



**Wisconsin Veterinary
Diagnostic Laboratory**
UNIVERSITY OF WISCONSIN-MADISON

2019 Summer Newsletter

Message from the Director



Hello from our team at WVDL! We hope you all are enjoying your summer and appreciate your time to read our quarterly newsletter. We have some important news about test changes, upcoming continuing education, and our staff spotlight focuses on Joel Larson, a microbiologist at the Madison Laboratory.

We are very excited to now be open for sample receipt on Saturdays! Sample receiving is open from 9 a.m. to 1 p.m. on Saturdays to receive and accession drop off samples as well as deliveries from couriers like FedEx and UPS. Please let us know if you would like a "Saturday Delivery" UPS label to get your time sensitive samples to WVDL.

We have two CE opportunities that I would like to highlight. First off, our annual Bovine Genetics Export meeting is slated for Thursday, July 25th, 2019. If you spend a significant amount of time with advanced reproductive techniques or the bovine artificial insemination industry, you will find this meeting helpful to troubleshoot regulatory testing requirements and emerging issues in the industry. The meeting is free, please contact us to be put on the mailing list. Second, we will be hosting a Veterinary Forensics Seminar and Autopsy Wet Lab on Sunday, November 3rd, 2019. We have seen a significant increase in forensic cases and are looking forward to sharing best practices and protocols with a fantastic guest speaker. See below for the link to register.

Please contact us with questions and have a great day!

Keith Poulsen
Director

Barron

First Annual Poultry Conference A Success - The First Annual Poultry Conference held May 29 in Cameron, Wis., was well received by all in attendance. Commercial poultry workers, backyard producers and Public Health Departments in both Wisconsin and Minnesota were invited to attend. Thank you to all who took part and stay tuned in the upcoming months for information on the 2020 conference!

Two New Team Members - Kathie Brown and Abby Suvada have joined the WVDL Barron team. Kathie, who grew up in Rice Lake, Wis., joins the staff as an Office Operations Associate in the Client Services department. She and her husband enjoy the outdoors and spending time with their three children and three grandchildren. Abby just completed her sophomore year studying Animal Science at UW-River Falls and is a student staff member helping out this summer. She loves spending time outdoors and riding her horse in her free time.

Client Services

Sample Receiving Open Saturdays - Beginning July 6, the WVDL-Madison location will be open on Saturdays from 9 a.m. to 1 p.m. to receive samples. Drop off samples in person, via a courier, UPS or Fed Ex. Additionally, any supplies, media or testing kits can be picked up during these hours as well. Take advantage of our [UPS Shipping Program](#) to ship any size package for a \$30 flat rate from anywhere in the U.S. Order Saturday delivery labels on Fridays through our [online service](#) (make sure to check the Saturday delivery option).

Microbiology

Mastitis Real-Time PCR Update - The WVDL has discontinued the mastitis panel real-time PCR testing on milk samples. This assay was used for the detection of *Staphylococcus aureus*, *Streptococcus uberis*, *Streptococcus agalactiae*, *Prototheca zopfii* type 2 and *Mycoplasma bovis*. WVDL will continue to test milk samples for *Mycoplasma bovis* and *Prototheca*

Johne's ELISA on Milk Samples - The WVDL will begin referring all requests for Johne's testing on milk samples to Texas A&M Veterinary Diagnostic Laboratory until further notice. Due to the small number of annual requests, the WVDL has decided to no longer stock the kits required to run this assay. WVDL's current ELISA manufacturer has applied for USDA approval for milk testing and when that approval is finalized, WVDL will reinstate Johne's disease testing on milk samples.

Johne's ELISA on Caprine Serum Samples - WVDL has finished evaluation of the VMRD ELISA assay for use on caprine serum. The manufacturer is currently working on obtaining USDA licensing for the use of the kit on caprine samples, but until that is completed, WVDL has conducted an evaluation using well established samples from our archive. The VMRD ELISA showed a diagnostic sensitivity of 100% and specificity of 96%. The interpretation of S/P ratio on the ELISA for caprine species is different than the bovine samples. Both set of interpretations will be included on result reports for easy interpretation.

