WVDL FALL NEWSLETTER

OCTOBER 2023

Wisconsin Veterinary Diagnostic Laboratory

UNIVERSITY OF WISCONSIN–MADISON

Message from the Director

Hello from WVDL

Thank you for taking time to read our Fall Newsletter. So far this fall, we have been quite busy with the academic year starting, World Dairy Expo, and preparing for our annual AAVLD/USAHA meeting in National Harbor, MD. We have several members of our team presenting their work in the quality assurance, bacteriology, virology, and molecular diagnostics scientific sessions and I am proud of how our entire WVDL team is represented.

In this newsletter, you will find some excellent information about antimicrobial sensitivity reporting, enteric disease PCR panels, and guidance on how to prepare for a possible resurgence of highly pathogenic avian influenza.

Please mark your calendars for our 2023 Bovine Genetics Export meeting on Thursday, December 7th from 9:30 to 2:30. This is a free event, and we offer 5 CE hours. Watch your email or contact us if you would like more information.

Finally, I would like to welcome Dr. Stephanie Ruppert to our team of diagnostic pathologists! We are very excited to have her on our team and are looking forward to working with her.

Keith

IN THIS ISSUE

Save the Date - 2023 WVDL Bovine Genetics Meeting

WHEN

Thursday, Dec. 7th, 9am-3pm

WHERE

445 Easterday Lane

Madison, WI

MORE INFORMATION

Agenda:

- Section updates from Virology, Serology, Molecular Diagnostics, and Bacteriology
- Updates from DATCP and USDA
- Special Guest Speakers:
  - **Dr. Howard Ketover** will talk about emergency animal handling and movement
  - **Dr. Alexa Burton** will talk about outcomes and findings from high genomic animals treated for pneumonia
- Updates from **Special Agent Scott Mahloch, FBI**

This meeting is free to attend and lunch will be provided. Five CE credits will be provided for attending. The WVDL will sending out an official email invitation with instructions for RSVP. Any feedback on needed agenda items or specific questions for our team to cover are much appreciated.

VIROLOGY UPDATE

Highly Pathogenic Avian Influenza (HPAI): Preparing for Fall Sample Submissions

HPAI continues to be on the forefront of WVDL's testing preparedness. We are closely watching the Fall migration of birds through the Mississippi Flyway and anticipate a busy Fall testing season for HPAI again. The Cornell Lab of Ornithology currently produces a live bird migration map: https://birdcast.info/migration-tools/live-migration-maps/. As a reminder, wild birds can be infected with HPAI and show no signs of illness. They can carry the disease to new locations

when migrating, therefore continue to pose a threat to the commercial poultry industry. The Eurasian H5 strain HPAI outbreak that started in the spring of 2022, continued into 2023, and is not yet over. While we have seen some relief last Fall and this past Spring 2023, quite a few cases were confirmed recently, all in the northern United States across multiple migratory flyways. All bird owners should review their biosecurity procedures and continue to stay vigilant to protect poultry and pet birds from HPAI.

MOLECULAR DIAGNOSTIC UPDATE

What You Need to Know About Avian Influenza PCR Testing at WVDL

Media: WVDL is able to send media for AI testing upon request. The form and a video on proper sampling technique can be found at https://www.wvdl.wisc.edu/forms/ under the "Supply Order Forms" header.

WVDL email contacts: One email address has been created to streamline point of contacts at WVDL. Please use AISubmissions@wvdl.wisc.edu when contacting WVDL about avian influenza questions and sample submissions.

WVDL submission form: https://www.wvdl.wisc.edu/forms/ We have recently updated the avian submission form with boxes to check for which category of testing is requested. **NOTE: this affects testing turnaround time, so please complete the boxes/sections on this form.**

Testing schedule: Routine testing is performed 3 days/week. The expected time for results to be reported is around 5-6 pm or later on testing day. Additional testing schedule will be added only when same day testing results are needed, thus advance arrangement is **required** for these types of testing. Please reach out to us as soon as you anticipate urgent testing needs for permitted movement, control zone surveillance testing, restocking, re-movement testing, or over-mortality threshold testing. If you have questions of needing additional information, please refer to resources available on the DATCP website:

- DATCP avian influenza webpage. Includes a biosecurity self-assessment, prevention tips, and other information about the disease.
- USDA Defend the Flock program. Biosecurity information and videos to learn about protecting your flock.
- DATCP's AI resource center. Fact sheets and brochures to learn about food safety, protecting your pets, and biosecurity.

