



# **C. E. KORD ANIMAL HEALTH DIAGNOSTIC LABORATORY**

## **LABORATORY USE GUIDE**

[Revised April 2019]

**Tennessee Department of Agriculture**

**Division of Consumer and Industry Services and Animal Health**

# C. E. KORD ANIMAL HEALTH DIAGNOSTIC LABORATORY

TENNESSEE DEPT OF AGRICULTURE, DIVISION OF CONSUMER AND INDUSTRY SERVICES AND ANIMAL HEALTH

<https://www.tn.gov/agriculture/consumers/pets/animal-health-diagnostic-lab0.html>

## U.S. POSTAL ADDRESS

**Kord Animal Health Diagnostic Laboratory**  
P.O. Box 40627  
Nashville, TN 37204-0627

## OVERNIGHT DELIVERY ADDRESS

**Kord Animal Health Diagnostic Laboratory**  
Porter Building, Ellington Agricultural Ctr.  
436 Hogan Road  
Nashville, TN 37220

## TELEPHONE NUMBERS

Information . . . . .	(615) 837-5125	Poultry Testing . . . . .	(615) 837-5128
Animal Health Services . . . . .	(615) 837-5120	Bacteriology . . . . .	(615) 837-5427
Agriculture Dept. . . . .	(615) 837-5103	EAC Security . . . . .	(615) 642-1972
Brucellosis Results . . . . .	(615) 837-5128	Billing . . . . .	(615) 837-5410
Tissue Receiving . . . . .	(615) 837-5410	Serum Boxes . . . . .	(615) 837-5231
Immunology . . . . .	(615) 837-5221	Fax Number . . . . .	(615) 837-5250

## INTRODUCTION

The C. E. Kord Animal Health Diagnostic Laboratory (KAHDL), a part of the Tennessee Department of Agriculture, Division of Consumer and Industry Services and Animal Health, provides no-cost diagnostic services for food animal producers and low-fee diagnostic services for companion animals.

The mission of the KAHDL is to provide accurate and timely diagnostic services to Tennessee veterinarians, animal owners, and commercial animal operations. Most submissions are made by veterinarians or with their knowledge and approval, which allows the laboratory to communicate test results rapidly and effectively to the veterinarian, who will ultimately determine factors such as treatment or prevention. Submissions are also accepted directly from animal owners, although use of a veterinarian is preferred. Laboratory personnel do not provide advice to animal owners regarding treatment, vaccination, or other medical or management factors; this should be provided by your veterinarian.

The KAHDL provides necropsy, biopsy, cytology, bacteriology, mycology, serology, immunology, parasitology, and molecular (PCR) diagnostic services for a wide range of animal diseases in many species of animals. The laboratory uses subcontractors for certain tests, such as toxicology. We also work with local private crematory services if an animal owner desires that service for their pet.

The KAHDL is fully accredited by the American Association of Veterinary Laboratory Diagnosticians (AAVLD) and is a member laboratory of the National Animal Health Laboratory Network (NAHLN), the Veterinary Laboratory Investigation and Response Network (Vet-LIRN), and the National Poultry Improvement Plan (NPIP).

If you have a question regarding a test, samples required, fees, or any other aspect of the laboratory, please feel free to call. Our staff of well-trained professionals and technicians are here to provide the highest possible quality of service and diagnostic results.

Bruce G. McLaughlin, DVM, MVSc  
Laboratory Director

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## LABORATORY HOURS

- 8:00 AM to 4:30 PM, Central Standard Time, Monday through Friday
- Observed Holidays:
  - New Years
  - Dr. Martin Luther King Jr. Day
  - Presidents' Day
  - Good Friday
  - Memorial Day
  - Independence Day
  - Labor Day
  - Veteran's Day
  - Thanksgiving
  - Christmas

## FEES

- Fees for the Kord Laboratory are determined by state statutes TCA 4-3-203, TCA 43-1-703, and TCA 44-7-403
- Horses, poultry, and livestock (animals used for food or fiber): **No charge for Tennessee residents for diagnostic tests**
- For all species, out-of-state submissions are charged at companion animal fee rates and charged per sample, not per submission
- Equine Infectious Anemia (Coggins): \$7/test
- Disposal fee for all species: \$25 per 250 lb. Animals are not accepted for disposal only.

