In This Issue

Director’s Corner
Mystery Photo
Disease Updates
- Rabies
- Blastomycosis
Bench Notes
- Winter weather closures
- Tissue submissions
- Case histories
- Horse necropsies
- Antimicrobial susceptibility testing
- Abortion submissions
Mini Case Report
- Canine enteric clostridiosis
Calendar
Staff Spotlight

A newsletter about diagnostic trends at the laboratory, animal health topics, interesting cases and new test offerings.

www.vdl.ndsu.edu

Feedback is always welcome. Please feel free to send your comments or suggestions to ndsu.vetlab@ndsu.edu and specify “newsletter” in the subject line.

NDSU Veterinary Diagnostic Laboratory

Happy New Year!
The earth has made a full rotation around the sun since I took over directorship last January. In that time, there have been many changes, along with opportunities to take on challenges that are currently affecting veterinary diagnostic laboratories across the country.

Several of the difficulties faced this last year include long-term staff retirements and faculty turnover, implementation of digital pathology, managing testing capabilities during global supply chain shortages, and the national outbreak of high pathogenic avian influenza.

Despite these hurdles, we hope we have met your needs. As always, please feel free to share your suggestions and comments on areas where we can improve. You can send us a message through the contact page on our website, or directly email ndsu.vetlab@ndsu.edu.

Best wishes for the upcoming year,

Heidi Pecoraro, DVM, Ph.D., Diplomate, ACVP
NDSU VDL Director and Veterinary Anatomic Pathologist

Mystery Photo

Thirty-five recently purchased feeder piglets show signs of ill-thrift and diarrhea. One 4-month-old female piglet is submitted for general investigation. There is thickened and necrotic mucosa in the large intestine.

What is your diagnosis and the etiology? Other enteric diseases in pigs?

Visit the NDSU VDL Website (www.vdl.ndsu.edu) to see the answers and read more about the case.
**Disease Updates**

**Rabies Virus**

The NDSU VDL conducted over 400 rabies tests in 2022. Of those, there were nine positive cases from North Dakota, South Dakota, and northern Minnesota. North Dakota had six rabid animals – two cats, two bats, one skunk, and one cow – from Barnes, Burleigh, Cass, Dickey, McHenry and Ransom counties.

Improper sample submissions resulted in 14 inconclusive results. Cross sections of the brain stem and cerebellum or hippocampus are required for a rabies test to be valid. Samples that are incomplete or too autolyzed cannot be validated.

The entire brain should be submitted fresh and whole for any neurologic case, but most especially for rabies testing. See the guide on the website on how to submit brain for rabies testing (www.vdl.ndsu.edu/wp-content/uploads/2019/10/Rabies-Guide.pdf).

Remember, rabies is 100% fatal once an animal or person starts to show clinical signs of infection.

**Blastomycosis**

In the last year, there have been over one dozen cats and dogs diagnosed with *Blastomyces dermatitidis* by the NDSU VDL.

Five of those cases have been dogs residing in North Dakota. The map shows the distribution of canine blastomycoses in North Dakota by county.

We reported two cats with ocular blastomycoses in our Summer 2022 newsletter.

In that newsletter, Dr. Quynn Steichen, the NDSU VDL veterinary pathology resident, reported that *B. dermatitidis* is found mainly in the Mississippi, Missouri, Ohio river valleys, and Mid-Atlantic states and that cases have been on the rise in Minnesota and North Dakota.

Moist soil conditions liberate *B. dermatitidis* conidia, which then can be subsequently aerosolized and inhaled by animals and people. After inhalation, spores travel to the lungs before disseminating throughout the body via blood or lymphatic vessels. The central nervous system and eyes are the most common sites of dissemination, and, thus, blastomycosis should be a differential in cases of sudden blindness and neurologic diseases.

**Winter weather closures** – Be sure to check our website and social media pages when there is a winter weather advisory. The NDSU VDL is closed whenever there is a campus-wide NDSU closure, as well as when 19th Avenue North in Fargo is shut-down. We will resume business as soon as we are able to reopen.

