Researchers develop fast test for invasive carp

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A Case Western Reserve University graduate student turned a research paper into a field test that quickly determines whether an Asian carp invading Lake Erie is sterile or can reproduce.

If proven successful, the technique could save money and time in the effort to keep the carp out of the Great Lakes, where the fish could grow unchecked and devour food supplies and habitat critical to native species..

Grass carp, the species *Ctenopharyngodon idella*, have been introduced throughout the Midwest and South to clear ponds choked with weeds. Also called the white amur, fertile fish are illegal to release or buy and sell in Ohio and surrounding states, but have been found in Lake Erie's western basin.

Hatcheries have breeding carp, but shock their eggs with drastic changes in water pressure. The shock results in a third chromosome in the eggs and makes the fish that grow from them sterile. These sterile fish can then be responsibly used for biological control of invasive aquatic plants, but fertile fish could result in growing populations that could devastate the Great Lakes ecosystems.

The U.S. Fish & Wildlife Service and the Ohio Department of Natural Resources monitor for—and are trying to prevent—the spread of grass, black, silver and bighead carp into the Great Lakes.

To confirm whether grass carp caught in the Great Lakes can reproduce, conservation agents must have an eyeball or fresh blood taken from the species tested at a United States Geological Survey laboratory in Wisconsin. The test equipment can run to more than $80,000 a USGS field biologist said.

But Katherine Krynak, who recently earned a PhD in biology at Case Western Reserve, led the effort to develop an inexpensive test that can be done in a boat or on shore in about 15 minutes with a conventional microscope.

Krynak worked with Case Western Biology Instructor Ronald Oldfield and Cleveland Metroparks Zoo's Pam Dennis, a veterinary epidemiologist; Mike Durkalec, aquatic biologist; and Claire Weldon, aquatic research coordinator. Their research is published in the journal *Biological Invasions*.

"With this technique, people can quickly recognize the problem animals and remove them from the population," Krynak said.

While doing her homework for a research paper, she found that Chinese scientists had discovered the shape of the nuclei in red blood cells of hybridized goldfish and Wuchang bream—members of the carp family— looked different depending upon whether the fish had a pair or three or more chromosomes. The proportion of abnormally shaped nuclei grew with the increasing number of chromosomes.

Krynak thought the cellular differences might be used to identify sterile grass carp from those that can reproduce, and therefore be used to monitor the species caught in the Great Lakes or rivers that feed them.

She discussed the ideas with Oldfield and they, Dennis, Durkalec and Weldon developed a technique they could use in the in the lab and field.

They smear a drop of fish blood on a slide, let it dry and fix it with methanol. They then stain the slide to bring out the nucleus, and, after rinsing and drying, view it under a standard microscope.

Like its relatives, sterile grass carp have a noticeably larger proportion of red blood cells with abnormal nuclei.

The researchers tested how well others could accurately differentiate sterile from normal fish. By viewing blood-smear slides, 14 of 15 staff and interns of the Natural Resources department of Cleveland Metroparks correctly identified the fish; a single intern incorrectly identified a single fish's reproductive potential.

Grass carp were first found in the Sandusky River, which flows into Lake Erie's western basin in 2012. Whether they came from fertile carp that were illegally or mistakenly brought into the area or originated from states such as Iowa, Missouri, Arkansas and Mississippi, which have permitted fertile fish since importation began in the 1960s, is unknown. But, the four caught had the potential to reproduce, and the minerals found in the bones of their heads, which can be used to track where the fish have been, indicated the fish had lived in the Sandusky all their lives.

Grass carp pose a number of threats because they eat soft-stem vegetation—the kinds of plants that dominate coastal marshes. The marshes are prime breeding grounds for game fish and act as filters that clear the water, said Eugene Braig, program director for the Aquatic Ecosystem Extension at Ohio State University.

Loss of the vegetation and native fish also poses a threat to ducks, geese and other large aquatic birds, the U.S. Geological Survey says.

"This test has the potential to be very helpful," Braig, said. "The results

are pretty convincing, but I would like to see it field-tested by management agencies."

Braig said that if this method comes to be recognized as more affordable, consistent, and reproducible, it could become a new standard practice for agencies charged with protecting the Great Lakes.

Provided by Case Western Reserve University