Laboratory Services Division
Animal Health Laboratory

AHL Newsletter

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In this issue:

AHL Newsletter
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Editor: **Maria Spinato,** DVM, DVSc, Diplomate ACVP, MBA
Editorial Assistants: **Helen Oliver, Sofija Jelacic**

The *AHL Newsletter* is published quarterly (March, June, September, December) by the Animal Health Laboratory, Laboratory Services Division, University of Guelph.

Its mission is to inform AHL clients and partners about AHL current activities, and laboratory-based animal disease events and disease trends. *All material is copyright 2022.* *Ideas and opinions expressed herein do not necessarily reflect the opinions of the University or the Editor.*

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Mailing address & contact information:
Animal Health Laboratory
Laboratory Services Division, University of Guelph
Box 3612, Guelph, Ontario, Canada N1H 6R8
Phone: (519) 824-4120 ext. 54538; fax: (519) 821-8072

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AHL holiday hours 2022/2023

Except for Sun. Dec. 25 (closed – no service), AHL-Guelph is open every day from Fri. Dec 23rd until Mon Jan 2, 2023 with limited services. The University of Guelph is officially closed during this period. * **Please note: closing date to run invoices will be Tuesday December 20, 2022.***

Thurs. Dec. 22	Guelph and Kemptville - All laboratory sections open with full service
Fri. Dec. 23	All laboratory sections open with limited services (Guelph - 9:00 am to 5:00 pm; Kemptville 8:30-4:30 pm)
Sat. Dec. 24	Guelph: specimen receiving, emergency mammalian postmortems, full bacteriology set-up, as well as clinical pathology testing; Kemptville closed
Sun. Dec. 25	Guelph and Kemptville laboratories closed
Mon. Dec. 26	Guelph: specimen receiving, emergency mammalian postmortems; Kemptville closed
Tues. Dec. 27	Guelph: specimen receiving, emergency mammalian postmortems; Kemptville closed
Wed. Dec. 28	All laboratory sections open with limited services (Guelph - 9:00 am to 5:00 pm; Kemptville 8:30-4:30 pm)
Thurs. Dec. 29	All laboratory sections open with limited services (Guelph - 9:00 am to 5:00 pm; Kemptville 8:30-4:30 pm)
Fri. Dec. 30	All laboratory sections open with limited services (Guelph - 9:00 am to 5:00 pm; Kemptville 8:30-4:30 pm)
Sat. Dec. 31	Guelph: specimen receiving, emergency mammalian postmortems, full bacteriology set-up, as well as clinical pathology testing; Kemptville closed
Sun. Jan. 1	Guelph: specimen receiving, emergency mammalian postmortems; Kemptville closed
Mon. Jan. 2	Guelph: specimen receiving, emergency mammalian postmortems; Kemptville closed
Tues. Jan. 3	Guelph and Kemptville - All laboratory sections open with full service

Guelph **drop box and fridges available 7AM to 10PM** and Kemptville **drop box and/or fridges are available 365/24/7** for specimen drop off.

For full details, please see AHL website – www.ahl.uoguelph.ca

Update from the Director

The view from the Director's office

Sometimes, it seems as if we are living in a modern version of the movie *Groundhog Day*. As the holiday season approaches again this year, there more warnings of respiratory illness outbreaks – influenza and RSV reappearing after a hiatus during COVID – more vulnerable people filling the hospital emergency rooms, and more concerns about how best to prevent the spread of infections while attempting to keep children in school, people at work, and maintaining holiday traditions and in-person celebrations. How we exit this reiterating scenario is anyone's guess, but humans are infinitely resourceful, and we'll probably figure it out somehow.

This issue's 'staff highlights' section shines the spotlight on Dr. Jim Fairles, our Client Services Veterinarian who is the 2022 Carl Block award recipient. This prestigious award is presented by Animal Health Canada to individuals who have contributed significantly to the advancement of animal health and welfare programs and initiatives throughout Canada. Jim has dedicated his professional career to these principles: first as a practicing food animal veterinarian; subsequently during his Client Services position here at the AHL; and finally, as a member of the many agriculture boards and veterinary organizations on which he serves. Congratulations Jim!

This year marks the 25th anniversary of the formation of Laboratory Services Division at the University of Guelph. It was 25 years ago that the Animal Health Laboratory and the Agriculture and Food Laboratory were devolved from the Ontario Government to the University of Guelph. Our motto for this year's celebration – suggested by several staff members – is "proudly serving". And indeed, we all are proud of the service that we continue to provide in support of animal health and welfare, food safety and environmental stewardship in this province.

From all of us at AHL, we send our very best wishes to you and your families for continued health and safety, and our hope that you have some time off to connect and relax during the holiday season.

Maria Spinato, Director

Animal Health Laboratory, University of Guelph, Guelph, ON.

Specimen Reception update

Jim Fairles

Animal Health Laboratory, University of Guelph, Guelph, ON

Cold weather shipping reminder

It's that time of year again when we need to start thinking about preventing samples from freezing. Specimens such as EDTA blood are rendered useless when frozen. Formalin will also freeze, creating artifacts in fixed tissue. It can be difficult to protect samples shipped during the winter from severe cold. Even 10% neutral-buffered formalin will freeze in harsh winter weather conditions. To prevent formalin freezing, add 1 mL of ethanol per 10 mL of formalin.

Samples that should not be frozen should be shipped inside insulated containers with minimal cold packs. Use of room temperature cold packs will help prevent temperatures from dipping too low. If you have any concerns about the best way to ship critical samples, please contact the AHL. ahlinfo@uoguelph.ca

Humane transport of live animals for postmortem

The AHL encourages submission of live animals for immediate euthanasia and postmortem, to ensure a high rate of diagnostic accuracy in poultry cases, and in outbreaks of diarrhea in young pigs and ruminants only. **We remind submitters that live animals must be transported humanely.** The Provincial Animal Welfare (PAW) Act requires that veterinarians report all suspected cases of animal abuse to the PAWS (Provincial Animal Welfare Services). AHL veterinarians will confer with PAWS inspectors to determine if an investigation is warranted in suspected cases of inhumane transport.

Examples of inappropriate restraint and transportation methods include:

- hog-tying calves, adult sheep or goats;
- baby pigs or chickens placed in sealed plastic tubs or styrofoam containers;
- pigs or chickens submitted in tied feed sacks;
- compromised animals in open air (back of a truck) in any type of weather.

Examples of acceptable transport containers include:

- dog or cat kennels for small pigs;
- cardboard boxes of appropriate size with ventilation holes for baby pigs or chickens;
- poultry crates;
- large dog kennels or enclosed bedded truck cabs for larger pigs, calves, small ruminants.

All containers should be of sufficient size to avoid crowding or smothering, and animals should be protected from extreme temperatures. Animals that cannot be transported humanely should be euthanized on-farm, and the practitioner is encouraged to collect appropriate samples for diagnostic testing. **Please contact the AHL and request a consultation with a pathologist if assistance is required for sample selection when performing on-farm postmortems**.

Staff highlights

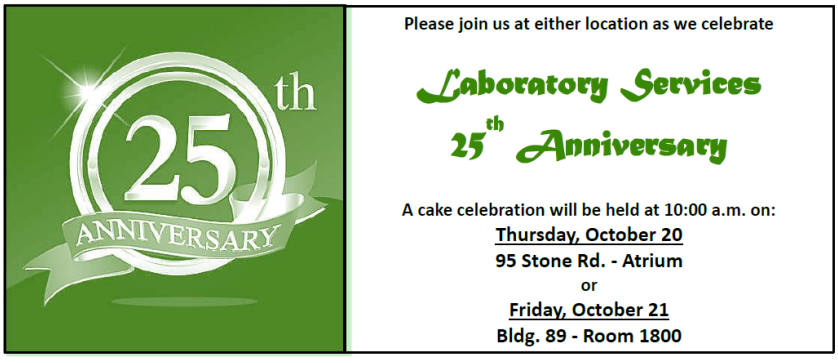
At the Animal Health Canada meeting in September, Dr. Jim Fairles was the recipient of this year's Carl Block Award. This prestigious award is presented to an individual for outstanding contributions in the field of livestock animal health in memory of Carl Block, a cattleman whose tireless dedication to Canadian agriculture and commitment to animal health will long be remembered.

"The vision of AHC is inspired by individuals who demonstrate outstanding contribution by their actions and behaviour. AHC is honored to bring to light the contribution of some of those individuals who often work in the shadow, but generate real change in the Canadian landscape of Animal Health and Welfare, and public-private collaboration" Dr. René Roy, AHC industry Co-chair

Dr. Jim Fairles receiving the Carl Block award, presented by Dr. René Roy and Dr. Maria Spinato

LSD 25th anniversary celebration

2022 is the 25th anniversary of the amalgamation of the Animal Health Laboratory and the Agriculture and Food Laboratory, forming the Laboratory Services Division at the University of Guelph. A celebration was held to mark this auspicious event, and to show appreciation for all staff whose contributions continue to be instrumental in supporting our 25th anniversary motto – "proudly serving". Congratulations LSD!