**Tissue submission** – Freeze artifact can severely impede evaluation of tissues and obscure important features that are necessary for an accurate diagnosis. During cold winter months with subfreezing temperatures, the addition of alcohol (isopropyl, methanol, or ethanol) at a mixture of 1-part alcohol to 9-parts formalin will help maintain integrity of the tissue. Additionally, the correct ratio of tissue to fixative solution (i.e., formalin, alcohol) is imperative for tissue preservation. There should be one part tissue to nine parts fixation solution. When sections are too large or there is not enough fixative in the containers, the tissue in the center of a sample becomes autolyzed (i.e., rotten), obscuring any lesions that may be present. Tissue sections should be less than 5 cm³ for postmortem cases. Larger biopsies can be placed directly in fixative solution but may need extra time for processing.

Tissue samples should never be larger than the opening of the container. Once the tissue is fixed, it becomes rigid and difficult to bend. Large tissue sections cannot easily be removed from containers with small openings. This results in damaged samples (and cranky pathologists). It may also incur additional processing fees due to the extra time it takes to remove the tissue from the container (e.g., dissecting tissues within containers, sawing open jars).

**Case histories** – Like other veterinary specialists, pathologists rely on patient history and clinical findings to make the most accurate diagnosis possible. Samples submitted without crucial information such as biopsy location or a description of the clinical signs noted prior to death can lead to misinterpretations and misunderstandings. Be sure to provide all the information requested on the submission forms, including any rule-outs. Additionally, pathologists are happy to review relevant clinical records, pictures, imaging, and other clinical data that will aid in making the proper diagnosis and, thus, treatment and prevention plans.

Continued.

**Bench Notes**

**Rabies Virus**

The NDSU VDL conducted over 400 rabies tests in 2022. Of those, there were nine positive cases from North Dakota, South Dakota, and northern Minnesota. North Dakota had six rabid animals – two cats, two bats, one skunk, and one cow – from Barnes, Burleigh, Cass, Dickey, McHenry and Ransom counties.

Improper sample submissions resulted in 14 inconclusive results. Cross sections of the brain stem and cerebellum or hippocampus are required for a rabies test to be valid. Samples that are incomplete or too autolyzed cannot be validated.

The entire brain should be submitted fresh and whole for any neurologic case, but most especially for rabies testing. See the guide on the website on how to submit brain for rabies testing (www.vdl.ndsu.edu/wp-content/uploads/2019/10/Rabies-Guide.pdf).

Remember, rabies is 100% fatal once an animal or person starts to show clinical signs of infection.

**Blastomycosis**

In the last year, there have been over one dozen cats and dogs diagnosed with *Blastomyces dermatitidis* by the NDSU VDL.

Five of those cases have been dogs residing in North Dakota. The map shows the distribution of canine blastomycoses in North Dakota by county.

We reported two cats with ocular blastomycoses in our Summer 2022 newsletter.

In that newsletter, Dr. Quynn Steichen, the NDSU VDL veterinary pathology resident, reported that *B. dermatitidis* is found mainly in the Mississippi, Missouri, Ohio river valleys, and Mid-Atlantic states and that cases have been on the rise in Minnesota and North Dakota.

Moist soil conditions liberate *B. dermatitidis* conidia, which then can be subsequently aerosolized and inhaled by animals and people. After inhalation, spores travel to the lungs before disseminating throughout the body via blood or lymphatic vessels. The central nervous system and eyes are the most common sites of dissemination, and, thus, blastomycosis should be a differential in cases of sudden blindness and neurologic diseases.
Horse necropsies – Reminder that the purpose of a postmortem examination is to determine cause of illness and/or death. Necropsy service is not to be used solely for carcass disposal. The NDSU VDL reserves the right to reject such cases. Private horse cremation is available, and necropsy is not required for this service. Check out the website for more information.

Changes to antimicrobial susceptibility testing (AST) – AST is no longer routinely performed on client-submitted (pre-paid) samples. In these cases, clients are not typically working with a licensed veterinarian; therefore, AST results will no longer be routinely provided. Instead, significant bacterial isolates will be reported to the owner with a note indicating that a veterinarian must request AST from the laboratory. AST will be performed for an additional charge and will be available for up to five days after a final report has been released.