## COMPANION ANIMAL FEES

### PATHOLOGY

- Histopathology/biopsy \$38.00 for first specimen, \$22.00 for each additional specimen
- Necropsy & disposal \$105.00
  - Toxicology add \$35.00
  - Spinal Cord add \$20.00
  - Save remains add \$25.00 (remains released only to licensed crematorium)
- Necropsy limb, organ, or head \$80.00
- Cytology \$30.00 per problem site
- Slide preparation for cytology \$15.00 per specimen
- Decalcification \$10.00 per tissue
- Special stains \$5.00 per stained slide
- Slide preparation/HE \$8.00 per slide
- Slide preparation/IHC \$10.00 per block (5 slides)
- Submitter special stain \$10.00 per slide (requested by submitter)

### TOXICOLOGY

- First test \$35.00
- Each additional test \$18.00

### SEROLOGY/IMMUNOLOGY/MOLECULAR

- FA, IFA, AGID, SN, etc \$25.00 per test type per submission, not per sample
- PCR testing \$50.00 per test type per sample

### BACTERIOLOGY/PARASITOLOGY

- Aerobic culture \$25.00 per swab, tissue, site, or sample
- Antimicrobial susceptibility \$25.00 per isolate
- Fungal culture \$25.00
- Anaerobic culture \$25.00
- Milk culture & susceptibility \$25.00 per submission and not per sample
- Stain (gram, acid fast) \$5.00
- Ectoparasite identification \$25.00

### MISCELLANEOUS CHARGES

- Leaky package/biohazard handling \$25.00
- Shipping and handling \$25.00

## GUIDELINES FOR SAMPLE SUBMISSION

- All submissions must be accompanied by an appropriate submission form that must be filled out completely.
- Reporting results:
  - An E-mail, telephone, or fax report will be given on any significant laboratory findings as they become available or if the result requires explanation by a veterinary diagnostician.
  - A written report will be sent to the referring veterinarian upon completion of the case. The preferred distribution method is by E-mail.
- A copy of the written report is sent to the owner if the owner delivered the tissues or animal directly to the laboratory or the submission is from a farm animal species and the owner's complete address is present on the submission form. E-mail delivery of the report is preferred; use of the USPS will significantly delay delivery.
- Please report any changes of E-mail, address, phone number, or clinic association to the laboratory at (615) 837-5125.
- Veterinarians utilizing laboratory services may choose to send payment with submitted specimens or establish a charge account with the laboratory. Accumulated charges will be billed to the clinic on a monthly basis. Accounts 90 days overdue are subject to a withholding of fee-based services until payment is received.
- Submission of samples, specimens, or animals to the KAHDL implies agreement with the policies of the State of Tennessee and the KAHDL and transfers ownership of samples and property to the KAHDL, unless otherwise determined by written agreement prior to submission. Remains are released only to a licensed crematory service. The Tennessee Department of Agriculture has mandated that the Technical Services Laboratory is a subcontractor for the KAHDL.
- Safety of laboratory personnel is paramount. Submissions with attached needles or leaking formalin or other fluids are subject to being discarded or may incur a biohazard fee. For primates, only formalin-fixed samples will be accepted (no fresh tissue or fluid samples from primates will be accepted).
- Animals that are submitted with suspicion of a gunshot wound must be accompanied by radiographs (X-rays) of the body part in question in two orthogonal views if retrieval of a projectile or confirmation is desired.
- Bone lesions or malformations are best diagnosed on radiographs. The laboratory does not have radiographic capabilities and has limited capacity to examine bone lesions.
- Formalin-fixed tissues are required for histopathology examination. Fresh tissue will not be accepted for histopathology.
- For rabies testing, the head or brain of the animal should be submitted to the Tennessee Department of Health rabies lab. Companion animal specimens sent to KAHDL will incur a necropsy fee to offset the costs of sample procurement, processing, and shipping to the Department of Health.

