

Intersex Largemouth Bass in Our Rivers

Sue Beede and Alison Field-Juma

In recent decades, scientists have discovered that the presence of human hormones and hormone-like chemicals in rivers, lakes, and streams can harm fish and their ability to reproduce. The development of an “intersex” condition in their sex organs indicates that fish may have been exposed to these chemicals. The intersex phenomenon is a serious concern because it has the potential to reduce reproduction among entire populations—in this case, of fish.

The term “intersex” is used to describe fish whose testes or ovaries also develop characteristics of the opposite sex. Intersex male fish develop eggs in their testes, called testicular oocytes. While intersex characteristics may naturally occur in some fish species, largemouth bass (*Micropterus salmoides*) normally develop as either male or female, and remain the same sex throughout their lifetime. A study published in 2013 reported that scientists found intersex male largemouth bass in the Assabet, Sudbury and Concord Rivers.

While temperature, season, and environmental stress may also contribute to intersex development, considerable evidence exists that disruption of the fish’s hormonal system, also known as endocrine disruption, may be the cause. Endocrine disruption can result from

exposure to hormones, either natural or manufactured; industrial chemicals that mimic or block a fish’s natural hormones can have the same effect.

In a now famous seven-year study in Ontario, Canada, scientists caused the resident population of fathead minnows to collapse by adding low doses of synthetic estrogen (the main ingredient in birth control pills) to a whole lake. The lake’s top predator, lake trout, began to starve as the minnow population crashed. Happily, the fathead minnow population recovered when they stopped adding estrogen.¹

Largemouth bass in the Assabet, Sudbury and Concord Rivers

In September 2013, the U.S. Geological Survey (USGS) and U.S. Fish & Wildlife Service published a study of nineteen National Wildlife Refuges in the northeastern United States examining freshwater bass for symptoms of endocrine disruption. The research was carried out in 2008-10.² The study included Great Meadows National Wildlife Refuge in Concord, Wayland, and Sudbury, and the Assabet River National Wildlife Refuge in Stow and Maynard.

Other studies of endocrine disrupting chemicals in the Assabet River were carried out in 2009-13 by the U.S.



Vin Antil

Largemouth bass caught on the Sudbury River by Chris Rodstrom of Stow.

Environmental Protection Agency (EPA) in collaboration with USGS, OARS and UMass Amherst. These will be discussed in future articles as soon as they have been published.

The National Wildlife Refuge study’s researchers looked primarily for two signs of endocrine disruption in the bass: intersex testes or ovaries, and measurable concentrations of an egg yolk protein called vitellogenin in the blood of male fish (it is usually not found in male fish). Researchers also evaluated the concentration and severity (defined below) of hormones and hormone-like chemicals in rivers, ponds, and reservoirs within or near the refuges. It was a “reconnaissance” study intended to identify problems that deserve follow-up, rather than to produce definitive results.

The scientists captured largemouth bass and collected water samples from five sites within our watershed: the Assabet River adjacent to the Assabet River refuge (ASR1); the Sudbury River (GRM1),

What is endocrine disruption?

Low-level exposure to some contaminants can disrupt animal reproduction and development by modulating, mimicking, or interfering with normal hormonal function. These contaminants include hormones created in the body, synthetic hormones (such as those manufactured for birth control or menopausal supplement), and industrial/commercial compounds that have some hormonal function (such as alkylphenols, pesticides, pharmaceuticals, and phthalates).

Source: toxics.usgs.gov

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Protecting, preserving, and enhancing the Assabet, Sudbury, and Concord Rivers, their tributaries and watersheds for the purposes of public recreation, water supply, and wildlife habitat.

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Environment is Too Easy to Cut!

The evenings are dark and the rivers are high (finally). The best thing for us to do—apart from analyzing water quality data, of course—is to work on our new river recreation maps so that they will be ready for next summer! But don't wait until summer to take a peek at our rivers—in the winter they are more visible with the leaves down, and utterly spectacular on sunny snowy mornings.