Any high increases in mortality or abnormal clinical signs should be reported to DATCP at: (800) 572-8981.

Bovine Molecular Diarrheal Disease Detection

Fall has arrived in Wisconsin and WVDL would like to highlight the Bovine Enteric PCR panels that are offered and performed on a weekly basis. More information on panels and individual PCR testing options can be found on our website including the bovine enteric disease panel submission form.

Please visit the "Submission Forms" tab at the WVDL website or follow this link here:
https://www.wvdl.wisc.edu/wpcontent/uploads/2020/10/BovineEntericDiseasePanel.pdf

Below highlights the the many different calf scour panels that WVDL has to offer. Please read on to see how to make the best choices from the available panels that provide the most appropriate testing for the calves you are working with and investigating.

Calf Scours PCR Panels for pre-weaned calves
Scours Panel A: Detects rotavirus, corona virus, and *Cryptosporidium*
Scours Panel B*: Detects rotavirus, corona virus, *Cryptosporidium*, and *Salmonella* genus. This panel is for **calves > 7 days of age**.
Scours Panel C*: Detects rotavirus, corona virus, *Cryptosporidium*, *Salmonella* genus, and *E. coli* K99 as well as intimin gene for enteropathogenic *E. coli.* This panel **SHOULD** be selected for **calves < or = 7 days of age ONLY** unless history and clinical signs suggest that older calves should be run under this test.

Juvenile and Adult PCR Panels
Enteric Panel*: Detects corona virus, *Salmonella* genus

**significant CT values (< or = 35) for Salmonella PCR are automatically cultured for serotype if an isolate is found. Antimicrobial susceptibility testing for Salmonella isolates are provided upon on request.*

SEROLOGY UPDATE

Reminder: The BLV AGID Kit Has Changed

WVDL has long used the BLV AGID kit from Veterinary Diagnostic Technology, Inc. (VDT; Wheat Ridge, CO). This company no longer produces the BLV AGID kit and we have now transitioned to using the Innovative Diagnostics (ID) Vet BLV AGID kit. This kit performed similarly as the VDT kit, but with a slight increase in sensitivity. As a reminder, clients will not recognize any changes in their results as the kit change did not change the reporting of results. Please contact WVDL with any questions or concerns.

Reminder: Increase in Positivity Rate for the BLV cELISA (ID Vet) for Cattle Less Than 9 Months of Age

Upon switching to the ID Vet cELISA kit, WVDL observed an increase in positive and inconclusive results. Upon analysis, we have found an increase in positive and inconclusive BLV cELISA results specific to cattle under the age of 9 months. For cattle under the age of 9 months, the positivity rate for BLV-specific antibodies is 17.3% for the ID Vet kit. Please be aware that clients may observe an increased positivity rate for cattle under 9 months of age as compared to last year. However, for cattle over the age of 9 months, the positivity rate has remained similar for both kits. Therefore, it is possible that the ID Vet BLV cELISA kit is more sensitive for maternal antibodies. It is also possible that the sampling group has changed between 2022 and 2023. We want our clients to be aware of the increased positivity rate with bovine serum where the animal is under 9 months of age.

PATHOLOGY UPDATE

Investigation of Brix Refractometry for Estimating Bovine Colostrum Immunoglobulin G Concentration

More interesting research on bovine colostrum evaluation for newborn dairy calves is available from the team at WVDL. Many dairy operations use a Brix refractometer to assess the quality of first-milking colostrum. This study investigated whether a digital Brix refractometer could be used in a model to predict colostrum IgG concentration and whether more than one %Brix threshold could be used for different colostrum IgG concentrations. It is important to be aware of how to manage colostrum programs on the dairy as well as preventing failure of transfer of passive immunity (FTPI). Calf-caretakers should strive to improve their colostrum feeding practices and keep in mind the well-being of the newborn calf. Smaller volumes may reduce unwanted side effects and shorten the time interval in which calves refuse to nurse, while still delivering an adequate mass of IgG to have successful transfer of passive immunity.

