WVDL SUMMER NEWSLETTER

2022

Wisconsin Veterinary Diagnostic Laboratory

UNIVERSITY OF WISCONSIN-MADISON

MESSAGE FROM THE DIRECTOR

Greetings from the WVDL, we hope you are enjoying your summer and have some time to relax and recharge. We have had a busy past few months and are excited to share news from the sections about best practices for sample submission, shipping changes, and changes in diagnostic testing options. We are still in the final phases of development for our new laboratory information management system and will be contacting clients soon when plans to move to the new system are solidified.

We are excited to be moving forward with the major renovation and expansion of the WVDL-Barron facility. The 2022 highly pathogenic avian influenza (HPAI) outbreak in Wisconsin emphasized the need for this project to better serve the Wisconsin Poultry Industry and veterinarians in northwest Wisconsin. We are currently in the selection phase for the architect and engineer for the project, which is being facilitated by the Department of Administration and UW Madison Facilities, Planning, and Management.

We are all very excited to welcome Dr. Maggie Highland to the WVDL as the Pathology and Professional Services Section Head. Maggie is a Wisconsinite and UW-SVM alumni. She did her pathology residency at UC-Davis and her graduate work at the University of Washington. She was most recently a diagnostic pathologist at Kansas State Veterinary Diagnostic Laboratory and her professional interests focus on small ruminant infectious disease.

For your calendars: December 1st, 2022 will be our Annual Bovine Genetics Export Meeting. We have moved this meeting to December because the summer season is too busy with vacations, fairs, and other events. This is a free meeting with CE available. If you are new to this meeting, please email us to put you on the invite list. WVDL will also be speaking or attending several meetings this fall including the National Association of Animal Breeders, The American Embryo Transfer Association, World Dairy Expo, The American College of Veterinary Pathologists, and The American Association of Veterinary Laboratory Diagnosticians. If you are planning on attending any of these events, please come find us and say hello!

Please feel free to contact WVDL with any questions or comments. Contact information can be found at the end of this newsletter.

Keith

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- **Pathology Update -** Welcome Dr. Maggie Highland, PEOPLE Program, International Wildlife Disease Conference
- **Serology Update -** AI Testing Update, *Mycoplasma* species Plate Testing Discontinued, Bovine Serologic Disease AGID Testing, Discontinuing Johne's & Brucellosis CF Testing
- **Virology Update -** HPAI Update
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CLIENT SERVICES UPDATE

KEEPING UP-TO-DATE SUBMISSION FORMS

Please be aware that WVDL submissions forms are updated on a yearly basis to meet client and testing needs. These forms many have more frequent updates when there are changes to testing availability as well as testing methodologies. Please find our most up-to-date submissions forms on the the WVDL website. Click here to visit the submission form webpage.

Please also remember that it is greatly appreciated when submission forms are legibly, clearly, and completely filled out. If there are any questions regarding testing submission, please feel free to contact the WVDL, contact information below.

BACTERIOLOGY UPDATE

SEMEN BACTERIAL AND FUNGAL CULTURES

The WVDL offers a quantitative test for bacteria and fungi from raw and extended semen. It is important to note that the Bacterial Count Culture on processed semen is a quantitative test reporting the amount of bacteria and yeast in colony forming units (CFUs) per mL. This test is also useful for the identification of pathogenic bacteria and yeast as well as being quantitative. This test should be used when assessing the cleanliness of the semen collection process or when identifying pathogens. When sending samples other than semen, such as swabs, the test is no longer quantitative and will be reported semi-quantitatively. Older submission forms may have a test option for Fungal Screen or Culture. This should not be used as filamentous fungi (mold) should not be present in semen unless gross contamination has occurred. Our microbiologists are trained to note if filamentous fungi are observed during the Bacterial Count Culture and therefore, please do not select Fungal Culture unless you have contacted the WVDL and discussed the purpose of this testing. Additionally, if you are culturing your own semen and are in need of identification of the isolate you have obtained, please check the box for Bacterial Identification and send in a freshly subbed isolate. If more than one isolate from one plate is needed to be identified, please circle all colony types you would like to have identified. Each colony will be charged a fee for identification. If numerous mixed flora or specific environmental pathogens are report and you need assistance with

interpretation, please contact the WVDL bacteriology section or our client services professional veterinarians (WVDL Madison Laboratory).