Test Cost Changes - The following tests have had significant increases in a reagent costs and/or personnel time that has required an increase in the test price. The Johne's Disease complement fixation assay will now cost \$8 per sample. The Brucellosis complement fixation assay will now be \$6 per sample. The Johne's Disease PCR pool test will now cost \$34 per pool. We apologize for this inconvenience.

Update on Antimicrobial Sensitivity Testing

Due to an increase in reagents and the implementation of testing for Staphylococcal Penicillinase and methicillin resistance, the WVDL will be increasing the cost of susceptibility testing for all species to \$28.00 for in-state and \$42.00 for out-of-state testing per isolate. The cost of susceptibility testing for mastitis pathogens will be \$19.00 for in-state and \$28.50 for out-of-state testing.

The WVDL is implementing additional testing for *Staphylococcus* susceptibility testing starting August 1, 2019. The first step is used to determine if the *Staphylococcus* isolate produces β -lactamase, which cleaves and deactivates penicillinase-labile penicillins, which include penicillin, amoxicillin, carbenicillin, ticarcillin, azlocillin and piperacillin. When the WVDL identifies that a *Staphylococcus* isolate produces β -lactamase, the interpretation for these drugs will be reported as resistant. However, cloxacillin, dicloxacillin, nafcillin and oxacillin are penicillinase-stable penicillins, and their interpretation will not be affected by the secondary test identifying Staphylococcal β -lactamase. The results of the β -lactamase test, will negate results given by our standard minimum inhibitory concentration (MIC) susceptibility test and therefore, clients need to pay attention to the results given as resistant, intermediate or susceptible rather than the MIC numerical value.

Additionally, the WVDL will be implementing susceptibility testing for the identification of methicillin-resistant *Staphylococcus* (MRS) and methicillin-resistant *Staphylococcus aureus* (MRSA) starting August 1, 2019. Although, we still utilize the nomenclature of "methicillin-resistant", methicillin is no longer available for treatment. Therefore, "methicillin resistance" is used interchangeably with "oxacillin resistance". Most resistance to oxacillin, in staphylococci, is mediated by the gene *mecA*, which directs the production of a supplemental penicillin-binding protein (PBP/PBP2a) during bacterial cell replication leading to resistance. The WVDL uses the Kirby Bauer disk diffusion method to identify oxacillin resistance. This test is slightly different depending on the species of *Staphylococcus* (as some species are tested against cefoxitin instead of oxacillin), but the results are the same if resistance is found.

When oxacillin/methicillin resistance is identified, the following antibiotics will also be reported as resistant: penicillin, ampicillin, oxacillin with 2% NaCl, cephalothin, ceftiofur, amoxicillin with clavulanic acid, cefazolin, cefovecin, ceftodoxime, cephalothin, imipenem, ticarcillin and ticarcillin with clavulanic acid. Oxacillin-resistant staphylococci are considered resistant to all penicillins, cepheems, β -lactam/ β -lactamase inhibitors and carbapenems, despite the numerical results given by our standard minimum inhibitory concentration (MIC) susceptibility test. Therefore, clients need to pay attention to the results given as resistant, intermediate or susceptible rather than the MIC numerical value. An interpretation will be provided for these additional tests, which will guide clients through the interpretation of their results. Please contact the WVDL if any questions arise.

Reminder for *Campylobacter fetus venerealis* & *Tritrichomonas foetus* testing

The WVDL offers a variety of testing options for these two venereal diseases, but submission of the samples is key to successful testing. For *Campylobacter fetus venerealis* culture, please submit genital washing in Weybridge Transport Media. The sample must arrive at the WVDL within 48 hours post collection and shipped at room temperature. With good collection techniques and proper sample handling, storage and shipment, we were able to reduce the amount of overgrown samples from 2.91% to 0.39% (see Table 1). The *C. fetus venerealis* culture is offered every day and Weybridge can be purchased through the WVDL.