OAHN Update – December 2022

Mike Deane, Tanya Rossi

Animal Health Laboratory, University of Guelph, Guelph, ON

This fall, OAHN has been working on new vet-focused resources, finishing the 2022 Varroa Monitoring Campaign, running the OAHN Equine webinars, and creating our normal quarterly vet and owner/producer reports. Additionally, we have been planning the OAHN AGM, set to take place (virtually) on January 25th. We are gearing up for a busy winter, with many more veterinary resources to come. To view the reports, go to OAHN.ca and navigate to the species you are interested in.

New Resources

- 2022 OAHN Public Health Update
- Rabbit Hemorrhagic Disease Surveillance Project
- OAHN Equine Webinars 2022
- OAHN Bovine Network Quarterly Update October 2022
- Highly Pathogenic Avian Influenza Resources 2022
- Aquatic Network Research Project: Biosecurity for aquatic animal facilities – content development for a website and workshop, thinkific platform and booklet

Video: Postmortem Examination in Swine

As part of the OAHN Swine Smallholder project, AHL pathologist Dr. Josepha Delay developed an in-depth explanation of how to perform a swine postmorterm, as well as providing information on the tools used and samples taken. Find the video here: https://www.oahn.ca/resources/postmortem-examination-in-swine/ (registration and log-in to OAHN is required).

New Reports

Most OAHN networks create reports once per quarter. To view any of the veterinary reports below, please click on the OAHN icon for each network, or go to OAHN.ca and navigate to the species you are interested in.

- Surveillance: Q2 Animal Health Laboratory data
- New strain of winter dysentery – Quebec
- *Mannheimia hemolytica* outbreak in a lactating dairy herd
- **Salmonella** Dublin diagnostics guide

- Fowl adenovirus microneutralization summary (September 2021 to August 2022)
- Poultry veterinary survey highlights – Q3 2022

- Disease discussion
- Laboratory diagnostic reports
- Ontario slaughter statistics
- OVC research
- CanSpot ASF surveillance update
- International disease topics of interest summary

- Habronemiasis ("summer sores") – do we have it in Ontario?
- Network member reports
- Syndromic and lab surveillance dashboard
- Equine research

- OAHN survey: CIRDC, parvovirus
- Imported dog distemper, titres study
- RHDV2 surveillance study
- Rabies update
- Avian flu order
- Pet pig, monkeypox resources for vets OAHN survey: CIRDC, heartworm, RHDV2

Ontario Interactive Animal Pathogen Dashboards

We at the AHL are pleased to announce the launch of the Ontario Interactive Animal Pathogen Dashboards!

What are the Ontario Interactive Animal Pathogen Dashboards (IAPD)?
The IAPD is an initiative by the Animal Health Laboratory (AHL) at the University of Guelph with funding from the Food for Thought program. These dashboards were developed to monitor and report current and historical trends in pathogens of special interest to the food animal industry. The project's goal is to support users in developing appropriate strategies to deal with endemic, emerging, or foreign animal pathogens in Ontario by keeping them informed with up-to-date laboratory diagnostic data.

Who can access the dashboards?
These dashboards are designed for individuals working in the food animal industry; therefore, access will only be granted to producers, veterinarians, and government officials working in agriculture.

How do I register?
If you would like access to the dashboards please contact us at iapd@uoguelph.ca and include your name, contact information, and affiliation with the food animal industry.

Samples of two dashboards depicting PRRSV submissions (**Fig. 1**) and *Salmonella* Dublin (**Fig. 2**) cases over the past 10 years are presented below:

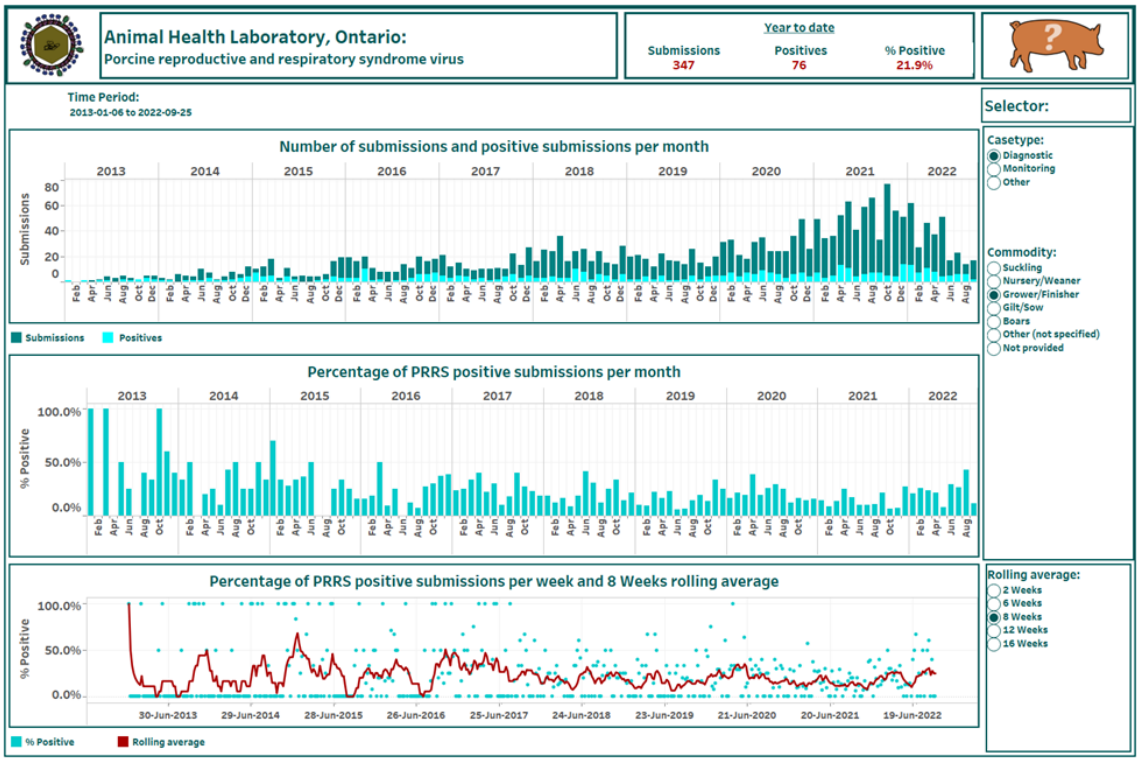


Figure. 1. PRRSV dashboard illustrating number of PRRSV PCR submissions, percentage positive and rolling average.

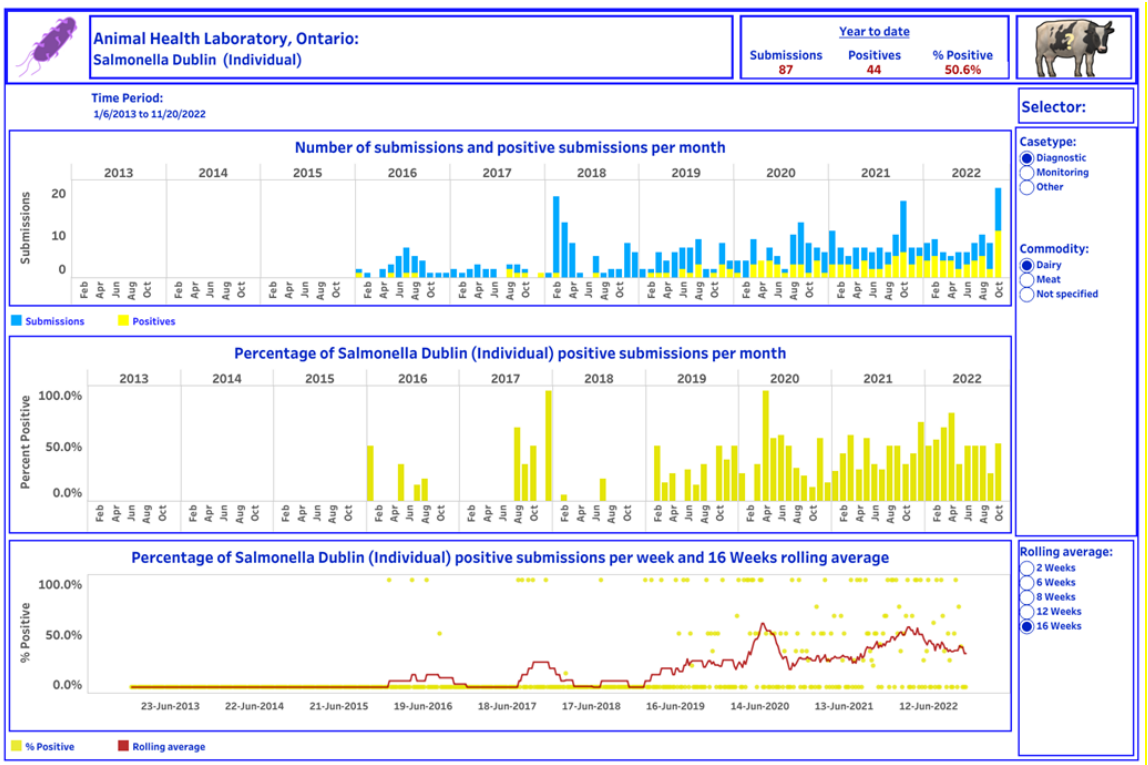


Figure. 2. *Salmonella* Dublin dashboard illustrating number of individual cow serum and milk ELISA tests, percentage positive and rolling average.

