



Intersex fish

Endocrine disruption in smallmouth bass

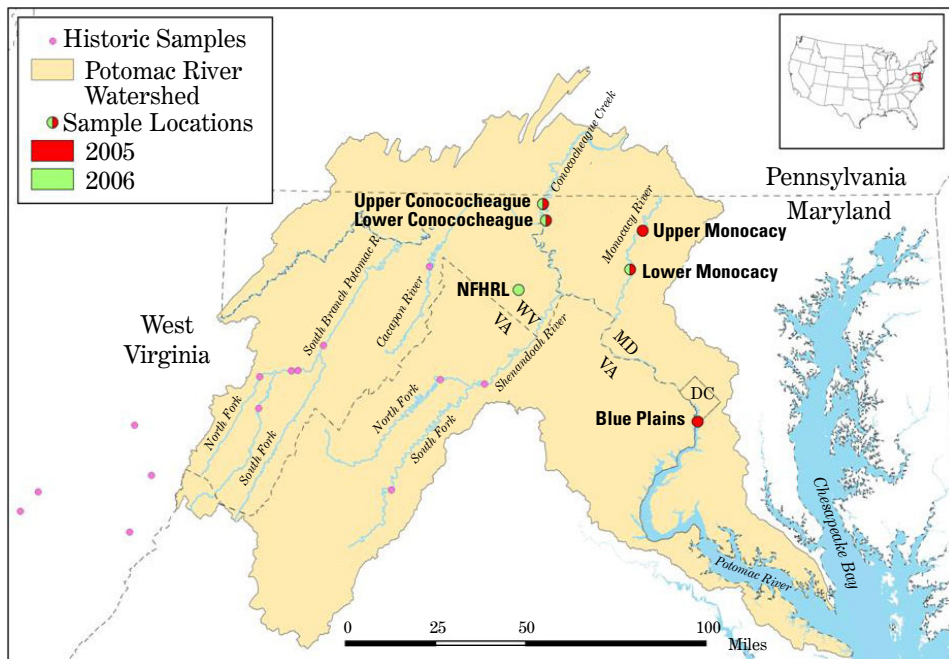
Background

Since 2003, scientists at the U.S. Geological Survey's National Fish Health Research Laboratory (NFHRL) in Kearneysville, WV have been evaluating the reproductive health of smallmouth bass in the upper Potomac River and its tributaries, including the Shenandoah River. They noted the presence of immature female germ cells (oocytes) in the testes of some of the male fish. This condition, a type of intersex, is evidence of a disturbance in the hormonal system of the fish (i.e., endocrine disruption). Further evidence of endocrine disruption occurs when we detect the presence of vitellogenin in the blood of male fish. Vitellogenin is a protein produced by female fish to form egg yolk and is normally absent in males. In addition to the effects on male fish, a substantial decrease of vitellogenin in females also suggests endocrine disruption.

The Problem

Intersex and abnormal vitellogenin in smallmouth bass from portions of the Potomac watershed pose a threat to fish resources. The USGS studies led to several research questions:

- Is there a relationship between intersex and abnormal vitellogenin and nearness to wastewater treatment plant discharges?
- How widespread are these problems within the Potomac watershed?
- What chemicals can be detected at fish sampling locations; which ones are considered to be endocrine disruptors; and do the concentrations relate to land use?



Map of sampling locations.

The Team

To try to answer these questions, a collaborative partnership of scientists from many organizations was formed. Toxicologists from the U.S. Fish and Wildlife Service's Environmental Contaminants Program at the Chesapeake Bay Field Office identified the sampling sites, coordinated the field work, and gathered the land use information. Biologists from the USGS NFHRL directed the field necropsies and performed the biological laboratory analyses. USGS chemists at the Columbia Environmental Research Center (Columbia, MO) estimated water concentrations of organic contaminants at the collection sites using long-term sampling devices. Fisheries biologists from the Maryland Department of Natural Resources provided local expertise on smallmouth bass populations and directed the collections. Biologists from the District of Columbia's Department of the Environment provided additional help

with collections and those from the University of Florida performed the vitellogenin analyses. The U.S. Fish and Wildlife Service and the U.S. Geological Survey funded the work through a jointly prepared proposal submitted in 2004.

The Survey

In the fall of 2005, biologists collected male and female smallmouth bass from two Potomac River tributaries in Maryland, the Monocacy River and Conococheague Creek. For each river, we identified one sampling location immediately downstream of a wastewater treatment plant discharge and one about 10 miles upstream. We collected about 10 males and 10 females from each location. We also collected largemouth bass near the discharge of the Blue Plains Wastewater Plant in Washington, DC. Fish blood and tissue samples were taken for analysis. Microscope slides were prepared of testes and ovaries.