Table 1: Amount of cultures reported as overgrowth and overgrowth post filtration and none isolated (N/I) after filtration

Result	Overgrowth	N/I after Filter
Jul '15 – Jun '16	1.43% (115)	7.20% (579)
Jul '16 – Jun '17	2.92% (251)	7.16% (616)
Jul '17 – Jun '18	1.21% (99)	4.95% (404)
Jul '18 – Jun '19	0.39% (31)	3.29% (261)

Additionally, the WVDL offers culture and PCR for *Tritrichomonas foetus*. For **BOTH** culture and PCR, please submit the genital washing in a Biomed InPouch™. Please do not submit in any other transport media including Biomed TF Transport Tube™ as we have observed a decrease in sensitivity using this transport media as compared to the InPouch™. For PCR, we can pool up to 5 samples for more cost effective testing. However, pooling should not be used when fulfilling a regulatory testing requirement. The InPouch™ is incubated at the laboratory for 24 hours prior to preparation for PCR and PCR testing is run on Tuesday and Thursdays.

Along with PCR, we have the direct observation/read and the 6-day culture. **When a direct observation/read is required, please do not roll down the InPouch™** as mixing the contents of the upper chamber with the lower chamber dilutes the sample and can make identification of a small amount of *T. foetus* more difficult. When only the culture is required, please roll down the pouch so the liquid is just in the lower chamber. This indicates to staff that the direct read is not needed and provides sufficient nutrients. The direct read and culture are offered every day and the samples must arrive at room temperature with 48 hours post collection.

Staff Spotlight: Joel Larson, Microbiologist

Where are you from? What high school and college did you graduate from? What is your degree in?

I was born in Madison and grew up in Fitchburg and Oregon, Wis. Graduated from Oregon High School and received a BS in Bacteriology from UW-Madison.

What is one thing on your Bucket List?

To complete the circuit of Lake Michigan by bicycle.

What book are you currently reading?

Travels with Foxfire

What is the one food that you will never bring yourself to eat?

Sweetbreads

What is your favorite movie soundtrack?

Anything Ennio Morricone was behind. Particular favorite would be For a Few Dollars More. Also strangely enjoy the soundtrack from Gettysburg.

If you could play a lead role in any movie, which character would you choose?

Ford Prefect

What three flavors of ice cream would be on your triple scoop cone?

Chocolate, Cookies & Cream and Peanut Butter-Chocolate



When you were a child, who did you want to be when you grew up?

Joe Montana

What pets did you have growing up?

Several dogs, mostly serially. Dozens of cats - when you live in the country they sort of show up and sometimes never leave.

What skill would you like to master?

Playing guitar. I have tried a dozen times, and just don't have a good feel for it. (Being tone deaf doesn't help.) It's pretty hard to get the old drum kit down to the beach to entertain people around the fire.

What conspiracy theories do you believe in?

That Mick Jaeger and Keith Richards conspired to get Brian Jones out of the band; directly leading to his accidental drowning.

Best concert you ever saw?

A Foghat concert - Blue Oyster Cult opened

What countries have you traveled to?

Canada, Mexico, South Africa, Ireland, England, Wales, Norway, Sweden, Denmark, The Netherlands, France, Germany, Austria and Switzerland

Favorite part of your job?

A little Apple polishing here. The job is pretty routine, but when a client calls and I am able to get something done for them (getting cases up from sample receiving to get on that day's run, etc.), it does feel pretty good.