To read this manuscript in full please visit this link:
https://pubmed.ncbi.nlm.nih.gov/37818390/

The authors would like to thank everyone at WVDL that contributed to this manuscript especially the staff at the WVDL - Barron Lab for their assistance, time and testing of colostrum samples evaluated in this study.

Evaluating Beef Cattle Colostrum and Transfer of Passive Immunity in Beef Cattle Neonates

Dr. Breuer has been investigating beef cattle colostrum quality evaluation as well as evaluation of transfer of passive immunity in beef cattle neonates. Read more about these researched findings below in the following publications:

- https://pubmed.ncbi.nlm.nih.gov/37596893/
- https://pubmed.ncbi.nlm.nih.gov/37549250/

BACTERIOLOGY UPDATE

Changes to Antimicrobial Susceptibility Testing

WVDL completed changes to antimicrobial susceptibility reporting on October 1. The changes are described below. There was also a visual change where susceptibility results are no longer reported as a table but as a list.

In an effort to advance Antimicrobial Stewardship, WVDL has changed how we report antimicrobial susceptibility testing (AST). Antimicrobial Stewardship at the diagnostic laboratory level includes, “positively affecting clinical outcomes, help maintain antimicrobial effectiveness, assist clinicians in using antimicrobial agents safely, and minimizing the selection of resistant pathogens, laboratories must use a standardized, well-defined method for performing AST.”1 WVDL uses, primarily, the broth microdilution method, which quantitatively measures the *in vitro* activity of an antimicrobial agent against a particular bacterial pathogen. Antimicrobial agents, in a serial dilution, are prepared and mixed with a standardized suspension of the bacterium. WVDL does not prepare these drug dilutions in-house, but rather relies on panels provided in 96-well plate format by Trek Diagnostics (ThermoFisher Scientific). These are incubated and the minimum inhibitory concentration (MIC), which is the highest dilution (lowest concentration) of an antimicrobial drug that completely inhibits bacterial growth, is determined.

Based on the MIC, the resistance, intermediate, or susceptibility of an organism, from a particular host species and tissue to a particular antimicrobial is established using the Clinical and Laboratory Standards Institute (CLSI) breakpoints. A breakpoint is established by CLSI utilizing microbiological characteristics, pharmacokinetic-pharmacodynamic (PK/PD) parameters, and/or clinical outcome data. Veterinary-specific breakpoints were established with particular attention to the product label. The MIC for the particular pathogen-drug combination is used against the CLSI breakpoint established for that pathogen-drug

combination to determine interpretative criteria which is susceptible, intermediate, or resistant (see example). The CLSI guidelines also allow for >S as an interpretations which indicates 'not susceptible' and <R as an interpretation which indicates 'not resistant'.

The CLSI guidelines are specific to a particular bacterium isolated from a particular host species' tissue. As an example, there are specific breakpoints for particular antimicrobial agents that have been established for bovine respiratory disease pathogens such as *Pasteurella multocida*, *Mannheimia haemolytica* and *Histophilus somni*. These breakpoints do not apply to these bacteria isolated from non-respiratory tissues from cattle. Additionally, these breakpoints do not apply to non-bovid species such that a *P. multocida* isolated from a cat would not get these same breakpoints. Therefore, the CLSI guidelines utilize a grouping system for interpretations of antimicrobial agents and their uses for veterinary pathogens.