After a lot of hard work to get important environmental programs funded, we were dismayed that the Governor's "9c" cuts to balance the state budget fell disproportionately on the environment. According to the Environmental League of Massachusetts, an average of 0.06% was cut from the overall state operating budget. However, the "9c" cuts to environmental agencies were a disproportionate 3%. Funding for water quality monitoring on the Sudbury, Assabet and Concord—that no one else does, that is essential for environmental protection and law enforcement—was one of the victims. This funding had been gained through the efforts of Senators Michael Barrett and Jamie Eldridge. We will try again next year.

While we work hard to garner grants to cover a portion of our water quality monitoring expenses, we are so very grateful that our supporters, volunteer Citizen Scientists, and our growing number of members continue to fill the breach with their contributions. In so many ways, we couldn't do it without you! Thank you.

We wish you all a very happy winter.

Alison Field-Juma
Executive Director

Congratulations to OARS Board member Ingeborg Hegemann Clark!

Stow Conservation Trust presented Ingeborg with their 2014 Land Conservation Award "in recognition of her decades of dedicated service to the Stow Conservation Commission and the Stow Conservation Trust, for her commitment to and enthusiasm for Land Conservation efforts in Stow and for her professional services throughout the Commonwealth of Massachusetts."



Don Rising

Ingeborg has started a series of articles in local newspapers that provide clear and enjoyable discussions of river science. Her first article "The Seasons of Our Rivers" resulted in this appreciation from a reader: "It was one of the clearest descriptions of the yearly fluctuations in base flow and the water table. [I] think it would be beneficial for all [my] board members to read this article."

If you would like to read the article, go to oars3rivers.org/our-work/speaking-up/inthenews. Keep your eyes peeled for the next one!

Images from 2014



Tsongas River Day 2014: Rochelle Lerner, Maynard High School teacher, and David Mark, Maynard resident and dedicated OARS volunteer, received Congressional Recognition for their work promoting environmental stewardship from Representative Niki Tsongas. Every year Rochelle brings a team of students to our river cleanups. Everyone enjoyed an autumn paddle on the Assabet River in Maynard and Stow.



The 11th season of our Water Wise Workshops! Free waterside workshops were enjoyed by children ages 6-10 in our communities. Hudson Boys & Girls Club children received certificates from Education Coordinator Bill Froberg and Summer Interns for finishing the Junior River Rangers workbook. Thank you to Intel for their support of the program.



State Senator Jamie Eldridge joined OARS and Clean Water Action to speak in support of the EPA's Clean Water rule. EPA's New England Region Administrator Curt Spalding received a "boatload" of letters of support and enjoyed an afternoon paddle on the Sudbury River at Great Meadows National Wildlife Refuge in a flotilla of canoes.



Sudbury River featured at the Holiday Pops concerts! Richard Sebring's original arrangement of "Veni, Veni, Emmanuel" was performed to a stunning display of his photographs and videos taken along the Sudbury River. "Sharing the winter splendor of the Sudbury River with so many people, through the magic of music and moving imagery, is a blessing," says Sebring, Principal Horn of the Boston Pops. We couldn't agree more! The performances were supported by OARS, Sudbury Valley Trustees, and the Wild & Scenic River Stewardship Council.



Invasive plant removal. 65 volunteers hand-pulled more than 600 bushels of water chestnut from the Sudbury and Assabet Rivers this July and August. We are grateful to ERM and EMC for sending volunteer teams and to the volunteers from our local communities. Thank you to Charles River Canoe and Kayak Service and to Laurie Ullman for providing canoes, to Dick Lawrence and Tom Largy for the use of their pick-up trucks and to REI for a grant to support this critical stewardship activity. This photo of an energetic EMC volunteer was taken in Saxonville along the Sudbury River last August.