Virology

Diagnostic / Surveillance testing:

- In April 2019, WVDL was notified that Veterinary Diagnostic Technology, Inc., the source of our BTV, EHD and BLV AGID kits as well as the BTV cELISA kit will no longer be in business. We are working thru validation processes of alternative vendors and with the export industry to expand acceptable sample testing methods. Meanwhile, confirmatory testing of BTV cELISA samples may be referred to NVSL.
- West Nile Virus (WNV) – Since 1999, health officials began a surveillance program to monitor the mortality due to WNV in avian species, testing primarily members of the Corvidae family (crows, ravens, and blue jays). Dead birds throughout Wisconsin are submitted to WVDL via public health departments. Virology personnel remove skin samples from dead birds and test for WNV by PCR. A county is closed after one bird is determined to be positive and no other birds will be tested from that county after that result. We are off to a slow start of testing compared to recent years. As of 7/1/19 we've tested 13 birds from 10 counties and have had 0 positives so far. For the same date in 2018 we tested 20 birds from 15 counties, and had our first positive on 5/21. In 2017 we tested 51 birds from 28 counties and had the first positive on 5/18/17
- During the summer months, we see an increase in the requests for testing porcine oral fluids. We recommend choosing the Porcine oral fluid PCR panel since it includes both tests required for DATCP. Tests included in this panel are: PRRS and SECD PCRs. SECD PCR tests include porcine epidemic diarrhea virus (PEDV), transmissible gastroenteritis virus (TGEV) and swine delta corona virus (SDCoV). Only rope extracted oral fluids will be accepted as an appropriate sample type for these tests. **This is what you need to know:**
 - The WVDL requests that veterinarians send in **oral fluids samples (min. of 5 ml)** from **rope testing kits**.
 - These rope-testing kits are currently available via email to supplyroom@wvd.wisc.edu
 - Testing will be done on Wednesday (PCR) with 24-hour turnaround. Samples need to be at lab by noon prior to testing day. The 24-hour turnaround time may be extended if samples require repeat testing.
 - PRRS vaccinated animals will most likely be antibody POSITIVE.
 - Please see DATCP website for up to date information on testing regulations https://datcp.wi.gov/Pages/Programs_Services/SwineMovement.aspx

Additionally, the Virology section has recently validated pooled blood swab and serum testing for PRRS PCR. This was done to meet the needs of the boar stud industry.

Test	Sample Type	Cost (in state)	Test set up day	Result Available

PRRS PCR (NA and EU strains)	Oral Fluid	\$45	Wednesday	Within 24 hrs.
SECD PCR Panel**	Oral Fluid	\$65	Wednesday	Within 24 hrs.
Porcine Oral Fluid Panel***	Oral Fluid	\$80	Wednesday	Within 24 hrs.
PRRS PCR (NA and EU strains)	Blood swab /serum pool (up to 5/pool) or indiv.	\$45	Mon./Wed/Fri.	Within 24 hrs.

- WVDL virology section is continuing to perform numerous canine and feline PCR's for UW Shelter Medicine program. Recently, canine influenza outbreaks occurred in California <https://www.vetmed.wisc.edu/shelter-medicine-influenza-dogs/> and Oregon. Additional information on Canine influenza for Veterinarians can be found here: <https://www.vetmed.wisc.edu/civ-h3n2-info/>

NAHLN (National Animal Health Laboratory Network):

- WVDL has seen an increase number of samples as part of the Seneca Valley Virus Pilot study which includes SVA testing along with FMD testing during specific foreign animal disease (FAD) investigations.
- As part of the NAHLN network, WVDL was asked to provide microbiology assistance to support the California Animal Health & Food Safety (CAHFS) Laboratory during the ongoing Newcastle disease outbreak in CA. Earlier this Spring, Marc Chipault (Microbiologist) in the Virology section traveled to Davis, CA to assist in molecular testing in the BSL-3 laboratory. Marc assisted in the laboratory for 2 weeks. An additional employee from WVDL will be going out to perform APMV PCR testing in July. More information on the CA outbreak can be found here: https://www.cdafa.ca.gov/ahfss/animal_health/newcastle_disease_info.html

Staffing - The Virology section recently had several student employees graduate from UW-Madison. Left to Right: Michelle Gotteiner, Sophia Koehler and Abby Rothering. Michelle and Sophia will be starting Veterinary School at UW-Madison this Fall and Abby was recently hired as an Associate Microbiologist in the Virology section. (if you look closely, they are spelling WVDL!) Congratulations graduates, we are so thankful for their help in the section!



