

Warm Springs Fish Health Center

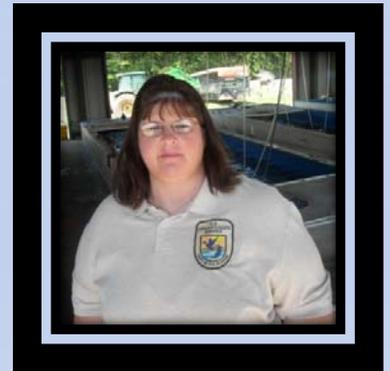
January to March 2013 Activity Report



Norm Heil, Project Leader
Brian Hickson, Fish Biologist
Devin Chappell, Fish Biologist
Allison Hernandez, Fish Biologist
Nikki Persons (below), Fish
Biologist- Arkansas field office



The Fish Health Center (FHC) is a component of the Warm Springs Regional Fisheries Center (RFC) and was developed to improve and enhance fisheries management. More specifically, the FHC provides a wide range of services from disease diagnostics to ploidy inspections.



Images are all courtesy of the US Fish and Wildlife Service unless noted otherwise.

Goals:

- Provide service, expertise and information that contributes to the health, survival, enhancement, restoration, and recovery of fish and other aquatic species in support of national and regional priorities.
- Supports hatchery operations to provide quality fish that contribute to ecosystem objectives.
- Assists in the development of management strategies through assessment and applied research to support the protection of wild stocks and recovery of threatened and endangered species.
- Educates key audiences about the elements of comprehensive fish health and its critical significance to healthy aquatic ecosystems.