- **Group A**: includes antimicrobial agents with VETERINARY-SPECIFIC breakpoints and interpretive categories that are considered appropriate for routine, primary testing for food and companion animals. These antimicrobial agents are considered first to report and use, and are preferred over using those with human medical breakpoints. These Group A compounds have demonstrated an acceptable level of correlation between *in vitro* susceptibility test results and clinical outcome.
- **Group B**: includes antimicrobial agents with veterinary-specific breakpoints and interpretive categories but are considered antimicrobials that should only be tested and reported as 'drugs of last resort'. The Subcommittee on Veterinary Antimicrobial Susceptibility Testing (VAST) considers these antimicrobials to be 'drugs of last resort' and concern exists for selecting for resistance, which could be transferred from animals to humans. The veterinary laboratory can report these at their discretion but are mostly used as antimicrobial resistance monitoring.
- **Group C**: includes antimicrobial agents that use HUMAN medical breakpoints and interpretive categories. These agents may perform adequately, but outcomes for many veterinary applications have not been demonstrated. The veterinary laboratory can report these at their discretion.
- **Group D**: include antimicrobial agents that are regulatory agency-approved for use in the specific animal species. Although quality control data is available, these antimicrobial agents DO NOT have CLSI-approved veterinary-specific or human medical breakpoints or interpretive categories. These agents may be approved for use in other animal species and have veterinary-specific breakpoints in those animals. However, it is not recommended to use breakpoints set for a particular animal species to be applied to a different animal species. This is because there are differences in dosages and pharmacokinetics between animals, people and between animal species. Thus, these agents should be reported selectively before extra-label use agents (Group D), but after agents in Group B.
- **Group E**: includes antimicrobial agents that are NOT APPROVED but may be used in an extra-label manner per the Animal Medicinal Drug Use Clarification Act of 1994 (AMDUCA) guidelines in the US. These agents maybe selective tested and reported and are often used for antimicrobial resistance monitoring. Group E may also include certain antimicrobial agents that are used only for a specific infection site (such as nitrofurantoin for treating urinary tract infections) in non-food-producing animals.1

Currently, WVDL provides interpretations based on CLSI guidelines as well some breakpoints supplied by Trek Diagnostics (ThermoFisher Scientific). The breakpoints supplied by Trek Diagnostics will no longer be used. WVDL will solely be using breakpoints supplied by CLSI. Therefore, WVDL will report mostly Group A and Group C antimicrobial agents based on the pathogen and what species and location on that host species the pathogen was isolated from. On occasion, some Group B, D and E antimicrobials may be interpreted with an MIC and interpretive criteria based on CLSI Vet01 1 and Vet09 2 guidelines. An example is applying *M. haemolytica* breakpoints for bovine respiratory disease to other members of the *Pasteurellaceae* family is acceptable such as *Biberstienia* and *Gallibacterium* species 2. As well, the CLSI Vet09 extrapolates the *Staphylococcus aureus* breakpoints and interpretive criteria for bovine mastitis so that Gram-positive cocci (but not *Enterococcus*) can be interpreted. 2 Interpretations for bovine respiratory disease, metritis and mastitis have been extrapolated for camelid, caprine, cervid and ovine species. **Therefore, WVDL would like to remind clients that there will be fewer antimicrobials reported with interpreted categories as we continue to move to reporting only CLSI and improving the visual look of the reports.** Veterinarians can always contact us for more information regarding AST or if additional antimicrobial agent breakpoints are needed.

For more detailed information on antimicrobial susceptibility testing please visit the WVDL updated guidelines here: https://www.wvdl.wisc.edu/wp-content/uploads/2023/04/Antimicrobial-Susceptibility-Testing.pdf

1 CLSI Performance Standards for Antimicrobial Disk and Dilution Susceptibility tests for Bacteria Isolate from Animals. CLSI, Vet01, Edition 5.
2 Understanding Susceptibility test Data as a Component of Antimicrobial Stewardship in Veterinary Setting. CLSI, Vet09, Edition 1.

Interpretive Category	Enrofloxacin Breakpoints (MIC, µg/mL)
Susceptible	≤ 0.5
Intermediate	1 – 2
Resistant	≥ 4

Example: *Escherichia coli* was isolated from a canine urine sample. The MIC for enrofloxacin was 0.25 µg/mL. Using the breakpoints listed above, the MIC for the isolate is categorized as susceptible because it is < 0.5 µg/mL.

CONTACT US

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