REMINDERS FOR TRITRICHOMONAS FEOTUS TESTING USING THE IN POUCH FOR CULTURE

As a reminder, **if you are requiring the Direct Exam**, which uses a microscopic examination of the In Pouch immediately upon arrival at the lab, please **do not roll down** the In Pouch and maintain the genital washing in the top compartment. This is done so that the sample is not diluted in the additional media found in the bottom compartment and allows for microbiologists to easily identify the pathogen if present. If you **are not requesting a direct exam**, and only need the 6-day *Tritrichomonas* culture, **please roll the contents of the top section of the In Pouch** down so that the parasites, if present, have enough media for growth for 6 days. As a reminder for the *Tritrichomonas foetus* PCR, please be sure to submit the genital washing in a Biomed In Pouch and not the transportation tube. WVDL data demonstrated a 6 CT loss when using the transportation tube instead of the In Pouch.

MOLECULAR DIAGNOSTIC UPDATE

NEW ZEALAND MYCOPLASMA BOVIS SEMEN PCR UPDATE

We would like to provide an update for *Mycoplasma bovis (M. bovis)* semen PCR testing with regard to New Zealand requirements. Upon providing data that has demonstrated our assay is sensitive, the WVDL has been granted approval to test semen samples in a single PCR reaction versus the original request of testing in duplicate per New Zealand protocol. This is a very positive outcome in cost savings for our clients who export to New Zealand.

There has been some updates to our current *M. bovis* semen PCR procedures to meet the New Zealand requirements. A larger starting sample volume is needed, and there were other changes required for the semen sample to be processed differently than for other PCR's. Thus, we require any semen samples that are to be tested for *M. bovis* (currently only needed for New Zealand export) have their own dedicated straws and submission forms. If other tests are required such as BVD or IBR, please include duplicate straws and submit on a separate submission form for those testing requests.

The key points to keep in mind for semen submission for New Zealand:
1. Submit a minimum of 4 small straws for *M. bovis* PCR

2. Submit *M. bovis* PCR submissions separately from other PCR requests (including submission form and straws)

WVDL has also been granted approval to test embryo collection fluid for *M. bovis* by PCR. These samples are processed and tested alongside semen samples and need to arrive at the laboratory by 11 am on Friday in order to obtain results by the following Tuesday. Collection fluid can be sent refrigerated with ice packs. We require 2-5ml of unfiltered collection fluid sent in leak-proof containers; screw caps are preferred.

SAMPLE REQUIREMENTS FOR SEMEN PCR

As the PCR assays are slowly conforming to the OIE standard, the minimum OIE requirement of 100 µl of semen for PCR detection of certain diseases has become a standard that warrants your consideration. The WVDL is looking at adopting a minimum of 100 µl of semen for all PCR assays, which will keep us in compliance with OIE requirements. To be in compliance, please submit **a minimum of 4 smaller (0.25 cc) or 2 larger (0.5 cc) straws** for PCR testing on semen. We will continue to accept 3 smaller straws for the remaining of 2022, but we would like to encourage all our clients to plan ahead and adopt this change as soon as possible prior to the requirement taking effect in 2023. For clients that have already been contacted and have adopted this change, we truly appreciate your flexibility.

STORAGE CONDITIONS FOR SERUM & WHOLE BLOOD SAMPLES

It is hot and sunny again and with that wonderful weather also comes an increased importance of proper storage conditions of serum and whole blood samples. Our section has seen an increase in testing failures in some of our assays which can be attributed to improper sample handling prior to receipt at the lab. This increase in initial testing failures may result in a slower turn-around time in results so we recommend storing whole blood and serum samples at refrigerator temperature as soon as possible after collection, and properly packed with cold packs for shipping.

BVD NASAL SWAB COLLECTION

Nasal swabs are an inexpensive sample type geared for high throughput BVD PCR testing. As a reminder, the swab shafts need to be broken off at the length that is below the top of the plate (see picture A).