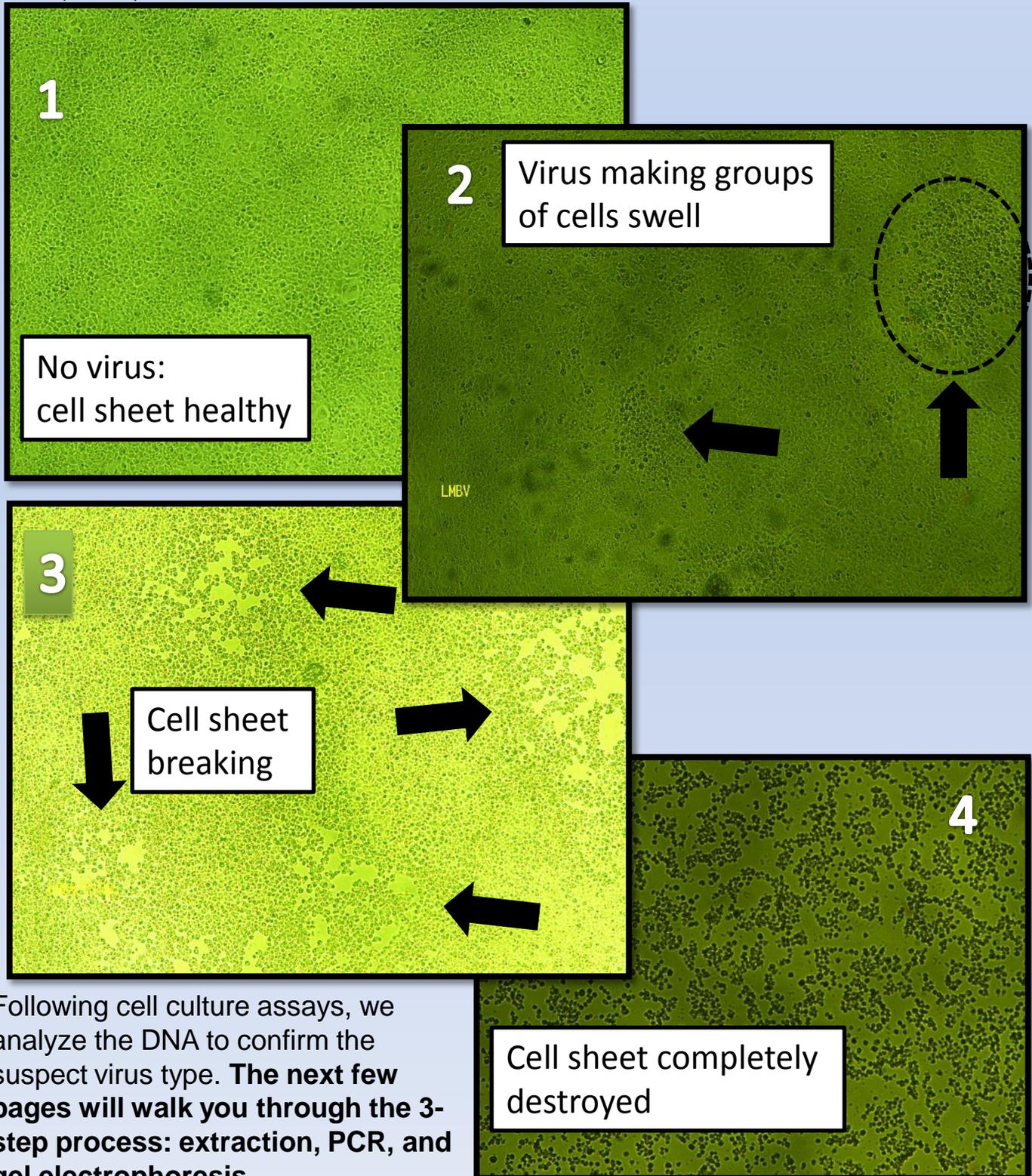


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<http://www.fws.gov/warmsprings/FishHealth/index.html>

Fish Health Diagnostics

All of the national health centers conducted ring testing in January. Ring testing is used as part of quality control and assurance that all of the health centers have cell lines sensitive enough to detect viral strains and that all staff are effectively conducting assays. The two fish viruses used this year were Large mouth bass virus (LMBV seen below on cell culture assays) and Infectious hematopoietic necrosis virus (IHNV).



Following cell culture assays, we analyze the DNA to confirm the suspect virus type. **The next few pages will walk you through the 3-step process: extraction, PCR, and gel electrophoresis.**

Fish Health Diagnostics

EXTRACTION:



Close-up view of the different sample tubes, processing tubes and pipettes used to apply or run the extraction processes.

When conducting PCR confirmation of suspect virus samples, the virus DNA must first be extracted. This piece of equipment is used to simplify and remove error from the extraction process.

The “Bio Robot” provides messages throughout the extraction to let you know where it is in the process. Following extraction, Polymerase Chain Reaction (PCR) is completed. During PCR, DNA is sliced up using temperature gradients and specific enzymes and it is replicated to create definite size bands. Band size is known for the pathogen that is being tested.