Part 5 – Antimicrobial susceptibility testing – Results reporting

Đurđa Slavić

Animal Health Laboratory, University of Guelph, Guelph, ON

AHL Newsletter 2022;26(4):9

In the series of articles about antimicrobial susceptibility testing, AHL published information about test methods, selection of bacterial organisms, selection of antimicrobials for antimicrobial susceptibility testing (AST) and laboratory results interpretation. In this fifth and the final article of this series, result reporting is discussed. The full document is available on the AHL website as LabNote 64: https://www.uoguelph.ca/ahl/ahl-labnote-64-antimicrobial-susceptibility-testing-ast

As per CLSI: "Veterinarians should understand that the presence of AST results for a drug on a laboratory report does not provide permission or the necessary justification for use of that drug. The presence of the drug on the panel does not indicate that it is legal or appropriate to use." It is the responsibility of a veterinarian to use the drugs as per their label recommendations, as laboratories do not provide separate panels for different commodity groups (i.e., beef vs dairy), and sometimes, one antimicrobial panel is shared with different animal species (e.g. bovine/porcine panel).
If the drug is excluded from the report, it is likely that:

1. The bacterial organism is intrinsically resistant to it and there is no need to perform AST (**Table 1**);
2. There are no CLSI guidelines available for that particular organism/drug combination; or
3. The drug is not approved for use in that food animal species. *AHL*

Table 1. Examples of intrinsic resistance in *Enterobacterales* and *Enterococcus* spp.

Organism	Antimicrobial agent
Enterobacterales	Clindamycin, fusidic acid, glycopeptides, macrolides, rifampin
Enterococcus spp.	Aminoglycosides, cephalosporins, clindamycin, trimethoprim, trimethoprim-sulfamethoxazole

Reprinted with permission from Clinical and Laboratory Standards Institute from: CLSI. Understanding Susceptibility Test Data as a Component of Antimicrobial Stewardship in Veterinary Settings. 1st ed. CLSI report VET09. Wayne, PA: Clinical and Laboratory Standards Institute; 2019.

References

1. CLSI. Performance standards for antimicrobial disk and dilution susceptibility tests for bacteria isolated from animals. 5th ed. CLSI supplement VET01S. Clinical and Laboratory Standard Institute; 2020.

2. CLSI. Understanding susceptibility test data as a component of antimicrobial stewardship in veterinary settings. 1st ed. CLSI report VET09. Clinical and Laboratory Standard Institute; 2019.

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RUMINANTS

Chronic proliferative rhinitis associated with *Salmonella enterica* subsp. *diarizonae* infection in a ewe

Margaret Stalker, Josepha DeLay, Amy Gaw

Animal Health Laboratory, University of Guelph, Guelph, ON (Stalker, DeLay), Temiskaming Veterinary Service (Gaw)

AHL Newsletter 2022;26(4):10.

An adult ewe experienced an exacerbation of dyspnea shortly after lambing. The producer had previously noted episodes of an "everted nostril" over the past few years. On physical exam, pink tissue protruded from the left nostril, with obstruction of air flow. The ewe was subsequently euthanized and the head was submitted to the AHL for further diagnostic workup.

On external examination, the left side of the nose appeared swollen, with an approximately 2 cm diameter pale smooth-surfaced pink mass protruding from the nostril. Sagittal sectioning of the head revealed a large 5 cm x 15 cm multilobulated tan-pink smooth-surfaced tissue filling the entire left nasal cavity, encompassing the turbinates and extending to the ethmoid turbinate region and into the nasopharynx (**Fig. 1**). The provisional diagnosis was enzootic nasal tumor.

Histologic sections of the mass revealed well-differentiated nasal respiratory epithelium overlying a fibrous stroma populated by scattered dilated glandular structures surrounded by large numbers of plasma cells, with fewer lymphocytes and neutrophils (**Fig. 2**). The glandular structures were lined by well-differentiated pseudostratified columnar epithelium with mucin secretion. Closer examination of the respiratory epithelium revealed myriad faintly visible intracytoplasmic bacilli (**Fig. 3**). Tissue Gram stains confirmed these as gram-negative bacteria.

A tentative diagnosis of chronic proliferative rhinitis was made, and a sample of frozen nasal epithelium was submitted for bacterial culture. Routine culture isolated 2+ *Salmonella* sp. from the nasal epithelium. The isolate was subsequently serotyped as *Salmonella enterica* subspecies *diarizonae* serotype 61:k:1,5,7 (SED).

Chronic proliferative rhinitis associated with intracytoplasmic *Salmonella* infection of nasal epithelium is an unusual disorder of sheep. The condition was first reported in the USA in 1992. Since then, cases have also been reported in Spain and Switzerland, again associated with SED infection. SED is considered a host-adapted bacterium of sheep, and 66.4% of US sheep flocks are culture-positive asymptomatic carriers of this organism. The bacterium is likely maintained in flocks through fecal-oral transmission, although colonization of the nasal cavity may indicate nasal shedding is also possible, as in this case. An opportunistic pathogen with sporadic reports of enteritis in lambs and abortion, SED is also potentially zoonotic, although human risk is more typically associated with exposure to infected reptiles that also carry the pathogen. *AHL*

References

1. Meehan IT, et al. Chronic proliferative rhinitis associated with *Salmonella arizonae* in sheep. Vet Pathol 1992;29:556-559.
2. Lacasta D, et al. Chronic proliferative rhinitis associated with *Salmonella enterica* subspecies *diarizonae* serovar 61:k:1,5,(7) in sheep in Spain. J Comp Path 2012;147:406-409.
3. Pritchard J. *Salmonella arizonae* in sheep. Can Vet J 1990;31:42.
4. *Salmonella* in US Sheep Operations, 2011. USDA-APHIS, Veterinary Services Info Sheet. June 2013.

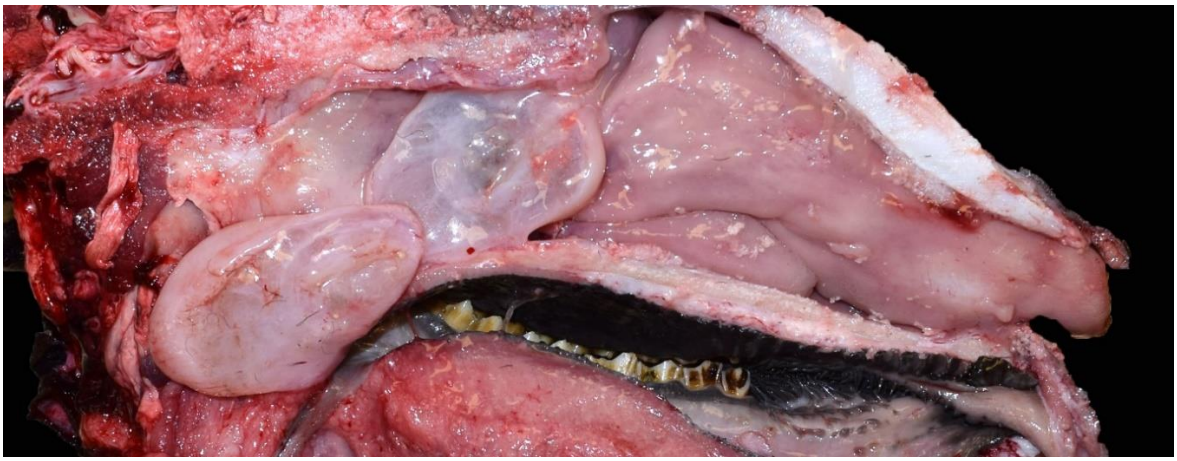

Figure 1. Pale pink edematous tissue filling and occluding the left nasal cavity of a ewe.

