (2008) in which selenium did protect against effects of MeHg on a visual startle response. Exposure of salmon parr to dietary MeHg (5 or 10 mg Hg kg^{-1} DW) for four months resulted in altered enzymes in the brain and behavioral changes. Fish fed 10 mg kg^{-1} MeHg had pathological damage,, significantly reduced neural enzyme activity (5-fold reduced monoamine oxidase, MAO, activity), and reduced overall post-feeding activity behavior (Berntssen et al., 2003).

Delayed effects may also occur after exposure of older stages. Alvarez et al. (2006) fed adult Atlantic croaker MeHg-contaminated food at three levels for one month. Fish were then induced to spawn and MeHg levels in the eggs were measured. Behavioral performance of exposed and control larvae was measured at different developmental stages. Behaviors analyzed included routine swimming speed and startle response (responsiveness, reactive distance, response distance, response duration, average response speed, and maximum response speed). Maternally-transferred MeHg impaired these behaviors, which were considered survival skills.

Some effects may be manifested years after the exposure. Fjeld et al. (1998) found very delayed effects. Embryos of grayling *Thymallus thymallus* were exposed to MeHg during the first 10 days of development. Three years later there was impaired feeding efficiencies and reduced competitive abilities in fish from groups that had accumulated Hg > 0.27 μg g^{-1}. Exposed fish were 15%–24% less efficient than controls, which caught 2–6 times as many prey items as exposed fish. Therefore, the embryonic exposures caused irreparable alterations in the brains of the fish. The concentration of 0.27 μg g^{-1} Hg, which appeared to be a threshold for this effect, is seen in eggs of piscivorous fishes in lakes with substantial atmospheric deposition of Hg.

Thus, it is clear that behavioral responses are very sensitive to low levels of a variety of environmental contaminants, and can have major ecological significance. Nevertheless these responses are not generally incorporated into ecological risk assessments or considered by regulatory agencies (Robinson, 2009). One reason is the lack of standardization of behavioral toxicity tests, and another is that the existence of delayed effects could make short term tests inaccurate and inadequate. There is also a general lack of field validation of these responses (Robinson, 2009). Prey detection and capture are much more complex and challenging in nature than in the laboratory. Furthermore, in the field, both predator and prey have been exposed and potentially impaired, yet few studies examine effects on both the predator and the prey simultaneously. Actual examination and quantification of fish behavior (other than avoidance) in the field is difficult. Newer technologies such as telemetry, ultrasound and infrared light beams, and “on-line” biomonitors may help in field validation and aid in adoption in ecological risk assessment. A confounding factor for a field validation is the presence of many contaminants at most field sites studied. It is rare to find a place that has only one contaminant. This may impede incorporation into ecological risk assessment frameworks. Nevertheless, it is of great interest from an ecological standpoint.

4 Field Validation: Studying Fish in the Field

There have been some studies in which fish were exposed and tested in the field to examine their performance. Hartwell et al. (1987) exposed fathead minnows to a mixture of metals (copper, chromium, arsenic, and selenium) and tested them for avoidance in an artificial stream. Control fish avoided much lower concentrations of metals than the exposed fish. Weber and Bannerman (2004) constructed a mobile field station into which stream water was pumped. Study sites were located in low, intermediate and high percent impervious surface watersheds, in which nonpoint source pollutant runoff would be low, medium and high. Stream water entered flow-through aquaria containing breeding pairs of fathead minnows. Reproductive behaviors including time spent conducting nest or egg care activities, spawning attempts, development of male secondary sexual characteristics, and male chasing of females were all reduced, related to the percent impervious surface in the watershed.

5 Field Validation: Fish from Contaminated Sites Studied in the Laboratory

An easier approach is to bring organisms into the lab from both contaminated and relatively unpolluted reference sites, and compare their behaviors under controlled conditions. Sopinka et al. (2010) compared aggression and dominance hierarchy formation in round gobies *Neogobius melanostomus* from polluted and cleaner sites in Lake Ontario and found that while stable hierarchies formed between pairs of fish from the cleaner site, dominance was less obvious among fish from the more contaminated area.