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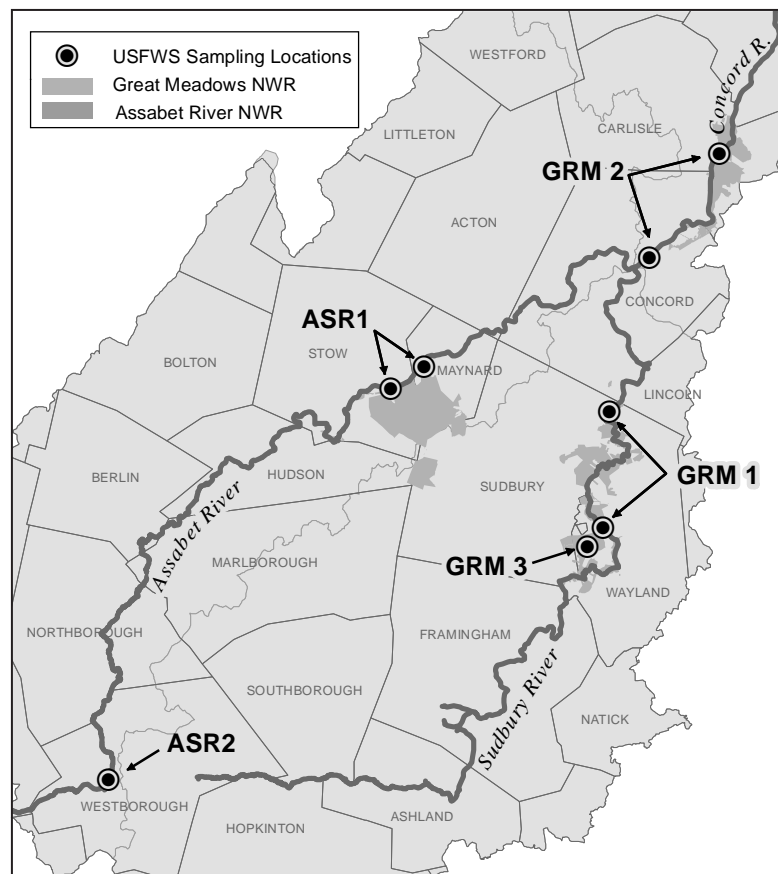
Concord River (GRM2), and Heard Pond (GRM3) within the Great Meadows refuge; and the Nichols Reservoir (ASR2) in Westborough, which is the headwaters of the Assabet River and the control site. Agency scientists caught the largemouth bass by electroshocking at the sampling locations (see map).

The study found intersex male largemouth bass in the Assabet, Sudbury, and Concord Rivers at the refuge sites. The percentage of intersex male largemouth bass captured at the river sites ranged from 9% to 75% and averaged 17% across the five sites. Intersex was defined in this study as the presence of one or more immature eggs in the testes of the males. The researchers also looked for the egg yolk protein vitellogenin in the blood of male fish as evidence of more generalized endocrine disruption or “feminization.” No intersex largemouth bass were found in Heard Pond in Wayland or at the A-1 Reservoir control site.

Seventy-five percent of the largemouth bass from the Sudbury River site (GRM1) were found to be intersex. This was the second highest percentage of intersex largemouth bass among all 14 northeastern national wildlife refuges where largemouth bass were captured. The sample size, however, was quite small. The Assabet River site (ASR1) had the ninth highest percentage, 33%, in this group.

The researchers detected no vitellogenin in the blood plasma of the 41 male largemouth bass from the five sites. In fact, vitellogenin was detected in male largemouth bass at only 6 of the 27 sites in the 14 refuges where they were caught. There is not necessarily a relationship between blood vitellogenin and intersex in male fish. Intersex is likely induced during early developmental stages while vitellogenin in male fish is less dependent on life stage.

No intersex females were found. This was not surprising, as study scientists have yet to find intersex female largemouth bass in their research.