In the fall of 2005 and in the spring of 2006, we installed passive water sampling devices at sampling sites for one to two months. These devices accumulate different types of organic contaminants, including both legacy and “emerging” compounds, allowing scientists to identify contaminants at very low concentrations.

The Results

We found female germ cells (oocytes) in the testes of 82% to 100% of the male smallmouth bass and in 23% of the males from the single largemouth bass collection near the Blue Plains Wastewater Plant in Washington, DC. The baseline prevalence of testicular oocytes in male smallmouth is uncertain but may be in the range of 14% to 22%; baseline for male largemouth may be closer to 0%. We found vitellogenin in the blood of 33% to 90% of the male smallmouth and 85% of the male largemouth. In Conococheague Creek, there was more than a tenfold decrease in the concentration of vitellogenin in the females collected downstream from the treatment plant vs. those collected from the upstream location.

Multiple environmental contaminants were found in the sampling devices, often with higher concentrations of wastewater chemicals (near the treatment plant outfalls). Pesticides currently used in agriculture were detected at all locations. Hormones were not detected in the passive water samplers. However, laboratory tests (yeast screening assays) suggested that estrogenic endocrine-disrupting chemicals were present at all locations. These tests do not identify single compounds, rather an overall response.

Based on the results of these samples, we cannot identify a single chemical or sources that may be causing the intersex and vitellogenin induction. Multiple chemical stressors that are not solely associated with agriculture or wastewater treatment plant effluent may be responsible.

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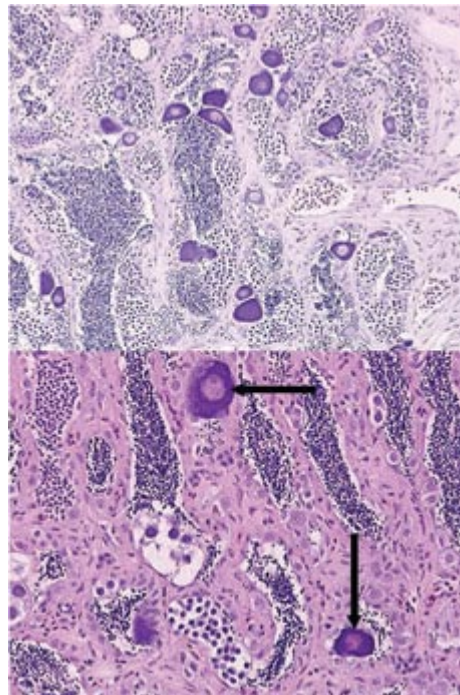
<http://www.fws.gov/chesapeakebay/>

Ongoing Research

To gain a greater understanding of the reproductive health of bass and the implications to the ecosystem in the Northeast U.S., sampling of fish from rivers near National Wildlife Refuges (from Virginia north to Maine) using a similar upstream/downstream design is ongoing. Further sampling is also being conducted in the mainstream upper Potomac River and in the tidal area downriver of Washington, DC.



Smallmouth bass. Illustration by Timothy Knepp, USFWS



Intersex (testicular oocytes). Most often observed as immature oocytes (arrows) within testes. Suggested as a biological indicator of endocrine disruption.

For More Information

Iwanowicz, L.R., V.S. Blazer, C.P. Guy, A.E. Pinkney, J.E. Mullican, and D.A. Alvarez. 2009. Reproductive health of bass in the Potomac, USA drainage: Part 1. Exploring the effects of proximity to wastewater treatment plant discharge. *Environmental Toxicology and Chemistry* 28:1072-1083.

Alvarez, D.A., W.L. Cranor, S.D. Perkins, V. Schroeder, L.R. Iwanowicz, R.C. Clark, C.P. Guy, A.E. Pinkney, V.S. Blazer, and J.E. Mullican. 2009. Reproductive health of bass in the Potomac, USA drainage: Part 2. Seasonal occurrence of persistent and emerging organic contaminants. *Environmental Toxicology and Chemistry* 28:1084-1095.

Guy, C.P., A.E. Pinkney, V.S. Blazer, L.R. Iwanowicz, D.A. Alvarez, and J.E. Mullican. 2009. Assessment of endocrine disruption in smallmouth bass (*Micropterus dolomieu*) and largemouth bass (*Micropterus salmoides*) in the Potomac River Watershed. CBFO-C09-03. U.S. Fish and Wildlife Service, Chesapeake Bay Field Office, Annapolis, MD. <http://www.fws.gov/chesapeakebay/envcont.html>

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Monocacy River below wastewater treatment plant outfall.



Barge electroshocking for smallmouth bass in Conococheague Creek