We now review experiments and results of predator/prey behavior in two species of fishes living in the contaminated urbanized estuaries of northern New Jersey. Impaired feeding behavior may be validated by examining the gut contents of specimens collected from the field. We have studied feeding behavior and predator/prey relations in fish from contaminated and clean estuaries and examined population and community level consequences. We collected organisms from the field and brought them into the lab to study their prey capture and predator avoidance behavior. We have also "switched" animals from the clean to the polluted site and vise versa or placed them in simulated environments in the laboratory, to see if the behaviors change to resemble that of the organisms from that site, which would suggest that behavioral differences are reversible and caused by the environment, rather than non-reversible or genetic differences among populations. It is also possible that environmental stressors trigger changes in gene expression that may lead to behavioral changes.

Mummichogs Fundulus heteroclitus

Our initial work was with common killifish (mummichogs *Fundulus heteroclitus*), a small estuarine fish that stays within a limited area for its entire life. Those from contaminated sites (initially Piles Creek off the Arthur Kill in northern New Jersey) were less active, less able to capture prey (grass shrimp *Palaemonetes pugio*), than those from reference sites in Tuckerton NJ. Piles Creek is a highly industrialized site near chemical companies, a power plant, and a sewage treatment plant, with elevated levels of metals and organic pollutants. Contaminants in the system include PCBs, PAHs, dioxins, chlorinated hydrocarbon pesticides, and metals such as copper, lead, zinc, cadmium, chromium, and mercury. These are typical contaminants in urbanized systems. In the 1990s, sediment values in Piles Creek (PC) were around 200 ppm for Pb and about 5.0 ppm for Hg. These values are high, but reflect a considerable decrease from levels present decades earlier (Bopp et al., 2006). Tuckerton is in a non-developed area that is part of a wildlife refuge, and has much lower levels of contaminants. While not "pristine", it has been used as a "reference site" in many studies. It is about 125 km south of Piles Creek and has slightly higher temperatures and higher salinity. Piles Creek fish also had reduced condition. Previous work had shown that the Piles Creek population had reduced life span and growth (Toppin et al., 1987); reduced feeding could be partly responsible for reduced growth and condition. After fish from Tuckerton were kept in aquaria in the lab with Piles Creek sediments and fed food from Piles Creek, their prey capture ability decreased to be equal to that of the Piles Creek fish and the level of Hg in their brains increased to that of the Piles Creek population. When Piles Creek fish were maintained in clean water, sediments, and food for six weeks, their prey capture ability increased slightly but not significantly, and their brain Hg did not decrease (Smith and Weis, 1997). The correlation of behavior with mercury does not mean that this contaminant is the definite cause of the behavioral impairment as there are many other contaminants at the site, including lead and PCBs, that are also known neurotoxicants and could contribute to behavioral deficits (Weber, 1996; Weber et al., 1997; Berger et al., 2001). Stomach contents of field-caught Piles Creek fish had far more sediment and detritus (which is not nutritious for them) and less live prey than Tuckerton fish (Smith and Weis, 1997; Weis et al, 2001a). Piles Creek fish also had reduced activity levels and were more vulnerable to predation by blue crabs than fish from TK. The poor diet and poor predator avoidance could help explain earlier observations that these fish do not grow as well or live as long as fish from reference sites (Toppin et al., 1987).

We noted abnormal neurotransmitter levels (reduced serotonin) in Piles Creek fish, which could be partly responsible for the altered behavior (Smith et al., 1995). Exposure of mummichog larvae to MeHg produced variable alterations in neurotransmitter levels that were inconsistent over time and were different in the two populations both before and after exposure (Zhou et al., 1999b). Newly hatched Piles Creek larvae, which were not behaviorally impaired, had levels of serotonin comparable to those in fish from the reference population, but did have higher levels of dopamine and its metabolites.

Piles Creek mummichogs also have greatly expanded and irregularly shaped thyroid follicles, increased epithelial cell thickness, and a trend of reduced levels of T3, which is the active form of the thyroid hormone in fish (Zhou et al., 1999a). The expanded thyroid follicles and increased epithelial cell height are a response to increased levels of TSH from the pituitary, responding to lower levels of thyroid hormones; thyroid hormones affect activity levels (Castonguay and Cyr, 1998)

When populations from many different sites were investigated for prey capture, their predatory ability on grass shrimp was found to be related to sediment and tissue levels of contaminants (Weis et al., 2001b). Using laboratory prey capture experiments, the number of grass shrimp captured was highest by fish from the sites with the lowest levels of contamination. Gut contents of field-collected fish revealed that grass shrimp made up the largest proportion of the diet in the "cleaner" sites whose fish had the highest capture rates in the laboratory. However, since the levels of contaminants at a site were highly correlated with each other, the role of specific contaminants on the behavior could not be determined.