We have been received swab shafts that have been broken above the plate surface (see picture B). These shafts would either puncture through the sealant foil, or get stuck to the foil. Swabs received in this manner have a much higher chance of contamination and individual sample integrity is compromised when the foil is removed with swabs stuck to it.

The key points to keep in mind, **please be sure to snap the shafts of the swabs so they lie below the top of the collection plate (as in picture A).** See the list of materials needed and collection procedure if you are interested in nasal swab collection.

1. Materials Needed for Nasal Swab Collecting:

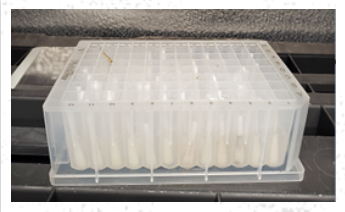
a. The following form and supplies can be found by visiting our website: http://www.wvdl.wisc.edu/index.php/forms/

- i. BVD PCR NASAL SWAB SUBMISSION FORM
- ii. BVD NASAL SWAB KIT

1. Polyester tip swab with Dacron shaft
2. 96-array deep well plate
3. Aluminum seal film
4. Re-sealable plastic bag

2. Collection Procedure

a. Using a single Dacron or polyester-tip nasal swab with a plastic applicator, (non-wood applicator) swab 2-3 inches inside one nostril with enough pressure to capture epithelial cells.
b. Put the swab in the 96 deep-well plate.
c. **Snap off the shaft so that the tip is below the top of the 96 deep-well plate and not protruding above the top of the top of the plate.** If necessary, use scissors to cut off the shaft, being careful not to touch the collection tip.
d. Wipe down the scissors between cuts with dry, clean cloth.
e. Do not add liquid to 96 deep-well plate.
f. Cover with aluminum foil.
g. Record the sample ID number on WVDL submission form in the corresponding well (an electronic spreadsheet can be used in addition to the WVDL submission form for ease of entry).
h. Mark the 96 deep-well plate with the corresponding submission form page number.
i. Place the 96 deep-well plate in a re-sealable bag provided and enclose the WVDL submission form in a separate bag.
j. Ship the plate and form with cold packs overnight, not on Fridays.
k. Refrigerate samples until packaging for up to one week. Do not freeze.
l. If shipping from within Wisconsin, we recommend UPS Ground as a fast and inexpensive submission handler.
m. A video of this collection procedure is located on our website at: http://www.wvdl.wisc.edu/index.php/forms/



Picture A. Proper swab collection

Picture B. Swab shafts are too long and sample integrity is compromised.