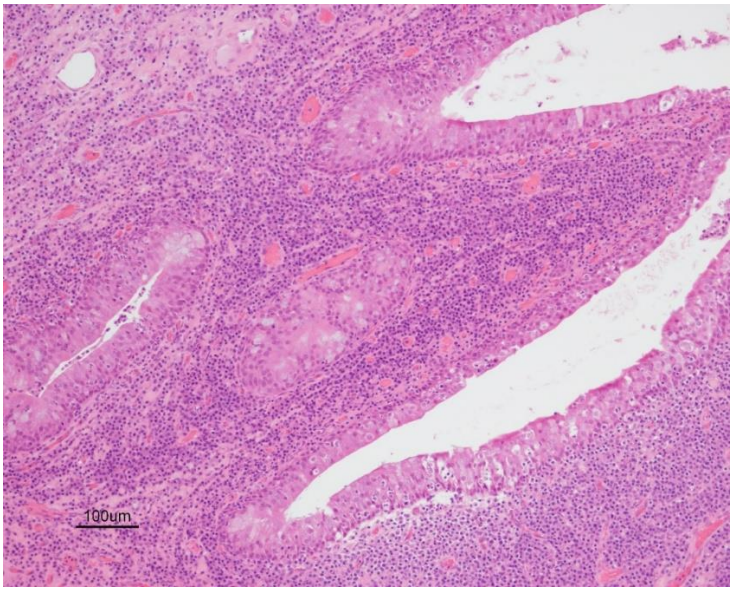

Figure 2: Chronic rhinitis with marked plasmacytic inflammation (H&E, 10X).

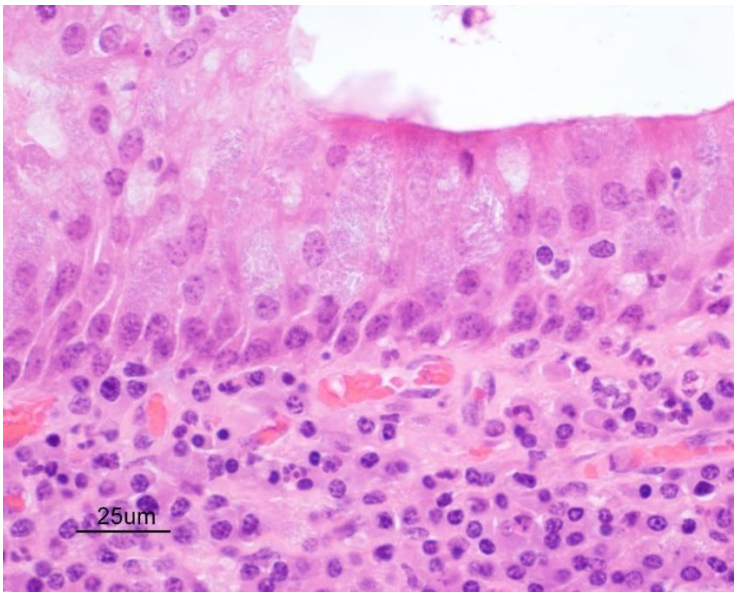

Figure 3: Numerous intracytoplasmic bacilli within nasal respiratory epithelium (H&E, 60X).

Ovine abortion due to *Actinobacillus seminis* and *Histophilus somni*

Meegan Larsen

Animal Health Laboratory, University of Guelph, Guelph, ON

AHL Newsletter 2022;26(4):12.

Several ewes aborted approximately one month prior to the expected due date. Twin fetuses and their placenta were submitted to AHL for postmortem examination. No significant gross lesions were noted. Microscopically, there was necrosis and neutrophilic inflammation in the placental chorionic villi with intralesional bacteria and evidence of vascular injury (**Fig. 1**). No significant microscopic lesions were seen in the two fetuses. Large numbers of *Actinobacillus seminis* and *Histophilus somni* were isolated from the placenta, and with supportive microscopic lesions of necrotizing and suppurative placentitis with vasculitis, abortion was attributed to infection with these two bacterial species.

Actinobacillus seminis is recognized as a common cause of epididymitis and associated infertility in rams. Metritis, mastitis, posthitis and polyarthritis have also been attributed to infection by this bacterium in sheep. *A. seminis* was first documented as a cause of abortion in an outbreak on a sheep farm in the United Kingdom in 1999, and was recently identified as a small ruminant abortifacient bacterium for the first time in the United States in a goat herd (personal communication with Maria Spinato regarding a report at the 2022 AAVLD Conference). *Histophilus somni* is a commensal bacterium of mucus membranes in ruminants and a possible, although uncommon, cause of abortion in small ruminants. Placental lesions with *A. seminis* abortion tend to be those of suppurative inflammation of the membranes and cotyledons, whereas *H. somni* infection is typically associated with vascular injury. Both microscopic lesions were seen in the placenta from the aborted lambs in this case, and thus both bacterial species are considered contributory etiologic agents. *AHL*

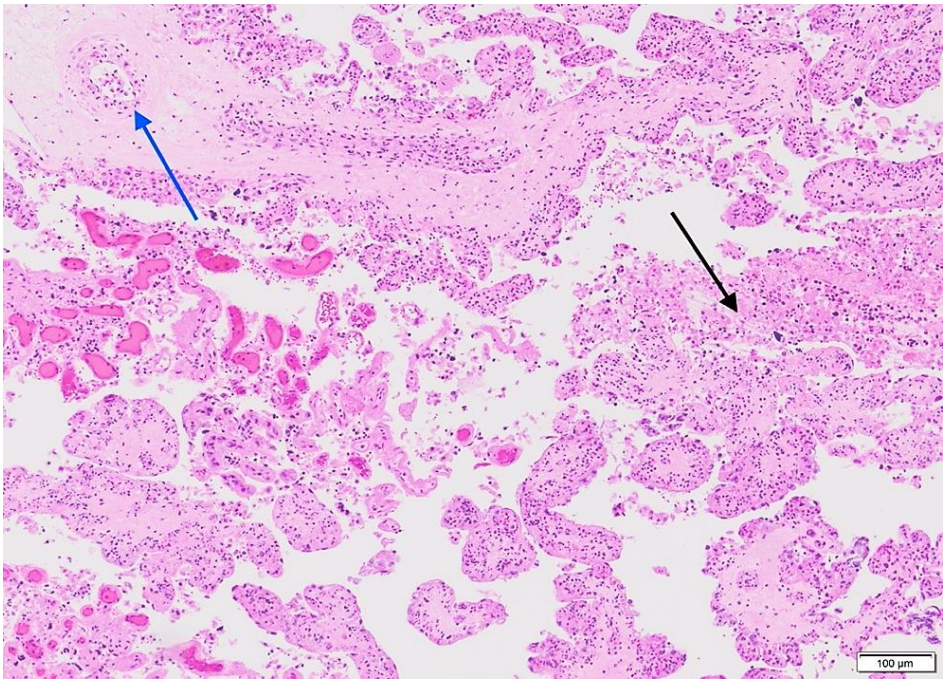

Figure 1. Fibrin and neutrophilic inflammation (black arrow) and inflammation of blood vessel wall (blue arrow) in histologic section of placenta. H&E.

Reference

1. Foster G, et al. *Actinobacillus seminis* as a cause of abortion in a UK sheep flock. Vet Rec 1999;144:479-480.

SWINE

Tackling negative diagnostic test results: Focus on respiratory disease

Josepha DeLay, Davor Ojkic

Animal Health Laboratory, University of Guelph, Guelph, ON

AHL Newsletter 2022;26(4):13.

New pathogens, and new roles for known pathogens, are at the forefront of swine disease diagnostics and research. As exciting as it is to suspect a new disease syndrome, it is important to be thorough in the approach to diagnostic testing, so as not to miss more common pathogens.

Results of diagnostic testing for clinical respiratory disease can sometimes be unrewarding and frustrating. Diagnostic test results are influenced by many factors, including the specific animals and tissues tested, the time point of sampling in the disease course, the specific tests and methodologies selected for a case, and the potential involvement of new or emerging pathogens.

Bias in sampling, test selection, and result interpretation can be easily introduced at all levels of disease investigation, and veterinarians and pathologists must be vigilant in avoiding bias at all times. The economics of diagnostic testing can play a role in introducing bias as well, as we try to minimize sample and test numbers to save money. To avoid these biases and to reach a valid conclusion, the approach to disease diagnosis must be logical, well planned, and thorough.

Important points to consider in any diagnostic workup include the following:

Select acutely affected, untreated animals for sampling for diagnostic tests. This applies for both live-animal and postmortem sampling. In animals with subacute or chronic disease, the pathogen may be present at reduced levels or may be cleared, potentially leading to negative test results and erroneous conclusions regarding the cause of disease. In samples from animals recently treated with antibiotics, bacterial growth will be negatively impacted by the drug, again producing misleading test results. The number of animals tested is also important. Choosing 3 animals for sampling will optimize the chances for identifying common pathogens among affected animals (finding the 'common disease denominator') while being relatively economical.