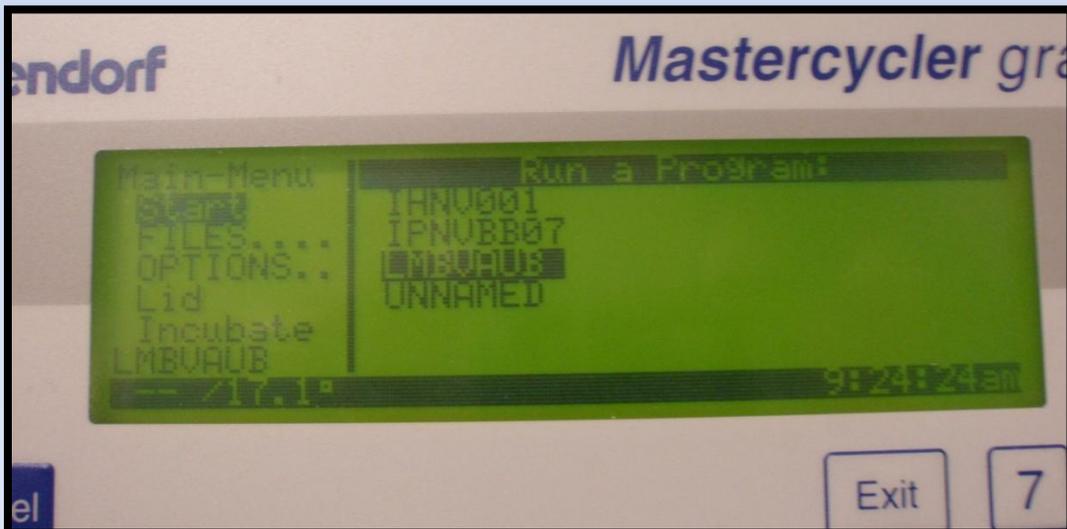
Fish Health Diagnostics

PCR:

PCR is performed using a thermocycler. Specific segments of DNA are replicated using enzymes that copy target DNA. The samples are placed into the thermocycler shown in the images below. It runs numerous cycles at different temperatures to split, separate, slice and amplify (replicate) the DNA strands into bands. Then the amplified DNA bands are used for confirmation assays.



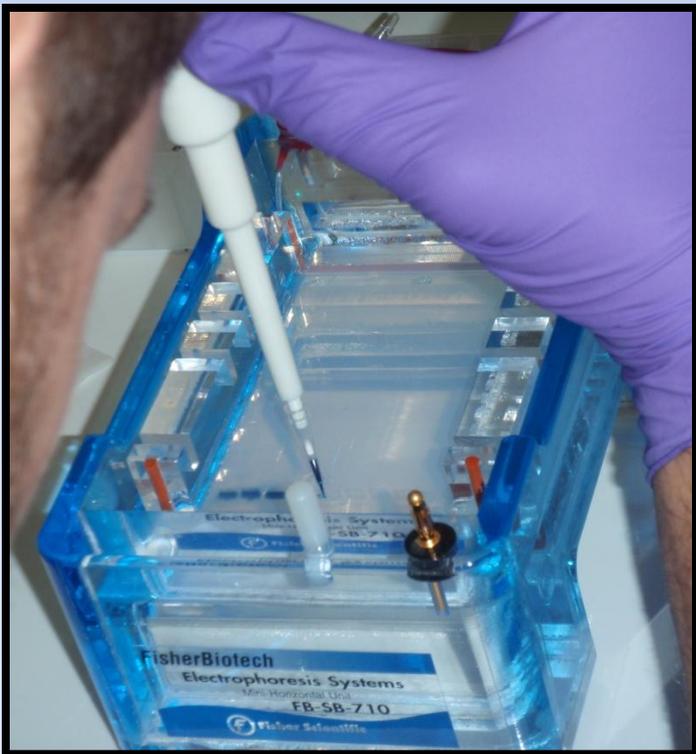
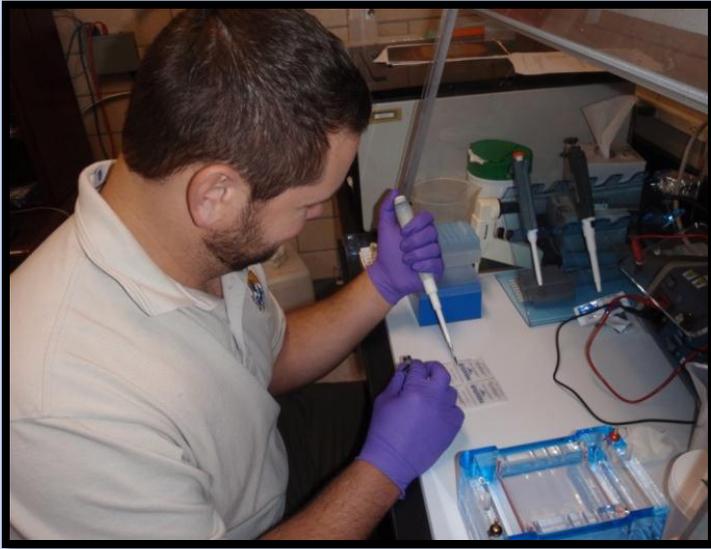
Sample tubes are positioned into heated plate then the heated lid is closed down over the tubes (essentially tubes are encased in an equally-distributed, temperature-regulated chamber).