PATHOLOGY UPDATE

WELCOME DR. MAGGIE HIGHLAND – NEW PATHOLOGY SECTION HEAD

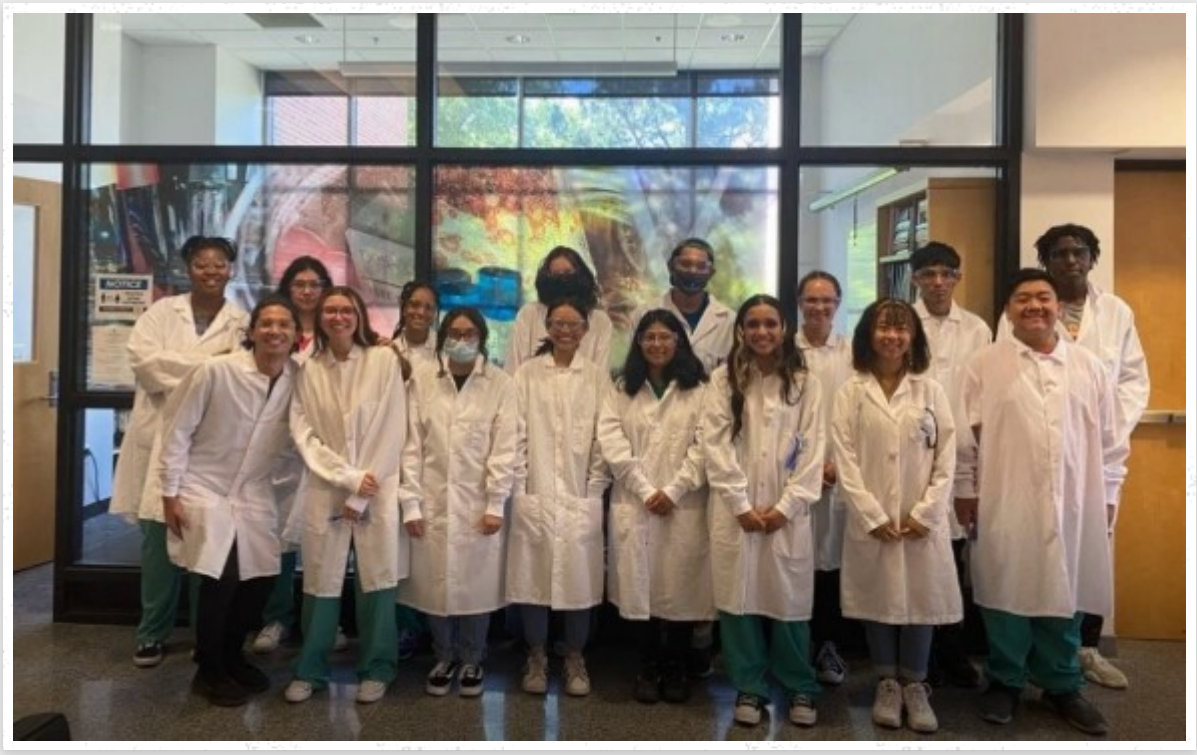
Dr. Highland joined the WVDL on June 1, 2022, to serve as the Pathology and Professional Veterinary Services Section Head and as a Diagnostic Pathologist. Dr. Highland received both her B.S. (School of Pharmacy, 1997) and DVM (2006) from UW-Madison. She completed 2 years of anatomic pathology residency training at the University of California-Davis (2006-2008) then returned to Wisconsin to complete 2 additional years of residency training as the Milwaukee County Zoo and Wildlife Anatomic Pathology Fellow (2008-2010). Following her anatomic pathology residency training, Dr. Highland completed a Ph.D. in Immunology and Infectious Disease as a USDA Ph.D. trainee within the Department of Veterinary Microbiology and Pathology within the College of Veterinary Medicine at Washington State University (2010-2016), then stayed in Pullman, WA for 3 years serving as a USDA-ARS Veterinary Researcher, specializing in small ruminant infectious diseases (2016-2019).

Prior to arriving back to her home state of Wisconsin in June 2022, Dr. Highland served as a diagnostician and necropsy laboratory section head within the Kansas State Veterinary Diagnostic Laboratory. She maintains a strong interest in small ruminant health and disease processes, particularly infectious disease processes and immunology.

WVDL & UW-MADISON PRECOLLEGE ENRICHMENT OPPORTUNITY PROGRAM FOR LEARNING EXCELLENCE (PEOPLE) PROGRAM

On July 14th, WVDL hosted a group of 15 high school students as part of the UW-Madison Precollege Enrichment Opportunity Program for Learning Excellence (PEOPLE) program. Students participated in a wildlife anatomy and dissection workshop and toured the Madison laboratory. WVDL staff members had the opportunity to share their career paths and details of

their current jobs as well as answer questions. Learn more about the program here https://peopleprogram.wisc.edu



2022 INTERNATIONAL WILDLIFE DISEASE CONFERENCE AT THE WVDL

Along with colleagues from the American Association of Wildlife Veterinarians, USGS National Wildlife Health Center, Wisconsin Department of Natural Resources, Arizona Game and Fish Department, and the Arkansas Game and Fish Commission, WVDL's Dr. Lorelei Clarke and Dr. Betsy Elsmo co-hosted a student wetlab on-site for the 2022 International Wildlife Disease Conference on July 23, 2022. Students from around the globe received hands-on experience investigating mock mortality events outdoors on the grounds surrounding the WVDL and performed postmortem exams on wildlife specimens that were donated by the Dane County Humane Society – Wildlife Center, USGS National Wildlife Health Center and Wisconsin DNR.