Take appropriate samples from the selected animals, and treat the samples kindly. For respiratory cases, examine the entire respiratory tract, including trachea, mainstem bronchi, and both lungs. Note the character and distribution of gross lesions, and communicate these findings on the submission form. Include an estimate of the percentage of lung parenchyma or tracheal mucosa involved in the disease process. For bacterial culture and PCR tests, sample at the margin of affected and unaffected lung parenchyma. Submit a fresh sample of trachea for potential testing. For histopathology, include trachea and multiple samples from both left and right lung (e.g., 1 sample from both cranial and caudal lobes from each lung), also capturing the interface between normal and abnormal lung. Be gentle when sampling, avoiding crush injury with forceps or scissors. Place fresh samples for culture and PCR in individual whirl-pak or other sterile bags that can be sealed, and keep these samples chilled. Place samples for histopathology in a sufficiently large screw-top formalin jar to ensure there is a 1:10 ratio of tissue:formalin to allow rapid and complete fixation.

Select specific diagnostic tests that will answer the questions for a particular clinical scenario, and include histopathology whenever possible. Initial test selection (various PCRs, bacterial culture) should be based on the clinical differential diagnoses. Histologic lesions can provide valuable evidence to confirm or exclude these differentials, and can be correlated with PCR and culture results. When test

results are negative, histologic features can help fine-tune additional testing. The lesions will categorize the disease process regarding pathogenesis and likely etiologies; e.g., inhaled vs blood-borne infection, bacterial pathogens vs epitheliotrophic viral injury, etc. Histologic samples also allow for direct pathogen detection using immunohistochemical (IHC) or *in situ* hybridization (ISH) techniques. These direct tests permit visualization of a pathogen within a lesion, and can confirm diagnoses suggested by PCR results.

A negative test result isn't the end of the story. In the face of histologic lesions and negative test results for more common pathogens, consider known emerging pathogens or novel pathogens. Current research at other institutions has identified involvement of porcine astrovirus type 4 (PoAstV4) in tracheitis and bronchitis in young pigs, and porcine hemagglutinating encephalomyelitis virus (PHEV) in tracheitis in grow-finish pigs. Porcine parainfluenza virus has been detected in some swine pneumonia cases, although associated lesions are not well described. Porcine respiratory coronavirus is recognized but infrequently investigated. Specific PCR tests for these viruses are available at external laboratories. We don't yet know the significance of these potential pathogens in the Ontario swine herd, but diagnosticians do consider these agents in compatible disease situations. For cases where available tests do not identify a pathogen to explain clinical disease and pathologic findings, metagenomic analysis by whole genome sequencing is an option for high-level investigation of novel pathogens.

Communication between the veterinarian and lab diagnosticians is important at every step of the diagnostic process. This includes communicating basic information, such as the clinical history and gross postmortem lesions, to more in-depth discussions when advanced diagnostic testing is warranted.
AHL

Bacterial arthritis in broiler breeder chickens

Andrew Brooks, Emily Martin, Siobhan O'Sullivan, Tanya Rossi

Animal Health Laboratory, University of Guelph, Guelph, ON

AHL Newsletter 2022;26(4):15.

Lameness due to bacterial infection is a common problem in poultry. This article summarizes the features of bacterial arthritis in broiler breeder chickens in 55 postmortem submissions to the AHL in 2022.

The clinical histories frequently described leg problems, lameness and swollen hocks. Several submissions also reported ongoing or persistent lameness, foot lesions, increased mortality due to culling, and reduced production. The age of the affected birds varied from 1 day to 37 weeks, with an average age of 20 weeks. The clinical problems involved both roosters and hens in approximately half of the submissions.

At postmortem examination, severe arthritis and tenosynovitis were consistently observed in the hock joints. The affected joints typically contained turbid, yellowish inflammatory exudate that extended along the gastrocnemius tendons (**Fig. 1A**). The shoulder and stifle joints were affected less frequently. Many birds also had lesions indicating septicemia, such as splenomegaly. Less common findings included tendon rupture, osteomyelitis (e.g., tibiotarsal bone), femoral head necrosis, sternal bursitis, and pododermatitis. Histologically, the affected joints and tendons typically contained severe inflammation with exudates of fibrin, necrotic heterophils and colonies of bacteria (**Fig. 1B**). Amyloidosis of the joint capsule, tendon sheath or liver was noted in some submissions with chronic infections.

Staphylococcus aureus was the most frequent pathogen cultured from the joints in this case series (48 submissions), followed by *E. coli* (18 submissions), *Enterococcus cecorum* (4 submissions) and *Pseudomonas aeruginosa* (1 submission) (**Fig. 2**). Mixed infections, with two or more pathogens isolated from the joints, were present in 40% of the submissions. Although *Pasteurella multocida* was isolated occasionally, in each circumstance the flock had been vaccinated against fowl cholera.

S. aureus is a common cause of arthritis, tenosynovitis, septicemia, osteomyelitis, omphalitis and gangrenous dermatitis in poultry. The diagnosis of staphylococcosis is confirmed by culturing *S. aureus* from the typical lesions. Staphylococci are ubiquitous in the environment and frequently colonize the skin, therefore, damage to the skin barrier may permit entry of bacteria and systemic infection. Immunosuppression due to conditions such as infectious bursal disease or chicken infectious anemia may also predispose to staphylococcosis. Prevention strategies include management practices that enhance host immunity, minimize damage to the skin and feet, and minimize environmental contamination. *Staphylococcus* infection may also be treated with antibiotics based on the results of sensitivity testing. The antibiotic resistance profile of *S. aureus* isolated from broiler breeder chickens at the AHL from 2020 to 2022 is summarized in (**Fig. 3)**. *AHL*

Figure 1. (A) Hock joint from a broiler breeder chicken with bacterial arthritis. The joint lumen contains a yellow, turbid inflammatory exudate. **(B)** Histopathology of the gastrocnemius tendon (*) with severe tenosynovitis due to *S. aureus* infection. The tendon sheath contains abundant inflammatory exudate composed of fibrin and necrotic heterophils (arrows). **Inset:** Colonies of coccoid bacteria (arrowheads) within the exudate. H&E.

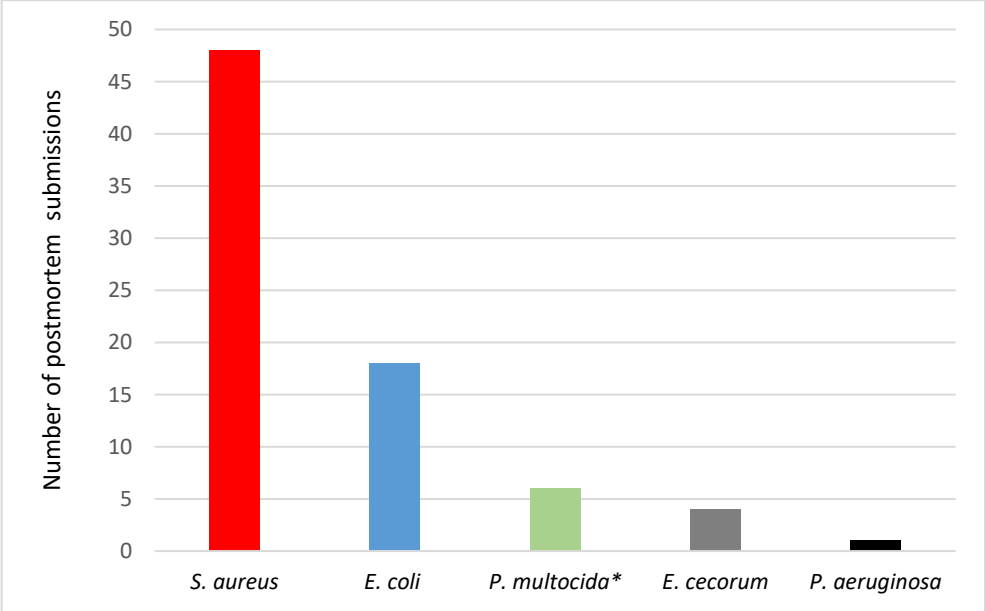

Figure 2. Bacterial etiologies of arthritis identified in broiler breeder chickens submitted for postmortem to the AHL in 2022. **P. multocida* isolates were from vaccinated flocks.