A pre-set program is selected and executed for an individual virus species.

Fish Health Diagnostics

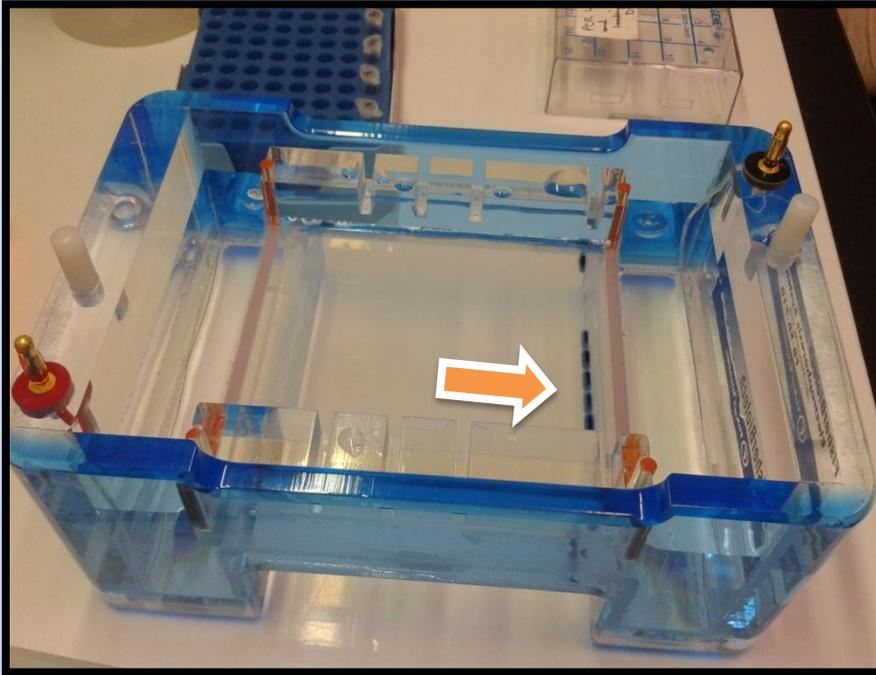
In this series of images, Devin is preparing a gel electrophoresis assay to run post-PCR samples that will determine presence or absence of a suspect virus DNA. A “ladder”, which is used to determine number of base pairs in samples, is also included in one of the wells to run alongside the samples. The band sizes (or number of base pairs) in the ladder are known and the pathogen band size is known, thereby providing us the positive/negative results by confirming the size of the sample DNA band.



Adding loading dye to the samples makes it easier to see them when pipetting into wells. As you can see, we work with very small amounts of material.