SEROLOGY UPDATE

UPDATE TO SEROLOGICAL AVIAN INFLUENZA (AI) TESTING

To ensure we are able to continue providing exceptional diagnostic services, on August 15, 2022, we will transition our testing method for the detection of Avian Influenza from agar gel immunodiffision (AGID) to an enzyme linked immunosorbent assay (ELISA).

As we have unfortunately experienced this year, both domestic and wild avian species can be severely and devastatingly affected by avian influenza viruses. The ISO certified and USDA licensed assay we have selected for detection of the virus is designed to measure the relative level of antibody to avian influenza in chickens, turkeys, ducks, ostriches, and geese sera. We look forward to better serving all of our avian producers by providing early identification and accurate surveillance of antibody presence for a wider range of species. We will continue to provide confirmatory testing via AGID and will work in partnership with NVSL and our WVDL-Madison facility to identify nucleic acid from avian influenza virus found in non-serum samples. The price of the avian influenza ELISA will be $3.00 per serum sample. The price for the AI AGID will also be $3.00 per serum sample.

We recognize this is an increase in test cost. The decision to increase cost was not taken lightly and involved several levels of consideration. We reviewed the cost of reagents and current market standards, considered our increasing demand for testing, and acknowledged potential supply chain issues. We concluded that the cost to run the AGID test method has increased and therefore, the AGID will be maintained for confirmatory purposes only. We determined that the ELISA test methodology will allow us to efficiently meet the growing testing load while continuing to provide highly specific results. We validated the use of several licensed avian influenza ELISAs allowing us to be proactive in regards to potential supply

chain issues. We care deeply about the health of your flocks and look forward to utilizing a test method that will aid our efforts of providing timely diagnostics.

If you utilize your own submission form, please update the test method requested by August 15, 2022. We appreciate your decision to partner with us and take great pride in supporting our nation's poultry industry. If you have any questions, please call the WVDL Barron laboratory at 715-637-3151.

MYCOPLASMA SPECIES PLATE TEST TO BE DISCONTINUED

Due to very low demand for the *Mycoplasma synoviae, meleagridis* and *gallisepticum* Plate Test, the WVDL will no longer carry the reagents needed for this test. The WVDL will continue to offer the ELISA and the hemagglutination inhibition (HI) serological test for these pathogens and these tests do satisfy the NPIP requirements as well as state requirements for Poultry Flock Programs. We will continue to accept the Wisconsin Individual Poultry Test Report Form for individual birds attending fairs and poultry shows. These individual poultry submissions will likely be tested via the HI test method, unless the ELISA is specifically requested. If you have any questions in regards to what testing is required for various purposes, please contact the WVDL Barron laboratory at 715-637-3151.

BOVINE SEROLOGIC DISEASE DETECTION USING AGAR GEL IMMUNODIFFUSION (AGID)

The WVDL continues to use AGID testing for BTV, BLV and EHD for regulatory testing only. These tests were reported as negative, weak positive and positive. Upon consultation with professionals within the bovine genetics industry, the WVDL will solely report negative or positive when reporting AGID serological results, so as there is no confusion as to the significance of the results. If you have any questions, please contact the WVDL Madison laboratory at 608-262-5432.

DISCONTINUATION OF JOHNE'S DISEASE AND BRUCELLOSIS COMPLEMENT FIXATION (CF) TESTS

Upon examination of the health certificates, the Johne's Disease or Brucellosis Complement Fixation (CF) tests are no longer needed. The Johne's Disease CF test has been discontinued. The Brucellosis CF test will be discontinued for on January 1, 2023 and may be available solely for confirmation testing. For Johne's Disease serological testing needs, please use the Johne's Disease/*Mycobacterium avium* subspecies *paratuberculosis* (MAP) ELISA. We also offer the Johne's Disease/MAP PCR and

liquid culture from feces. The Johne's Disease Liquid Culture is approximately a 9 week culture and should only be used when required for health certificates.