In more recent work, Goto (2009), examined mummichogs from another polluted site near the Arthur Kill in Staten Island and found that the fish had 2-3x less food in their stomachs than fish from a reference site. The grass shrimp ingested by these fish were half the size of those eaten by the reference population, and the polluted site fish ate more polychaete worms instead. They apparently replaced decapods in their diet with polychaetes (instead of sediment and detritus, as done by Piles Creek fish). These fish appear to be less impacted than Piles Creek fish and eat a better diet, since they can capture more grass shrimp, albeit smaller ones that are probably easier to catch, handle and digest. Being less impaired is consistent with contaminants causing poor prey capture (Weis et al., 2001b); the sediments in the Staten Island creek are less contaminated than those at Piles Creek (Goto, 2009).

Grass shrimp from Piles Creek were captured by mummichogs just as frequently as shrimp from the reference site (Tuckerton) (Smith and Weis, 1997), showing that the predator avoidance ability of the shrimp was not impaired by the conditions at Piles Creek. Bass et al. (2001 studied the *Palaemonetes pugio* populations in Piles Creek and the reference site Tuckerton, and found that the grass shrimp were both larger in size and more numerous at the polluted site than Tuckerton. However, laboratory studies in which juvenile shrimp from both populations were maintained in aquaria with sediments and water from both sites (Piles Creek on Piles Creek, Piles Creek on Tuckerton, Tuckerton on Piles Creek and Tuckerton on Tuckerton) showed that Piles Creek shrimp did not grow faster than Tuckerton shrimp, and that maintenance in an aquarium with Piles Creek sediment did not enhance the growth of either population of shrimp. The larger size and greater population density of the shrimp at the polluted Piles Creek is consistent with reduced top-down control, since their major predator, mummichogs, are not only poor predators (Smith and Weis, 1997), but are both smaller in size and less abundant at Piles Creek than Tuckerton (Bass et al., 2001).

Bluefish Pomatomus saltatrix

After bluefish spawn in the Atlantic Ocean in the spring, the young move into estuaries. Young-of-the-year bluefish, voracious piscivores with high lipid content, may reside in contaminated estuaries during their first summer before moving back to the ocean in the fall. As a result of this exposure they may bioaccumulate high levels of lipophilic contaminants including PCBs, pesticides and MeHg that biomagnify up the food web. We hypothesized that bluefish from contaminated estuaries may have reduced feeding and growth compared to those that spent the summer in cleaner environments, and may be less competitive. Young-of-the-year bluefish were collected from another contaminated site in industrialized northern NJ, the Hackensack Meadowlands and from Tuckerton in the early fall and their length, weight, stomach contents, and PCB and mercury levels measured. The Hackensack River runs approximately 72 km through New York and New Jersey and empties into Newark Bay. The river flows through and drains the New Jersey Meadowlands, and is surrounded by a highly urbanized region. Two centuries of industrial pollution have left the water and sediment severely polluted and, while conditions have improved over the past few decades, urban runoff, sewage treatment plant discharges, sewer overflows and sanitary landfills continue to impair the habitat quality. Hackensack fish were significantly smaller overall (Fig. 1), had elevated levels of contaminants, and many (73%) of the fish had empty stomachs (Candelmo, 2010). The 27% with food in their stomachs is considerably lower than the percentages of 63%–92% reported in numerous other studies of young-of-the-year bluefish (Juanes and Conover 1994; Buckel and Conover, 1997; Buckel et al., 1999; Gartland et al., 2006). Reduced feeding could be responsible for their smaller size. The high frequency of empty stomachs does not reflect scarcity of these prey species at Hackensack, as they were caught readily in seines and trawls. Reduced feeding could be responsible for the smaller size of the bluefish. The mean PCB concentration in Hackensack bluefish was 2–3 fold greater than that of the menhaden *Brevoortia tyrannus* and mummichogs *Fundulus heteroclitus* found in their stomachs, showing biomagnification taking place from prey to predator. In contrast, bluefish from Tuckerton were significantly larger (14.9% longer and 44.4% heavier) than Hackensack bluefish; and most (75%) had prey (mostly mummichogs and menhaden) in their stomachs. They had lower levels of contaminants in their tissues although their mean PCB body burden was six-fold greater than that of the mummichogs and menhaden in their stomachs (Candelmo, 2010).