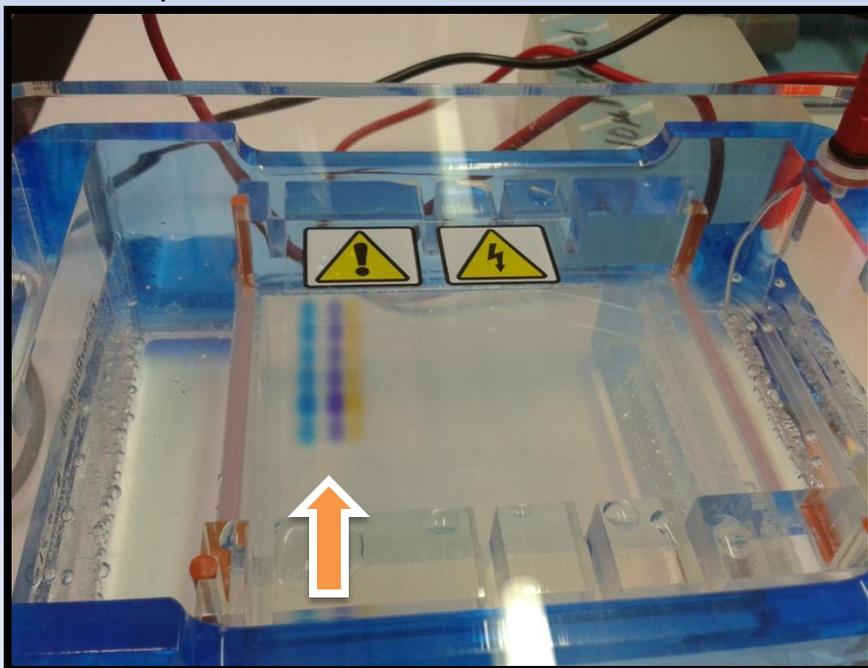
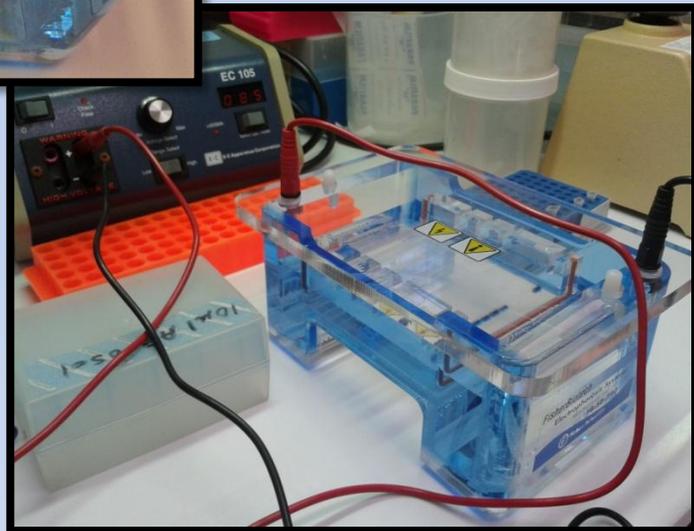
Fish Health Diagnostics

Gel electrophoresis:



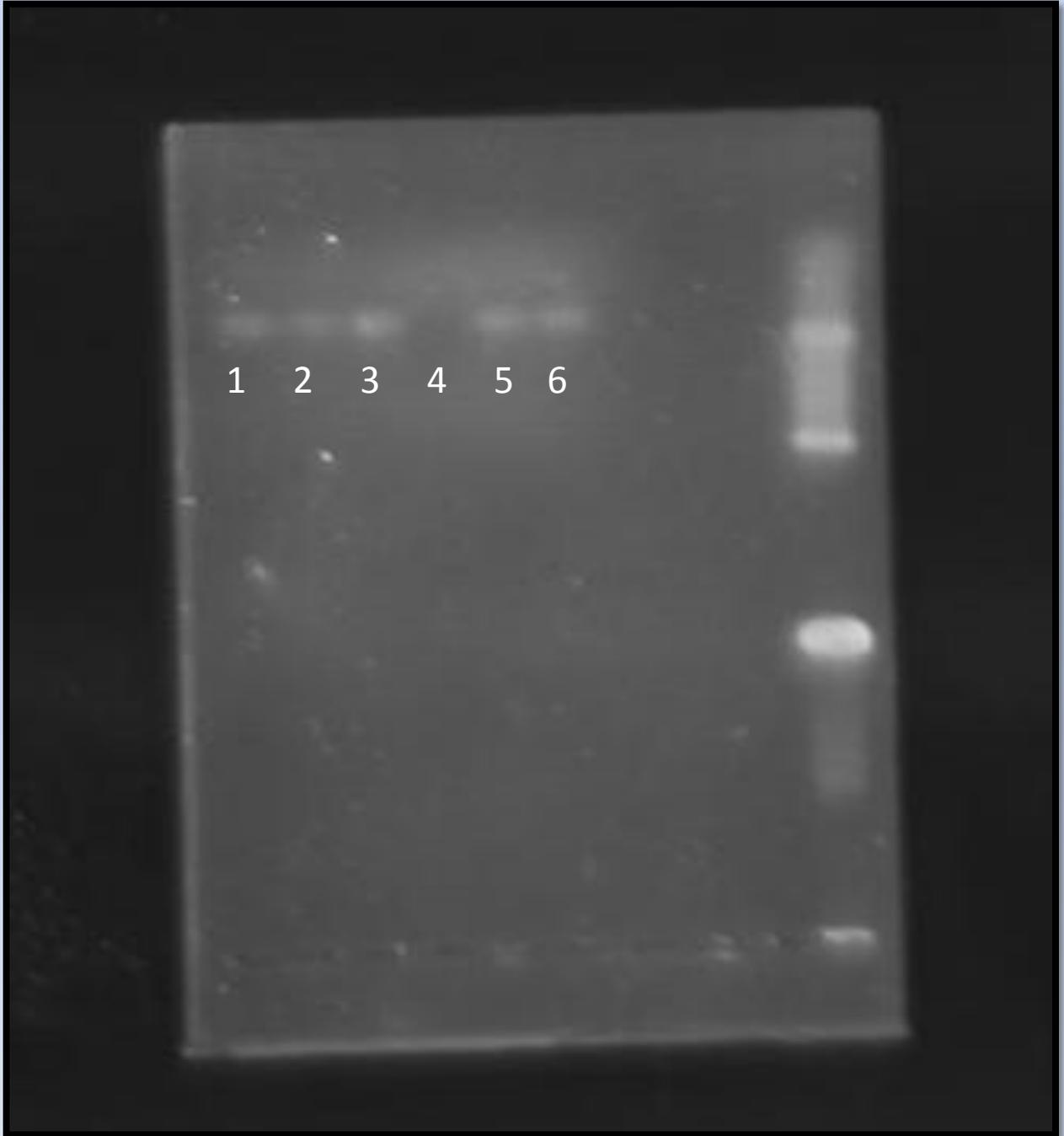
Gel electrophoresis apparatus is shown below. Electric current is sent through the gel to make the DNA sample strands migrate through the agarose gel. Smaller bands migrate faster.

Above: Loaded gel- ready to be run. You can see arrow pointing to the sample wells. The well at the top of the image, and separated from the samples is the ladder. Below: you can see the colored bands of the loading dye running through the gel. When the yellow (smallest) band gets to the top (or right side of image), the electrophoresis is complete.