The study also looked at the severity of the intersex features, using a “severity index” based on the number of immature eggs and their distribution (whether single or clustered together) in the testes of the male fish. All of our sites ranked as “low” severity. Nevertheless, scientists found at least one immature egg in the testes of each intersex male fish collected from the Assabet, Sudbury, and Concord River sites. The male largemouth bass at the Assabet River site (ASR1) had the highest intersex severity score of the three sites with intersex male fish, followed by the Sudbury River site (GRM1). The intersex severity of the male largemouth bass captured at the other 12 refuges was also, with the exception of one lake in Virginia, considered “low.”

Searching for causes

The researchers wanted to find out if the fish were being exposed to hormones, particularly estrogens and androgens, in the waters they live in. To see if these substances are present, researchers tested

water samples collected from the same sites where they caught the largemouth bass.

Estrogens are the primary female sex hormones synthesized by all vertebrates, and include estradiol, estriol, and estrone. They are both naturally produced and manufactured for use in oral contraceptives and hormone replacement therapy. They are excreted from the human body and are present in wastewater, which is discharged to our rivers. Previous studies have shown that estrogens can induce intersex in some fish species and harm the ability of fish and other animals to reproduce. Exposure to very low concentrations of estrogen (just 1-10 parts-per-trillion, the same as 1-10 nanograms in a liter of water) has been shown to induce intersex characteristics in laboratory and wild fish.

In this study, scientists measured the total estrogenicity of the water samples, i.e., the total of estradiol, estriol, estrone, ethinylestradiol (synthetic estrogen), and

estrogen-mimickers such as bisphenol A (BPA). They detected a concentration of “Exhibited Estrogens Equivalents” of 2.2 parts-per-trillion in the water sample taken from the Assabet River site. This showed the highest concentration of estrogens observed among all the samples tested in all nineteen northeastern refuges in the study (apart from those collected in the discharge zone of a wastewater treatment plant). None of the samples collected at the Assabet or Sudbury River sites were collected in the discharge zone of the rivers’ five wastewater treatment plants.

This is a significant finding because it is more than twice the concentration of estrogens (1 part-per-trillion) that a number of scientists in the field consider the maximum concentration that will not affect the reproductive health of fish. However, fish species vary greatly in their sensitivity to estrogens, so further research is needed.

Water samples were also tested for the presence, though not the quantity, of androgens such as testosterone. Androgen is the broad term for any compound (usually a steroid hormone) that stimulates or controls the development and maintenance of male characteristics in vertebrates. The primary and most well-known androgen is testosterone. Only water samples from the Sudbury and Concord River sites were

tested for the presence of androgens and both sites tested positive. This indicated only their presence, not the concentration.

What can we conclude?

The study made two important findings about the Assabet, Sudbury, and Concord Rivers. First, there are intersex male largemouth bass at the wildlife refuges in all three rivers. Among the three rivers, the Sudbury River site within the Great Meadows refuge had the highest percentage of intersex males, 75%, but with a very small sample size (only four fish). The Assabet River at the Assabet River refuge, with three intersex males out of nine fish sampled (33%), is more conclusive. According to study scientists, a sample size of approximately 12 males is needed to have confidence in the prevalence of intersex at a site.

Second, the Assabet River water had the highest concentration of estrogens measured in the study with the exception of water samples collected in the discharge zone of a wastewater treatment plant. The concentration of 2.2 parts-per-trillion Exhibited Estrogens Equivalents exceeded what is termed the “predicted-no-effect concentration.” Concentrations above 1 part-per-trillion can cause intersex in males, and other forms of endocrine disruption.

This study raises the possibility that wastewater is causing intersex fish, but there may be other causes and contributing factors as well. More research is needed to determine causality.