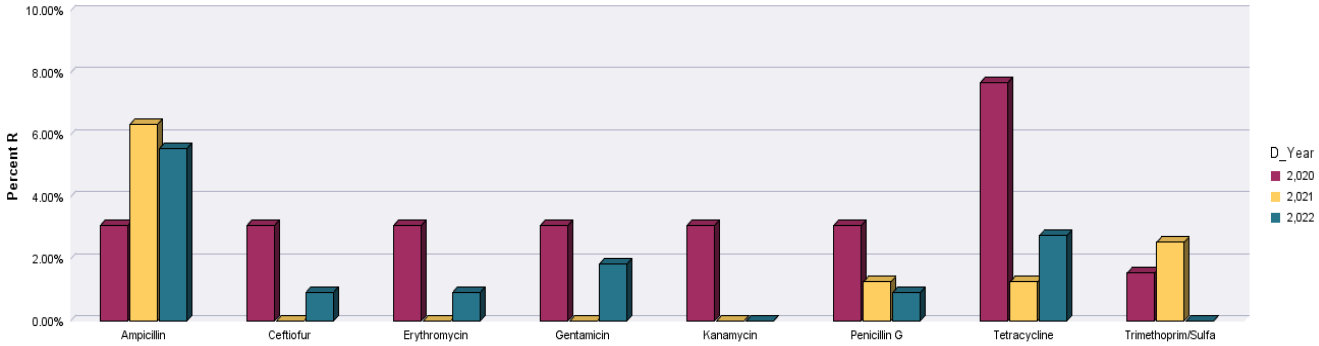

Figure 3. Antibiotic resistance profile of *S. aureus* isolated from broiler breeder chickens at the AHL from 2020 to 2022.

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Ventral opisthostoma tear with exsanguination in a Brazilian salmon pink bird-eating tarantula (Lasiodora parahybana)

Sara Pagliarani, Heindrich Snyman

Department of Pathobiology, University of Guelph, Guelph, ON (Pagliarani), Animal Health Laboratory, University of Guelph, Guelph, ON (Snyman)

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An approximately 9-year-old Brazilian salmon pink bird-eating tarantula (*Lasiodora parahybana*) was noted to be having a difficult molt with leakage of hemolymph from the ventrum. The animal was found moribund the following day, and was administered isofluorane and phenobarbital for euthanasia before being placed whole in formalin (**Figs, 1, 2**).

Histologically, there was a single ~ 5.0-6.0 mm wide full thickness defect within the integument along the ventral midline. The exo- and endo-cuticle along the remaining edges of the integumentary defect were partially separated from each other and elevated from the underlying stroma, with irregular fragmentation and tattering of the edges. The defect contained small amounts of fine granular to homogenous eosinophilic fluid with enmeshed fragmented yellow-brown plant substrate material (**Fig. 3**). Moderate numbers of admixed hemocytes were also present, with hemocytes also extending along the lateral margins along the sub-epidermal connective tissue (**Fig. 4**). No obvious bacteria were detected in the sections.

As was observed during clinical examination, this spider contained a regional integumentary defect along the ventral opisthosoma/abdomen. These defects are considered one of the most common causes of captive spider mortality. Gross lesions associated with this finding are usually those of epidermal discoloration, abrasion, and loss of surface structures to exoskeletal fractures. While the cephalothorax

and the limbs are considered solid and resilient structures, the opisthosoma, due to the absence of the more rigid chitinous exocuticle layer, is far more easily breached. Any damage to the endo-cuticle, epidermis, or exoskeleton can result in loss of osmoregulatory capacity, hemolymph loss and shock (exsanguination), in addition to opportunistic infections and in many cases, death of the spider.

Whereas major injuries are usually detected promptly, small injuries may not be, and exsanguination then often goes unnoticed as the cause of deterioration and/or death. While wounds over the dorsal opisthosoma are likely to cause more rapid hemolymph loss from the heart and surrounding pericardial sac, wounds at the base of appendages and the ventral opisthosoma seem to lose hemolymph slower and a spider may take several days to gradually exsanguinate, with little to no premonitory clinical signs.

Larger ruptures, such as in the current case, are easier to identify; however, quantification of fluid loss can still be difficult to estimate, especially considering that enclosure substrate will readily and rapidly absorb the relatively clear leaked fluid. The defect in the current case contained proteinaceous fluid and hemocytes which probably suggests that this rupture/tear occurred before death. Although foreign plant material (probably enclosure substrate) was embedded within the fluid, and there appeared to be some degree of hemocyte host inflammatory response, there was no obvious bacterial component. Therefore, the deterioration and death of this spider was probably the result of hydrodynamic/hydrostatic derangement rather than secondary bacteremia and sepsis. In most invertebrates, the first step towards wound healing is the formation of a hemolymph clot which rapidly prevents further hemolymph loss and entraps microbial pathogens before they can colonize other adjacent organs.

It is unclear whether or not the current case was the result of trauma during the actual shedding process or any other possible concurrent traumatic event (e.g. a fall). There were no obvious accompanying degenerative or inflammatory changes described in the histologic sections; therefore this finding is considered to be an incidental event. *AHL*

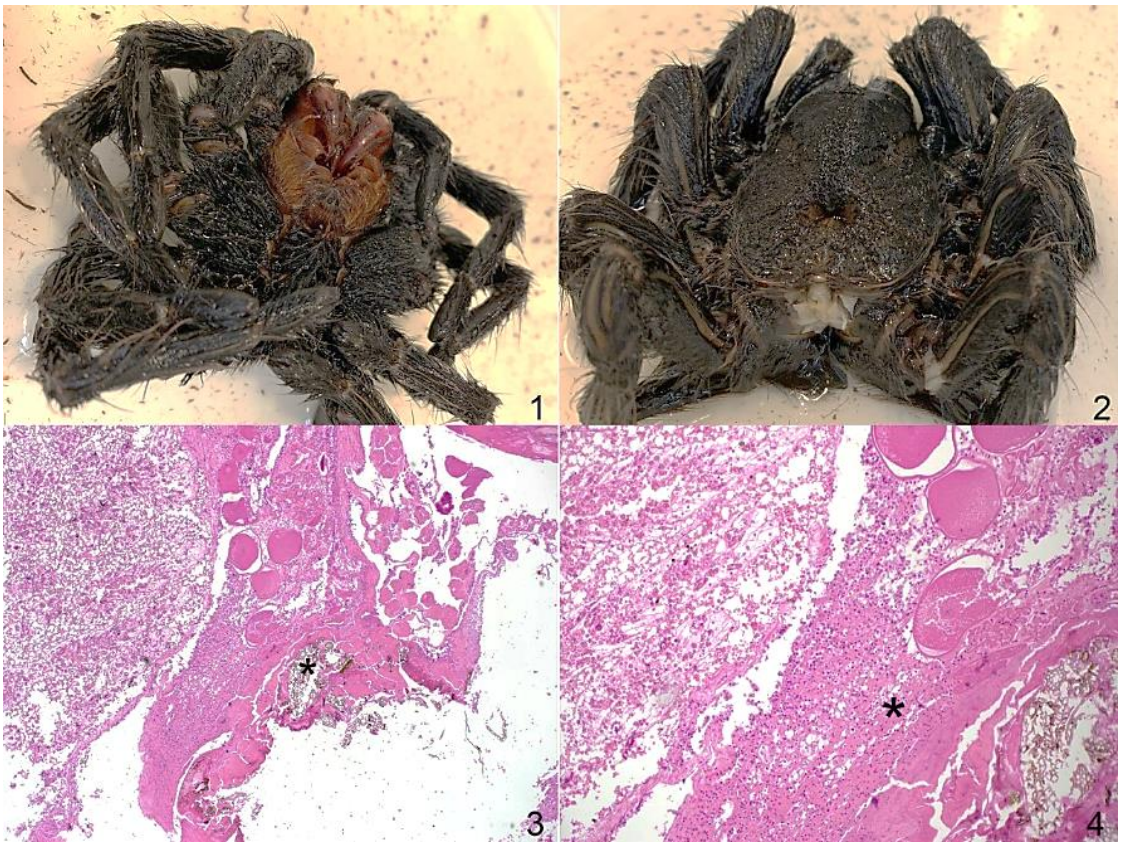

Figure 1. Full body, ventral view, Brazilian salmon pink bird-eating tarantula.
Figure 2. Full body, dorsal view, Brazilian salmon pink bird-eating tarantula.
Figure 3. Ventral opisthosoma, Brazilian salmon pink bird-eating tarantula. The opisthosoma contains a focal region of defect with eosinophilic material and embedded plant substrate fragments (*). H&E.
Figure 4. Ventral opisthosoma, Brazilian salmon pink bird-eating tarantula. Hemocytes are also present within the opisthosomal defect (*), and extend along the lateral margins of the sub-epidermal connective tissue. H&E.