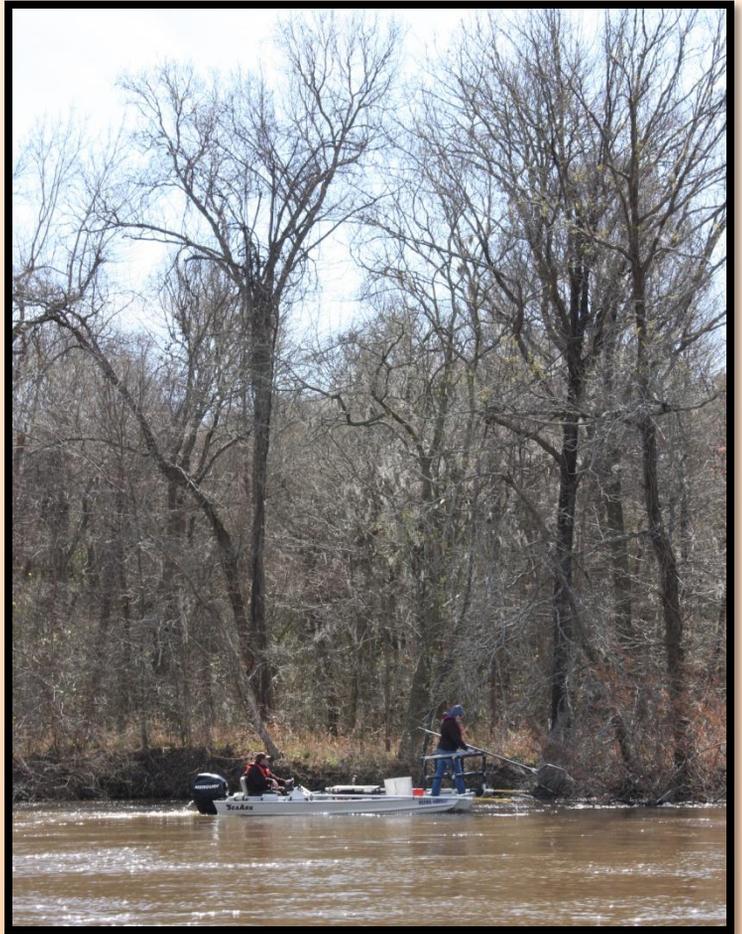


Fish Health Diagnostics

Ring testing results of PCR and gel electrophoresis confirmation of LMBV. You can see the light bars that were positive for five of the six samples (#4 was a negative control). The column of bars on right side is the ladder (degraded in this assay- usually you would see a clear separation of bands).



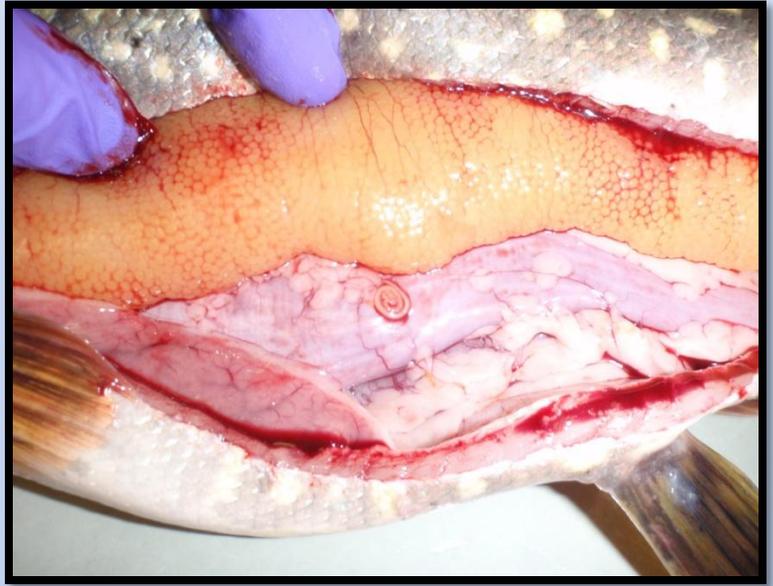
Wild Fish Health Survey



Devin went to Savannah to assist with Refuges Inventory & Monitoring Program to survey fish populations in the Savannah River. While there, he collaboratively used the fish collected for National Wild Fish Health Survey.



Other FHC Activities



Allison travelled to La Crosse, Wisconsin in January to take Intro to Fish Health training. Having worked nearly three years as a fish health biologist previously, the course was useful in filling out the knowledge gaps. The course is put on by NCTC and held at the LaCrosse Fish Health Center. The team-taught course was led by Corey Puzach who composed a well-organized and informational training for all participants, most of which were hatchery personnel. There were classroom lectures in the mornings and hands-on laboratory sessions in the afternoons. Allison was excited to necropsy two fish that she had never worked with previously. In addition to the class tours of the fish health building, participants were provided a tour of the new eDNA lab that was recently built on to the facility. The class also took a field trip to the USGS Upper Midwest Environmental Sciences Center to learn about collaborative research between agencies.



Other FHC Activities



Devin received a plaque and pin from the Regional Office for 10 years service with the United States Government.