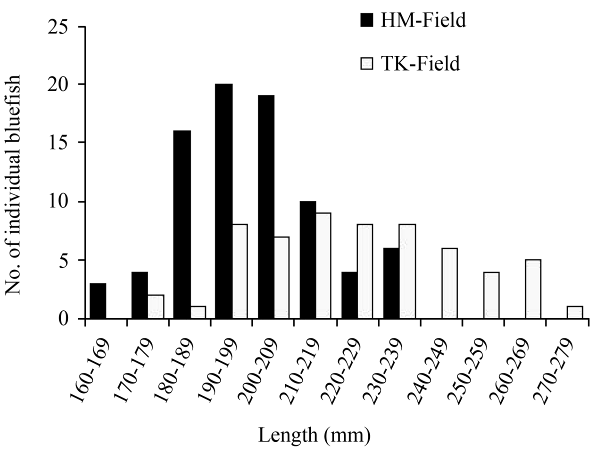

Fig. 1 Length frequency distribution of field collected bluefish in the fall after spending the summer in Hackensack (HM) or Tuckerton (TK)

The levels of PCBs and DDT in menhaden and mummichogs found in the Hackensack bluefish stomachs was higher than that of these fish caught in trawls and seines, suggesting that the more contaminated fish were easier for predators to capture (Fig. 2). This supports our earlier laboratory data that contaminated mummichogs were impaired in predator avoidance and were easier for blue crabs to capture. If bluefish are preferentially foraging on the more contaminated prey, higher levels of contaminants are being trophically transferred.

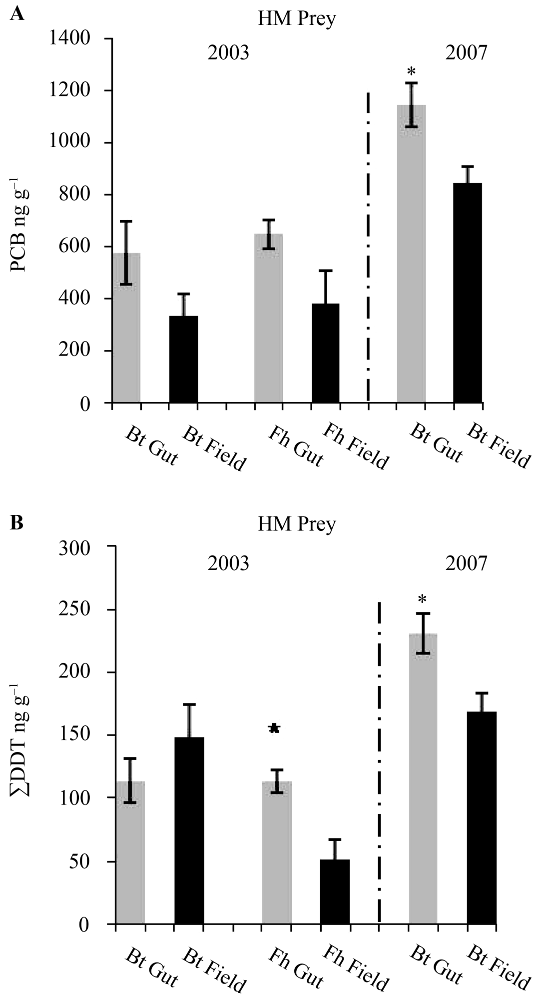

Fig. 2 PCB and DDT concentrations in prey fish

A. PCB content of mummichog (*Fundulus heteroclitus* = Fh) and menhaden (*Brevoortia tyrannus* = Bt) collected from bluefish stomachs and those captured from the field at HM and TK. Bars with asterisks indicate significant difference of PCBs in prey fish from gut vs fish from the field. **B.** DDT content of mummichog (Fh) and menhaden (Bt) collected from bluefish stomachs and those captured from the field at HM and TK. Bars with asterisks indicate significant difference of DDT in prey fish from gut vs fish from the field.