For Brucellosis serological assays, the WVDL offers a variety of tests including the Brucellosis Buffered Acidified Plate Antigen (BAPA), Antibody Card Agglutination (Card), Standard Tube Test (STT) and the Fluorescence Polarization Assay (FPA) tests. Please check the health certificate requirements for the country or state that the testing is needed for. The WVDL will automatically perform confirmatory testing on any non-negative Brucellosis tests. Please contact the WVDL with any questions or concerns you may have.

VIROLOGY UPDATE

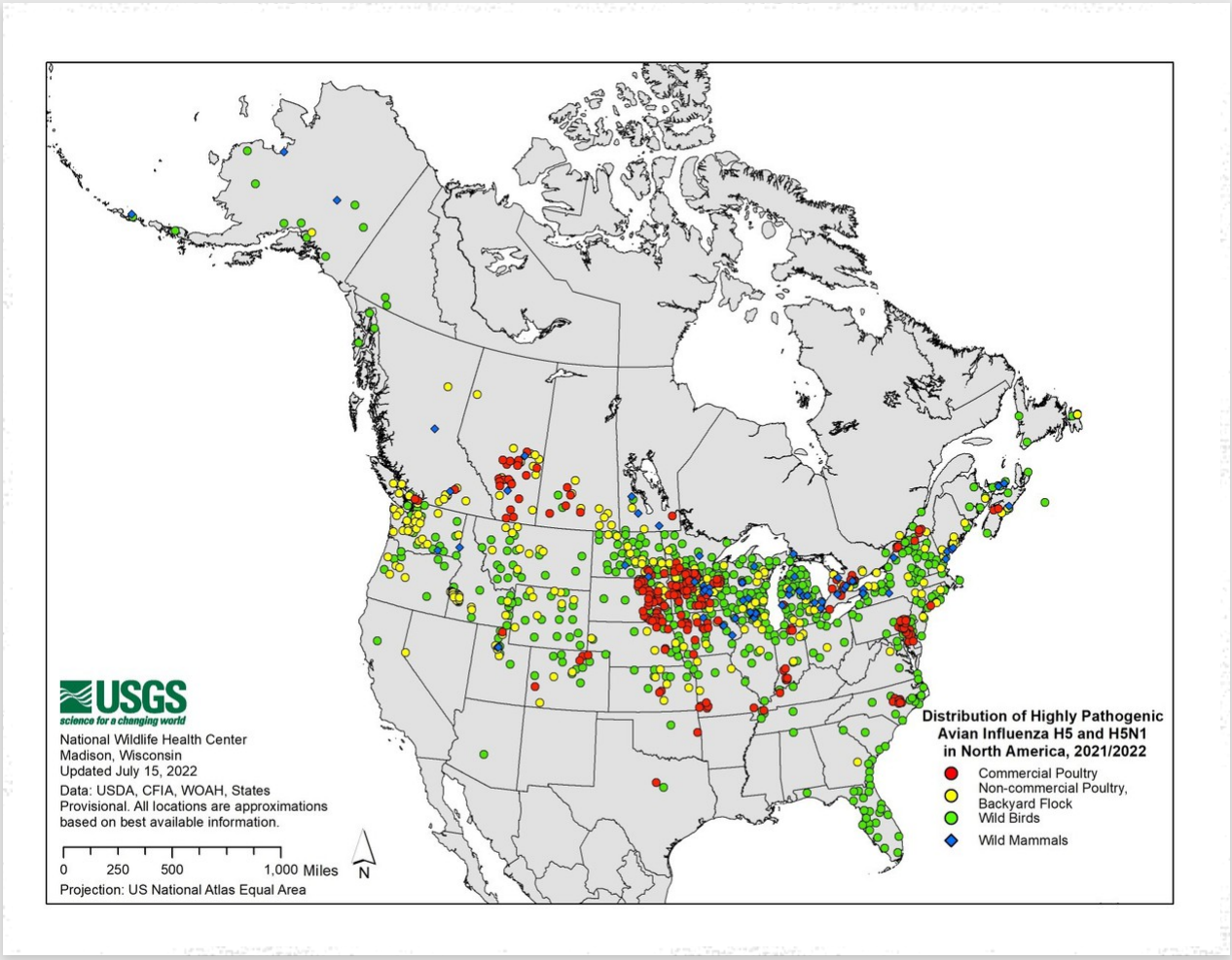
HIGHLY PATHOGENIC AVIAN INFLUENZA (HPAI) UPDATE