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Equine pastern leukocytoclastic vasculitis

Josepha DeLay, Meegan Larsen, Teresa Reitz

Animal Health Laboratory, University of Guelph, Guelph, ON (DeLay, Larsen), Rideau St. Lawrence Veterinary Clinic, Prescott, ON (Reitz)

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Pastern leukocytoclastic vasculitis is an uncommon but distinct histologic entity in horses, and contributes to the clinical syndrome of pastern dermatitis in this species. Frequent involvement of non-pigmented skin, and occurrence of many cases in summer months has spurred speculation that vascular injury is aggravated by sunlight. However in some studies, pigmented skin may also be affected, and lesions may also develop in other seasons. The lesions may be painful, resulting in lameness.

Between January 1, 2010 and November 1, 2022, AHL pathologists have diagnosed pastern vasculitis in 10 horses, with 3 cases identified in 2022. All horses were >2 years of age, as is typical for this condition. Lesions may affect 1 or more limbs. The diagnosis relies on histologic identification of vasculitis or vasculopathy in biopsies from affected skin, in addition to the clinician's description of compatible skin lesions; these include lesion characteristics, distribution and targeting of non-pigmented skin.

A recent case diagnosed at the AHL involved a 16-year-old gelding with chronic recurrent hindlimb pastern dermatitis. Lesions primarily, but not exclusively, affected non-pigmented skin, and consisted of erosions and ulcers with serous exudate (**Fig. 1**). The lesions typically developed in summer and were non-responsive to various topical antibiotic, antibacterial, and anti-inflammatory therapies. In biopsies from affected skin, small blood vessels in the superficial dermis had thickened walls expanded by fibrin, small clumps of necrotic cellular debris, and rare neutrophils, consistent with leukocytoclastic vasculitis (**Fig. 2**). Fibrin thrombi filled lumens of a few blood vessels. Fibrin and free intact or fragmented erythrocytes surrounded some affected blood vessels, indicative of vessel compromise. Concurrent epidermal injury included hydropic degeneration of some basal epithelial cells, erosion and ulceration, reflecting ischemic injury resulting from vascular damage.

The cause of pastern vasculitis is unknown. The condition is thought to represent an immune complex disease and, as discussed above, may be exacerbated by exposure to sunlight. Secondary bacterial dermatitis is common due to epidermal injury and breach of the skin barrier.

As for many dermatologic conditions, knowledge of the anatomic distribution and appearance of clinical lesions is very important for correlation with histologic lesions and in reaching a working diagnosis. In cases of pastern leukocytoclastic vasculitis, communicating the lesion distribution to the veterinary pathologist is especially important, as this particular condition often involves distal limbs, and especially white-haired regions. A clear description of the primary skin lesions will aid in interpretation of histologic lesions, and in reaching a diagnosis. Biopsies should include acute lesions, if possible. Assessment of potential bacterial, fungal, or parasitic contribution to pastern dermatitis is also important in biopsy evaluation.

Therapeutic interventions are multifaceted and include management efforts to keep skin of the distal limbs clean and dry, preventing exposure to ultraviolet light by wrapping the distal limbs, and potential use of systemic or topical immunosuppressive and immunomodulatory medications. *AHL*

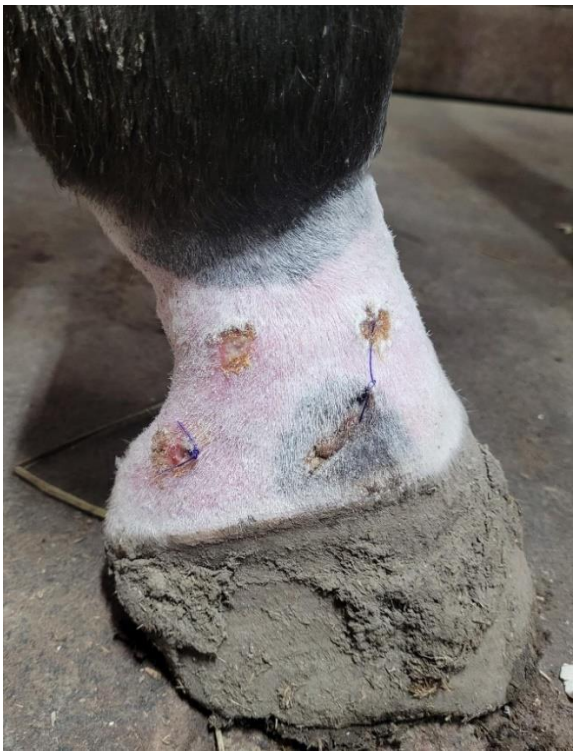

Figure 1. Multifocal erosive to ulcerative pastern dermatitis, mainly involving non-pigmented skin. Note that biopsies were taken at the margin of lesions to include both affected and surrounding unaffected skin, as indicated by suture placement.

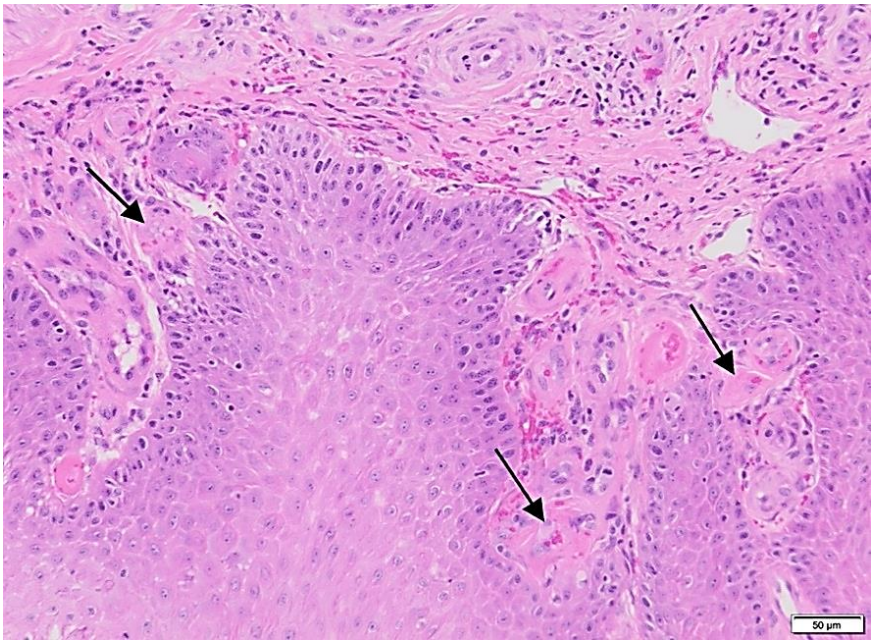

Figure 2. Superficial dermal blood vessels with mural thickening, perivascular fibrin and fragmented erythrocytes (black arrows). H&E.

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Echinococcus multilocularis: A fatal tapeworm infection in a dog

Amanda Mansz

Animal Health Laboratory, University of Guelph, Guelph, ON

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A 2-year-old male neutered Boxer dog presented to the referring veterinarian for abdominal distention and lethargy. Through various tests, including PCR, the dog was diagnosed with multiple hepatic/abdominal *Echinococcus multilocularis* parasitic cysts. After attempted treatments with alcohol ablation of the cysts without recovery and continued decline, humane euthanasia was elected.

At postmortem, there was compression of all abdominal organs by three large fluid-filled cysts (**Fig. 1A**). All cysts had a mottled white and red, nodular, heavily vascularized fibrous capsule. The cyst walls were thick and spongy with numerous fluid-filled pockets, and multiple foci of necrosis. The largest cyst measured 22.0 cm x 17.0 cm x 18.0 cm, and two of the three cysts nearly replaced the left medial and caudate process of the caudate lobes of the liver (**Fig. 1B**). The third cyst was located within the mesentery of the abdomen. There were many abdominal adhesions and generalized enlargement of all abdominal lymph nodes.

Histological sections of the cysts revealed massive regions of necrosis, and inflammation characterized by fragmented, eosinophilic hyaline membranes with numerous deeply basophilic calcareous corpuscles. Outlines of metacestode protoscoleces with few distinguishable refractile parasite rostellar hooks, fibrin, cellular debris, epithelioid macrophages, neutrophils and fibrosis were also present.

E. multilocularis is a zoonotic tapeworm found in North America and Europe, and is considered endemic in wildlife within defined regions of Canada. Wild canids (primarily foxes and coyotes) serve as the definitive host for the parasite. Adult tapeworms reside in the small intestine of these species, and the eggs passed in feces are immediately infective to intermediate hosts (mainly small rodents). Parasitic cysts filled with larvae grow within the abdominal organs of the intermediate rodent hosts. The rodent is then ingested by a wild canid, and the lifecycle begins again.

