

## VHS Collection Protocol

### Contact Information:

The University of Toledo, Lake Erie Center 6200 Bayshore Rd. Oregon, OH 43716

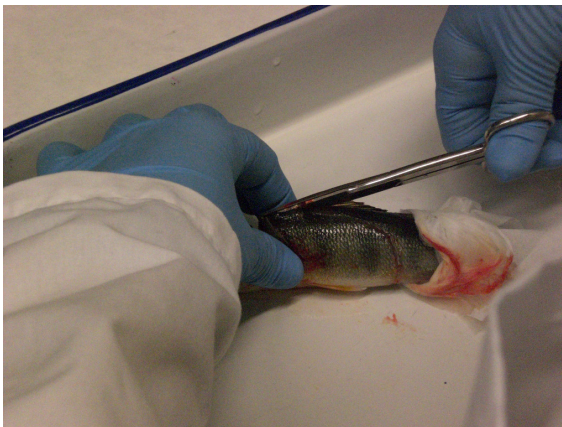
Director: Dr. Carol A. Stepien (carol.stepien@utoledo.edu) 419-530-8360

Ph.D. Student: Lindsey Pierce (lindsey.pierce@utoledo.edu) 304-920-0431

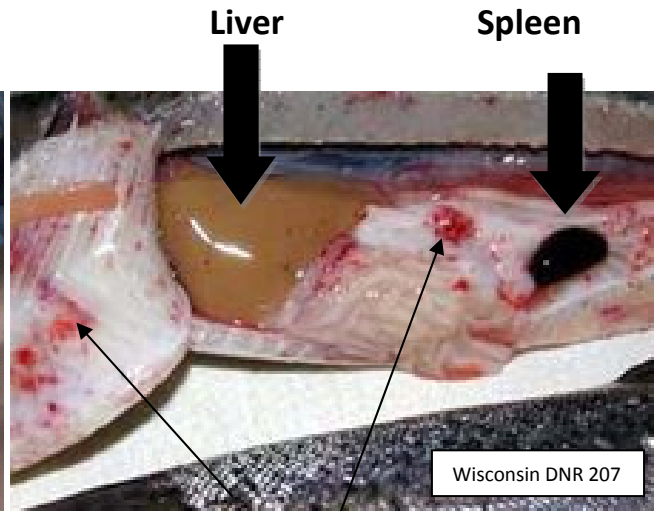
**Species of Concern:** *Yellow Perch, Walleye, Drum, Smallmouth bass, Round goby*

**Information:** RNA later is stable at room temperature for 1 year. Once introduced to tissue, it will last for 1 week not on ice, and 1 month if kept on ice (regular freezer). Once tissue is placed into solution, please contact the Lindsey so samples can be retrieved ASAP. If more than 1 species is infected per site, please contact Lindsey so more tubes can be provided.

1. Prior to collection:
  - \*label tubes with fish number and species (any will work as long as they are the same between samples) with an **L** for “liver” and another separate tube with **S** for “spleen”.
2. Once fish are caught, put on sterile gloves provided, make cut with sterile razor blade provided from the anus up to the head and pull back flesh to expose insides (see below).



Dissection



External hemorrhages (VHS symptom)

3. Remove dime-sized portion of liver with the provided sterilized razor blade, chop into 3-4 pieces, and place in **L** tube containing RNAlater (prefilled with 1.5 mL). Open new sterile razor blade and do same with spleen and place in **S** tube with. Repeat above, each fish with separate sterile razors. Dispose of alcohol pads and razors in provided 50mL conical tube.
4. Mark tubes with a star \* if signs of VHS are apparent (see Figure 2 of hemorrhages).
5. Screw cap back onto tube and place into appropriate container (cooler if possible).
6. Please call Lindsey ASAP for pick up or shipping information. If shipping, please send conical tubes containing biohazard material for proper disposal.