Pathology Sciences

2019 Veterinary Forensics Seminar and Autopsy Wet Lab - WVDL will be sponsoring a veterinary forensics seminar and wet lab on Sunday, November 3rd from 8 a.m.-5 p.m. This year's chosen speaker will be Dr. Melinda Merck who is a leader in the veterinary forensics field. She will provide a wealth of knowledge and an engaging experience for all those in attendance. The goal of this seminar is to provide training in an environment that promotes cohesiveness and teamwork in forensic casework amongst the target audience while enhancing their knowledge and skills in handling animal cruelty cases. The target audience for this seminar includes but is not limited to: Wisconsin DVMs, CVTs, SVM students, animal welfare groups, animal control, and law enforcement. The link to more information and registration is below; the seats will be limited and the wet lab is restricted to only 25 participants so registering as soon as possible is recommended.

The link to our registration is: <https://apps.vetmed.wisc.edu/cereg/>

Osmolality and Total Solids Testing Available at WVDL - The WVDL is excited to launch new testing to enhance diagnostic capabilities for veterinarians working up pre-weaned calf diseases. Regardless if dairy calves are fed milk, milk replacer, or a combination of both, calf raisers are increasingly adding more and more additives to the diet in an attempt to improve calf health and reduce the dependence on antimicrobial drugs. Depending on the type of carrier (often dextrose) used for the additives, this practice can lead to increased osmolality that often results in delayed gastric emptying and increased risk of abomasal disease and motility disorders.

Throughout validation and clinical experience with calf liquid feed analysis at the WVDL, we recommend that osmolality should always be run in conjunction with percent total solids testing to allow for better interpretation and ability to guide management decisions for the calf raising facility. On occasion, we have observed conflicting results with previous samples tested at various referral diagnostic laboratories. This is likely due to some of the components of the milk replacer solution, particularly the lactose, coming out of solution in storage. It is also possible that the samples were not initially mixed appropriately according to the manufacturers recommendations, leading to the incorrectly low osmometry results. We know that lactose can form crystals that are hard to get into solution, which is why it is best practice to heat the sample to 110° F and vigorously stir (no visible floccules) to make sure there is a homogenous solution prior to testing.

To avoid these problems and provide veterinarians and calf raisers with reliable diagnostic testing, the WVDL has purchased a freeze point depression osmometer that utilizes a strict protocol to control sample preparation (reheating in a water bath, stirring to achieve a homogenous solution) prior to testing. The WVDL now offers testing of waste milk (additives included), milk replacer, and electrolyte solutions for percent total solids and osmolality. The WVDL uses the Association of Official Agricultural Chemists (AOAC) approved method for percent total solids testing of milk replacer and milk samples. We performed an extensive validation on the percent total solids and osmolality testing to make sure the WVDL results are accurate and repeatable. It is important to point out that calf milk replacer solutions always have a higher osmolality than milk even when both solutions contain the same percent of total solids because it contains more lactose and sodium than milk.

Please contact us with questions regarding best practices for sample collection and submission to WVDL, or interpretation of results for management of calf nutritional problems.

Price structure

- \$10 accession fee added to each submission
- **1 to 5 samples:** \$35.00 (Osmolality) + \$7.00 (Total solids) = \$42.00 per sample
- **6 or more samples:** \$15.00 (Osmolality) + \$7.00 (Total solids) = \$22.00 per sample

Pricing examples

- **3-day feed trial** (2 feedings per day, total of 6 samples):
 - \$90 + \$42.00 totals solids + \$10 accession fee = \$142.00
- **5-day feed trial** (2 feedings per day, total of 10 samples):
 - \$150 + \$70.00 totals solids + \$10 accession fee = \$230.00

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www.wvdl.wisc.edu

Our mailing address is:

445 Easterday Lane
Madison, WI 53706
info@wvdl.wisc.edu

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