WVDL continues to provide new FAD investigation and surveillance testing for HPAI as the outbreak winds down, with post cleaning and disinfection of previously infected premises recently completed. Staff had been performing testing 7 days a week from mid-March to early May to support movement testing within multiple control zones, before we moved to 6 days a week and eventually 5 days a week and now as needed. Staff in multiple sections throughout WVDL have modified their schedules and workloads to accommodate the additional testing with rapid turnaround times. Between March 2022 and July 2022, WVDL performed over 3,341 HPAI PCR tests on poultry, with 22 positive premises identified in Wisconsin (1 was tested at MN VDL due to proximity of the lab to the premise).

WVDL worked with WI DNR and Dane County Humane Society to be the first state in the US to identify a HPAI infection in multiple red fox kits and bobcats. Since then, HPAI infection had been identified in several mammals including opossum joeys across several states. These infections represent sporadic spillover events into wild mammals that are sharing the landscape with HPAI-infected wild birds. Ingestion of birds infected with HPAI is presumed to be the most likely source of infection in these mammals.

Nationwide, as of 7/25/22, approximately 100,058 HPAI PCR tests have been run by 44 NAHLN labs across 31 states that were activated for this outbreak. A little over 10,000 additional HPAI PCR tests have been run on wild birds. The virus has been detected from coast to coast, and 3 lineages were identified as separate introduction to different flyways, with multiple re-assortments of the viral genome over time. The re-assorted virus has become the predominant circulating virus strain.

There will be several hot washes (internally at WVDL, externally with DATCP and nationally with NAHLN partners) to capture and discuss the strengths and areas for improvement during the outbreak.



SPECIAL EVENTS

BOVINE GERMPLASM MOVEMENT PLAN (BGMP)

Funded by the USDA National Animal Disease Preparedness and Response Program (NADPRP), a multidisciplinary group representing AAVLD and NAHLN, industry, and SAHOs held a 2-day discovery meeting in Madison, Wisconsin on May 19 and 20. The NADPRP funded project will develop a bovine germplasm movement plan (BGMP) to maintain intra- and interstate movement of semen, embryos, and high genomic merit cattle in the event of an FAD, specifically foot and mouth disease.

The core team is led by Keith Poulsen, Director of the Wisconsin Veterinary Diagnostic Laboratory and Danelle Bickett-Weddle, CEO of Preventalytics and includes Ailam Lim (WVDL), Darlene Konkle, Wisconsin State Veterinarian, and Charles Brown II from PEAK Genetics. They assembled a working group from 14 different states and 2 Canadian Provinces to discuss the current state of the bovine semen and embryo industries as well as the diagnostic testing and regulatory movement barriers that would occur in an FMD outbreak in North America.

The meeting was attended by 31 people in-person and 18 virtual attendees with joint sessions followed by break-out working groups. The working groups identified gaps and tasks to develop a guidance plan to maintain beef and dairy breeding programs and North American

genetics as global leaders. We were fortunate enough to have industry sponsors for food and beverage and want to show our appreciation to PEAK, ABS, ST Genetics, TransOva, Semex, and Sunshine Genetics. Overall, the hybrid meeting was well received and the core team received great feedback including this quote from an industry stakeholder participant:

“Thank you both very much for putting on a very informative and productive meeting. While there is a lot of work to do, there are lots of good and competent folks leading the charge. I feel confident we can put together a program that will provide a solid foundation, should something happen.”

The stakeholder groups will meet virtually to follow up on tasks with in-person opportunities at AAVLD/USAHA meetings in 2022 and 2023. The core group aims to exercise a draft plan in late 2023.

Group photo from May 20, 2022 at the University of Wisconsin-Madison Campus.

Break out group with industry, diagnostics, and animal health official representatives talking about resources needed to address gaps to draft a BGMP.

Industry partners from bovine artificial insemination and embryo production companies discussing enhanced biosecurity plans.

CONTACT US

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