Domestic dogs are also considered to be a definitive host; however, deviations to the life cycle can occur, causing domestic canines to develop cysts (often hepatic) with intermediate parasite stages. *E. multilocularis* poses a potentially fatal zoonotic risk, as humans can become infected and develop parasitic cysts by ingesting eggs shed by an infected domestic dog. *AHL*

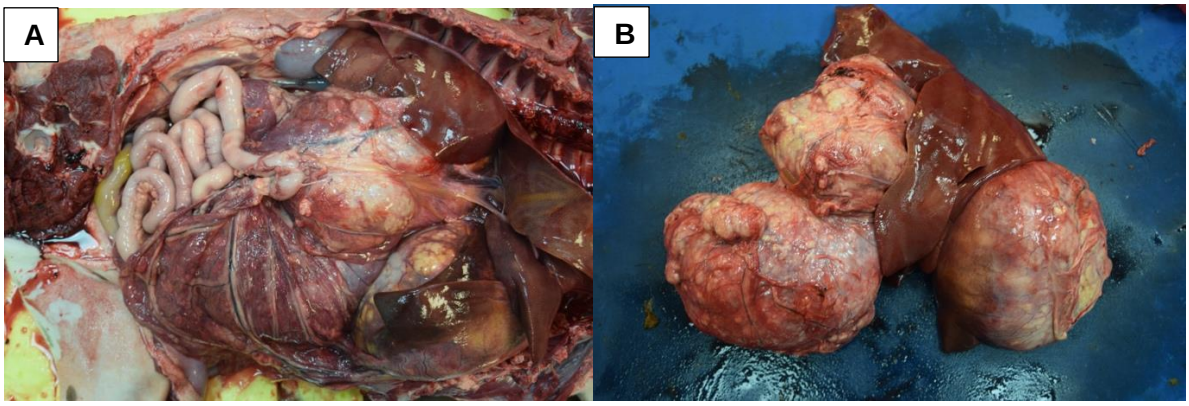

Figure 1. *E. multilocularis* cysts**. A.** Multiple hepatic and mesenteric cysts in situ within the abdominal cavity. **B.** Two large cysts replacing liver lobes and a third mesenteric cyst.

A case of Chagas disease in a puppy adopted from the southern United States

Rebecca Egan

Animal Health Laboratory, University of Guelph, Guelph, ON

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Shortly after arriving from Texas for adoption in Ontario, a six-month-old Dachshund mix puppy was presented to a veterinarian with a 1-day history of anorexia accompanied by lethargy, soft stools, and straining to defecate. Clinical signs did not resolve with symptomatic treatment, and the puppy went on to develop hypothermia, hypoglycemia and poor oxygenation. Stabilization was attempted, but the puppy died due to cardiopulmonary arrest. The puppy was presented to the AHL for postmortem examination which identified 150 mL of thoracic effusion, a lesser amount of pericardial effusion, and extensive pale mottling of the heart that was visible from the epicardial surface. Auricles appeared symmetrical and of normal size. On cut section, the right ventricular free wall was thickened to 4 mm and the LV:RV thickness ratio was 2:1 (**Fig. 1A**). The myocardium was pale, and there were no grossly visible structural defects. Other notable findings included pulmonary congestion and edema, 150 mL of serous abdominal effusion with edema expanding intra-abdominal adipose and connective tissues, and accumulation of ample hemorrhagic contents throughout the jejunum. Microscopic examination revealed severe pancarditis characterized by widespread infiltration of neutrophils, eosinophils and mononuclear cells occupying greater than 50% of the tissue sections (**Fig. 1B**). Multifocally, individual myofibers contained variably sized, intracytoplasmic oval to elongate pseudocysts measuring approximately 150 um x 50 um that harboured numerous 2-4 um round to oval protozoal amastigotes (**Fig. 1C**). The inflammatory infiltrate separated and sometimes obscured cardiomyocytes that exhibited occasional necrosis. A heart sample was submitted for PCR testing which confirmed the presence of *Trypanosoma cruzi* organisms.

Trypanosoma cruzi is a zoonotic pathogen that can infect many mammals, and is known for causing Chagas disease (also known as American trypanosomiasis) in dogs and humans. The parasite is endemic in parts of South America, Mexico, Central America, with presence also documented in the southern United States, and is transmitted by a biting fly vector or though ingestion of infected bugs or

contaminated feces. Chagas disease can manifest with acute and/or chronic phases, and the incubation period can range from approximately 5 to 42 days. In addition to non-specific febrile illness, acutely affected dogs can present with anorexia, lethargy, vomiting, diarrhea, lymphadenopathy, hepatomegaly, and/or splenomegaly and reports of acute clinical myocarditis seem to be less common. This case highlights the importance of obtaining travel history as part of a clinical work-up, since animals travelling between different geographical locations can carry pathogens not native to the area in which they reside.
AHL

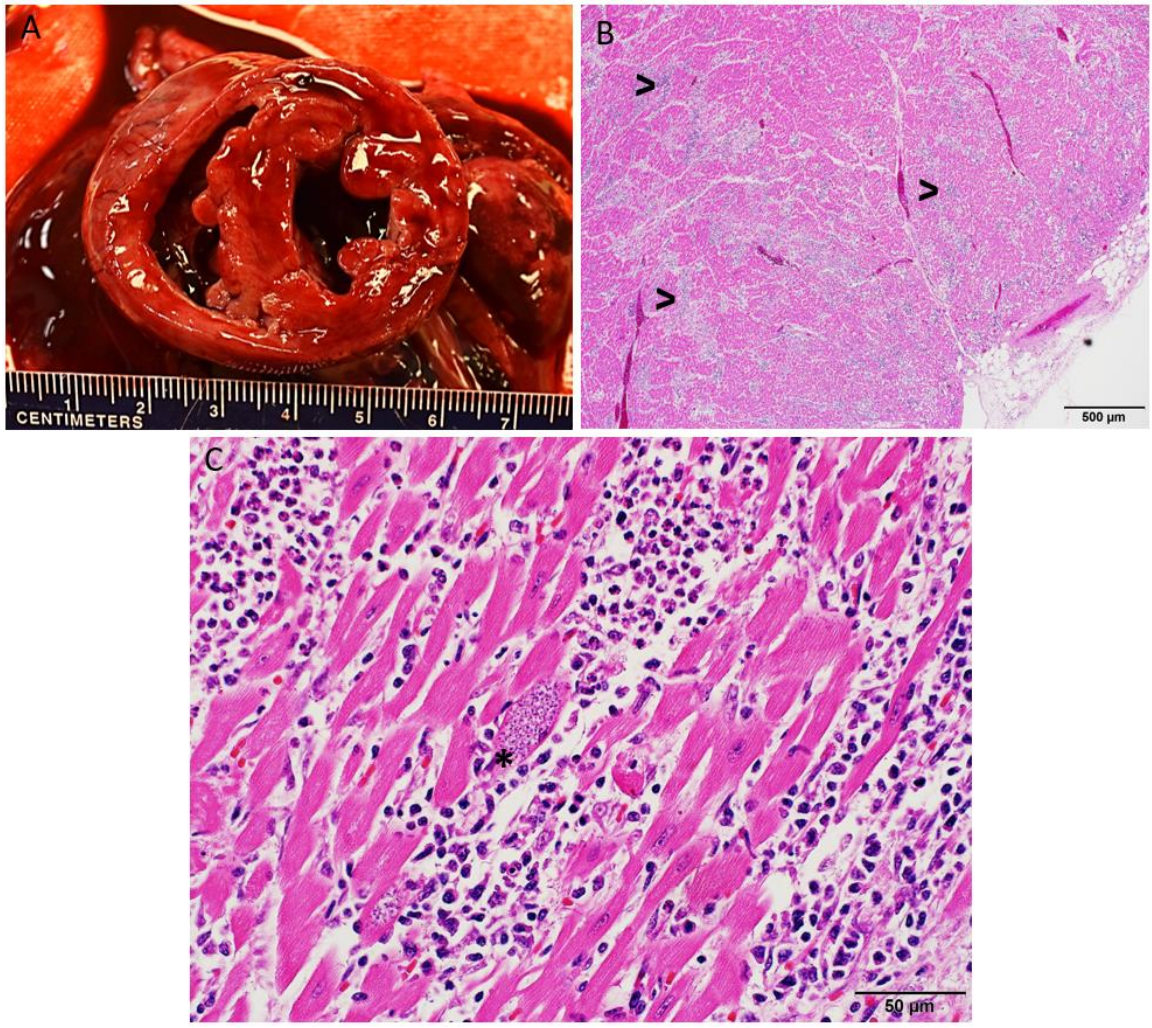

Figure 1. Gross and histologic lesions of *T. cruzi* in a puppy. **A.** Cross-section of the heart showing thickening of the right ventricular free wall and pallor of the myocardium. **B.** Microscopic section of heart demonstrating widespread inflammatory infiltrates throughout the wall (>) (H&E, 4x). **C.** Microscopic section of heart capturing predominately neutrophilic inflammation surrounding a cardiomyocyte harbouring an intracytoplasmic pseudocyst containing numerous 2-4 um protozoal amastigotes (*) (H&E, 40x).

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