Next steps

This study was a preliminary survey designed to prioritize refuges for re-evaluations. Both the Assabet River refuge and the Great Meadows refuge made the list of the seven priority refuges needing comprehensive follow-up studies. Among these seven refuges, the study ranked the Assabet River refuge as a moderate priority and the Great Meadows refuge as a low priority for follow-up. At this time no additional fieldwork or analysis is planned by the U.S. Fish & Wildlife Service. We now await the completion of the EPA study that focused on water quality and endocrine disruption in fish in the Assabet River.

This kind of research is relatively new, assisted by improved techniques to measure very low concentrations of pollutants. It is also quite expensive. It is important that the public and private sectors continue this research so that the causes and impacts of endocrine disruption can be understood and any necessary action can be taken.

References

1. Kidd, K.A., *et al.* 2007. Collapse of a fish population after exposure to a synthetic estrogen. *Proc. National Academy of Sciences* 104 (21): 8897-8901.
2. Iwanowicz, L.R., *et al.* 2013. Evidence of estrogenic endocrine disruption in smallmouth and largemouth bass inhabiting Northeast U.S. National Wildlife Refuge waters: a reconnaissance study. U.S. Geological Survey and U.S. Fish and Wildlife Service. On-Refuge Investigation 51410-1261-5N44 Final Report.

For more information about endocrine disruption see: www.oars3rivers.org/threats/water-pollution/edcs.

Largemouth Bass Results in the SuAsCo Basin

Sample Site	Waterbody	National Wildlife Refuge	Town(s)	Males caught	Females caught	Intersex males
ASR1	Assabet River	Assabet River	Stow & Maynard	9	11	3 (33%)
ASR2	Nichols Reservoir (Assabet headwaters; control)	Not in a refuge	Westborough	10	10	0 (0%)
GRM1	Sudbury River	Great Meadows	Sudbury & Wayland	4	8	3 (75%)
GRM2	Concord River	Great Meadows	Concord, Carlisle & Bedford	11	11	1 (9%)
GRM3	Heard Pond (adjacet to Sudbury River)	Great Meadows	Wayland	7	13	0 (0%)

Thank You to our 28th Annual River Cleanup Sponsors, Donors, and Volunteers!

On Saturday, September 20, more than 150 volunteers came out to make a difference in their communities. Mountains of trash were pulled from the rivers and their banks in towns up and down the Assabet, Sudbury, and Concord Rivers.

Thank you to all who participated!



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Visit www.oars3rivers.org for photos of the cleanup.

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Maynard: Maynard Pizza House, Pizza Express
Northborough: Monti's Pizza,

Northboro House of Pizza
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Sudbury: Papa Gino's
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Towns:

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Thanks to our site leaders, scouters, and boat haulers: Sue Beede, Don Burn, Pat Conaway, Mike Duclos, Bill Fadden, Allan Fierce, Bill Froberg, Dick Lawrence, Marty Moran, Priscilla Ryder, Drew and Kathy Simmons, Lee Steppacher, Pete Tobin, and Laurie Ullman



On the morning of Saturday, December 13 (39°F), sixteen OARS and Stow Conservation Trust volunteers came out to clean up the Kalousdian Sanctuary property owned by OARS in Stow. The Town of Stow holds a Conservation Restriction on the property. Neighbors assisted with access and an impromptu pick-up truck and the Stow Highway Department removed all the trash and debris. A big thank you to everyone!

Welcome, New Members!

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OARS has been protecting the natural resources of our watershed for 28 years, thanks to the help of your generous donations and volunteer hours. How will we protect it in the future? One way is to ensure the sustainability of OARS and its programs. We hope that you will consider including OARS in your will and estate plans. If you would like more information on including OARS in your estate planning, please contact Julia at jkhorana@oars3rivers.org or (978) 369-3956.

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OARS is your local river conservation organization. Established in 1986 to protect the Assabet River, OAR added the Sudbury and Concord Rivers to its mission in 2011 and became OARS.

OARS has over 900 members, a dedicated board of directors, a small professional staff, and a large corps of active volunteers. Our work benefits all communities in the Assabet-Sudbury-Concord watershed.

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