Abortion work-ups – Many animals are pregnant this time of year. Unfortunately, that also means abortions can occur in herds and flocks. In general, in-house postmortem examinations of aborted fetuses have higher rates of agent recovery and identification compared to field-performed necropsies.

If an aborted fetus cannot be submitted to the NDSU VDL for pathologist examination, the following tissues should be submitted for optimal results:
- Fresh and formalin-fixed placenta
- Whole fetus/neonate (recommended) or fresh and fixed tissues, including:
  - Fresh brain, heart, lung, liver, kidney, spleen, abomasal fluid, thymus, thyroid, lymph node, fetal eyeball/vitreous humor (for nitrate analysis), placenta and any tissue with a suspected lesion
  - Formalin-fixed placenta, brain, thymus, heart, lung, liver, kidney, spleen, conjunctiva, adrenal gland, skeletal muscle and any tissue with a suspected lesion
- Feed and water
- Maternal sera (acute and convalescent)

Postmortem examination revealed a 30 cm segment of distal jejunum and ileum, extending to the ileal cecal region, that was dark red and hemorrhagic.

Microscopically, within the small intestine, was diffuse coagulative necrosis of the intestinal mucosal villi characterized by loss of differential staining and retention of cellular architecture. Transmurally, the mucosa, submucosa, tunica muscularis and serosa were markedly expanded by hemorrhage, edema and fibrin. There were also moderate numbers of large bacilli scattered throughout the necrosis.

Transmural necrosis and hemorrhage, along with large bacilli, were consistent with clostridiosis. Cultures and PCR identified Clostridium perfringens Type F as the cause of enteritis and subsequent death.

C. perfringens is a gram-positive, spore-forming anaerobe that mediates disease by the production of protein toxins. Historically, C. perfringens has been classified based on its ability to produce varying combinations of four major typing toxins: alpha, beta, epsilon, and iota. However, additional toxins, such as enterotoxin, NetB, NetF, NetE, beta2, may play a critical role in virulence of C. perfringens, and thus the traditional typing (Type A-E) has been expanded to include Type F and G.

C. perfringens Type F produces alpha toxin and enterotoxin (cpe), but not beta, epsilon, or iota toxins. The alpha toxin acts on the cell membrane (lecithinase) and produces hemolysis and necrosis of cells. Enterotoxin is secreted and freed into the intestinal lumen where it binds to the intestinal epithelium and exerts cytotoxic action, ultimately causing villous shortening and intestinal epithelial desquamation.

Type F strains are the causative agent in human food poisonings, and possibly play a role in enteric diseases in horses, dogs, and other species. C. perfringens type A is the causative agent in canine hemorrhagic diarrhea syndrome.

Enteric clostridiosis does not typically require antimicrobial therapy. Clinical signs are due to the toxins produced by C. perfringens overgrowth, rather than the organism itself. Replacement fluids and electrolyte administration are recommended over antibiotics.
Staff Spotlight

Kicking off the new year’s staff spotlight is one of our microbiologists. Sarah Gefroh, MLS(ASCP)SM has been with the VDL since 2021 but has worked as a medical laboratory scientist (MLS) for almost 20 years. Sarah brings her microbiology expertise and medical laboratory knowledge to the VDL every day. She is also our social media specialist and is responsible for keeping up our posts on Facebook and LinkedIn.

Do you fold your pizza? Sure

Favorite Christmas present when you were a kid? A balance beam made by my grandpa.

What is the worst fashion decision you ever made? I decided in the 7th grade that I needed a more fashionable hairstyle and I decided to trim my bangs myself. It was NOT a good decision.

If you could time travel, where is the first place you would go? Back to 7th grade to stop myself from cutting my hair!

Would you rather be at the beach or in the mountains? Beach

Calendar: Winter-Spring Closures

January 16 – Martin Luther King Jr. Day
February 20 – Presidents Day
April 7 – Good Friday
May 29 – Memorial Day

Staff Spotlight

Sarah Gefroh
VDL Microbiologist
(© Photo by Kelly Benson, VDL Chemist)