Young-of-the-year bluefish were collected from Tuckerton at the beginning of the summer for a laboratory experiment in which they were fed daily for four months with menhaden and mummichogs from either Tuckerton or Hackensack. After four months in the laboratory, the bluefish were measured, tested for swimming activity level and feeding rate, and were sacrificed for chemical analysis. Hackensack-fed bluefish displayed significantly reduced feeding (Fig. 3), spontaneous activity, and growth compared to the Tuckerton-fed bluefish. Tuckerton-fed fish had about twice the swimming speed as Hackensack-fed fish (Fig. 4) (Candelmo et al., 2010). The Hackensack-fed fish had poor schooling behavior, frequently leaving the school and swimming up and down the sides of the test aquarium. The reduced feeding and growth support the data on size and empty stomachs seen in the field-collected fish. The Hackensack-fed fish also contained highly elevated levels of PCBs, pesticides, and total mercury that had been trophically transferred to them during the feeding experiment; these levels were higher than those of the field-collected fish in the fall. While total mercury was measured, previous studies and the fact that concentrations biomagnified considerably suggest that it is mostly MeHg.

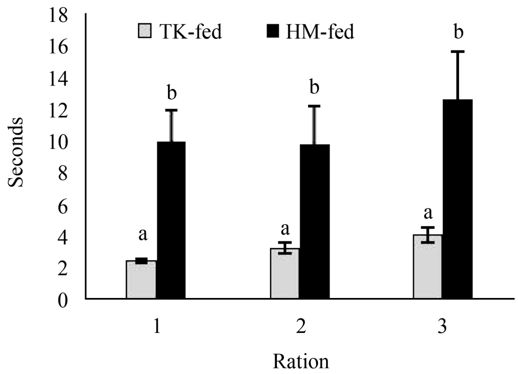

Fig. 3 Time taken for the control (TK-fed) lab bluefish and the exposed (HM-fed) bluefish to consume the ration of mummichogs

Trial 1 was the first ration thrown in, trial 2 was the second ration thrown in after the first was consumed, trial 3 was the third ration. Letters represent mean values that are significantly different from each other.

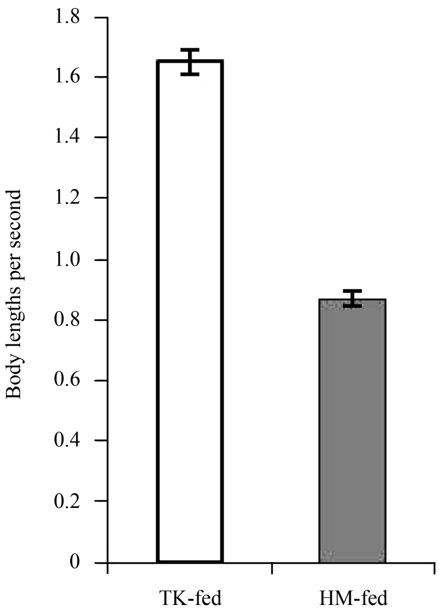

Fig. 4 Mean swimming rate of the control (TK-fed) and contaminated (HM-fed) bluefish

We had previously found altered thyroid and neurotransmitters in the contaminated Piles Creek mummichogs (Smith et al., 1995; Zhou et al., 1999a, b). We investigated neurotransmitters and the thyroid glands of bluefish from Tuckerton and Hackensack. Bluefish from Hackensack and the Hackensack-fed laboratory fish had enlarged, irregular thyroid follicles, lined with thickened epithelial cells, and were often depleted in colloid compared to fish from Tuckerton. This was similar to the condition seen earlier in the mummichogs. Neurotransmitter levels were measured in the brain of bluefish from both sites. The dopamine (DA) metabolites dihydroxyphenylacetic acid (DOPAC) and homovanillic acid (HVA) were significantly lower in Hackensack fish than in Tuckerton reference fish. In addition, the dopaminergic activity level (ratios of DOPAC: DA and HVA: DA) of Hackensack bluefish were significantly lower than those of Tuckerton fish. These results suggest disruption to both the synthesis and metabolism of dopamine. This is in contrast to alterations of serotonin seen in the Piles Creek mummichogs (Smith et al., 1995). These neurological and hormonal alterations may underlie the observed altered behavior. Exposure to endocrine-disrupting and neurotoxic contaminants including PCBs and mercury appears to be associated with biochemical and physiological changes that result in behavioral impairments during a period of rapid growth and development of bluefish in contaminated estuaries.

There may be significant ecological consequences of the altered behavior. In the fall, these fish will migrate back to the ocean and migrate south, along with fish that had spent the summer in cleaner estuaries. Their impaired feeding and poor schooling behavior, along with their smaller size, would appear to make them more likely to starve or to be captured by a predator. The altered behavior and reduced growth may have a substantial impact on their fitness, migratory competence, overwinter survival, and recruitment success. Size-dependent winter mortality can play a key role in year class strength and overall stock size; in general, larger individuals are more likely to survive their first winter (Conover, 1992; Hurst and Conover, 1998). In addition, during winter migration and their prolonged period of reduced feeding, lipid utilization may cause PCBs and MeHg stored in lipids to be redistributed to sensitive organs such as liver and brain and lead to further detrimental effects, as reported in other fish (Boon and Duinker, 1985; Jørgensen et al., 1999). A major unanswered question is whether, once in the ocean, they can depurate enough of their toxicant body burden and improve their fitness before they starve or are captured by a predator. Even with depuration, the neurotoxic effects may be permanent due to effects on neuronal development, including growth and differentiation.

6 Summary and Conclusions

In two very different kinds of fish species, one resident (mummichog *F. heteroclitus*) and one migratory (bluefish *P. saltatrix*), inhabiting two different contaminated estuaries of northern NJ, we have seen similar behavioral and ecological alterations. Both species have reduced feeding/prey capture due to reduced appetite or motivation to feed. In the contaminated field sites, *F. heteroclitus* eats largely detritus and sediment of poor nutritional value, while many *P. saltatrix* are found with empty stomachs. Both species have reduced growth at the contaminated sites, which is a logical outcome of decreased feeding. Both species appear to have impaired predator avoidance ability as well, as seen in *F. heteroclitus* in the lab experiment with blue crab predators, and in the field where specimens with higher contaminant levels were found in bluefish stomachs than swimming in the estuary. More contaminated bluefish demonstrate impaired schooling behavior and slower swimming in the lab, which is likely to make them more susceptible to predation. Impaired predator avoidance by more contaminated individuals is a mechanism for increasing trophic transfer of contaminants up the food chain. Underlying changes in the thyroid glands have been seen in both species, which show expanded follicles and thicker epithelial cells, an indication of lowered thyroid hormones. Altered neurotransmitter levels have been noted in both species, but the particular neurotransmitter affected in the mummichogs is serotonin, while in bluefish it is dopamine. Why different neurotransmitters were affected is an open question, but may be due to species differences or the concentrations of particular contaminants at Piles Creek vs Hackensack. Exposure to PCBs has been associated with dopamine disruption, while mercury exposure is associated with serotonin. Hackensack bluefish were found to accumulate substantially higher concentrations of neurotoxic ortho-substituted PCB congeners than Hackensack mummichogs, which could explain the lack of response from dopamine in the mummichogs (Candelmo, 2010). Additional studies on other fish species and other contaminated sites are needed to learn whether the similar suite of responses seen in mummichogs and bluefish is typical of fishes in contaminated environments, or whether these behavioral changes may be responses to the particular mix of industrial contaminants found in these sites in northern New Jersey.

A great deal has been learned from the laboratory assay approaches discussed in the first part of this review. Many chemicals have been shown to alter (impair) fish predator and prey behaviors. If similar changes occur in populations in contaminated environments, they can have effects at the community level, since they affect organisms at two trophic levels. In many cases, more highly contaminated prey are more likely to be captured, resulting in increased trophic transfer to predators. Much more research remains to be done on how fish behaviors are altered in the "real world" of polluted environments and how trophic relationships, and therefore community ecology, can be altered by pollution-related changes in fish feeding and predator avoidance.

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