## SAMPLE SUBMISSION BEST PRACTICES

**Serum** - Collect blood aseptically into a sterile dry tube, refrigerate immediately, allow to clot, centrifuge, and transfer serum aseptically into a second tube. Serum must be fresh, clear, non-hemolyzed, and uncontaminated. Use of red top Vacutainers®, B-D, or other non-EDTA/heparin sterile commercially available tubes is preferred. Do not let whole blood freeze or be exposed to direct sunlight or high temperature before decanting serum.

- Label each tube (not stopper) with **tube number** and **vet code**. Ensure that labels are legible (use an indelible marker). Avoid using animal names for sample labels. Keep your own master list of sample numbers to avoid duplication and confusion. Number samples consecutively, and include a master list with the submitted samples; keep a copy of the master list. Put tubes in a box or tape them to cardboard so they will not be lost in the packing materials.
- Call the lab if sending more than 20 samples to permit preparation for high-volume testing.
- Submit at least 1.0 mL of serum for each test requested. Refrigerate the serum until shipment.

**Fresh tissues** should be collected aseptically, placed individually in well-labeled Whirl-Pak® bags, and refrigerated immediately. Refrigerated samples should be shipped with sufficient cold packs and padding. Ship samples in a leak-proof insulated container or a sturdy mailing carton that complies with postal or commercial carrier specifications. Submit samples early in the week to avoid holding over the weekend by the carrier. **FedEx®** or **UPS®** are preferred. Shipping by the USPS may result in significant delays.

**Cytology** slides should be made as soon as possible after collection; review at least one slide before submission to ensure that specimens are of good quality. Slides should be sent in a protected plastic or cardboard slide mailer to avoid breakage. There is a slide preparation fee for fluids sent to the lab and these are seldom diagnostic because of the deterioration of cell features in fluids over time.

**Complete and legible form(s)** (including a complete history) must accompany all submissions. All regulatory charts must include the submitting veterinarian's signature, vet code, and animal identification.

KAHDL is proficiency tested and certified by the National Veterinary Services Laboratory (NVSL) or the USDA National Animal Health Laboratory Network to perform:

- Anaplasmosis enzyme-linked immunosorbent assay (ELISA)
- Agarose gel immunodiffusion (AGID) for bluetongue, bovine leukemia virus, and equine infectious anemia
- Avian influenza ELISA and AGID
- *Mycobacterium avium* subsp *paratuberculosis* (Johne's disease) ELISA
- Johne's disease, avian influenza, foot and mouth disease, classical swine fever, African swine fever, and exotic Newcastle disease by polymerase chain reaction (PCR)
- Pseudorabies serologic screening by gB ELISA

#### Ear notch specimens for BVD:

- Materials and equipment needed:
  - Ear notching tool that yields a 1 cm x 1 cm notch. We recommend purchasing two or more tools
    - Nasco: 1-800-558-9595. Order ID medium C0024N
    - Dairy Health USA: 1-800-276-7933. Order ID medium 440-31
    - Stone Livestock ID & Show Equipment: 1-816-231-4020. Order ID medium 7125
  - Red top blood tube: Vacutainer® 7 mL draw, 16 mm x 75 mm collection tube with no additives.
  - Shipping container that holds individual tubes in slots.
  - Kord Animal Health Diagnostic Laboratory submission form.
  - Disinfectant for rinsing notching tool: 10% bleach (100 mL [3 oz] bleach in 900 mL [27 oz] water).
  - Clean rinse water: 3-5 gallon bucket. Change bucket water every 20-30 notches.
- Collection and submission procedure
  - Label red top collection tubes with consecutive numbers beginning with #1 and keep a list of the corresponding animal ID or tattoo numbers. This method of labeling significantly reduces the turnaround time in the laboratory.
  - Dip the notching tool in disinfectant, and then rinse away disinfectant with copious quantities of clean water. CAUTION: Residual disinfectant on the notching tool will yield false negative results, so thorough rinsing with clean water is required.
  - Collect an ear notch that measures approximately 1 cm x 1 cm (3/8 in. x 3/8 in.) from a clean portion of the ear. Only fresh ear notch samples are acceptable for the antigen-capture ELISA test. **The ear should not be cleaned with anything but water. Disinfectant of any kind (e.g. Novasan) can interfere with the test.**
  - Place the fresh ear notch into a sterile, clean red top Vacutainer® collection tube. Do not use a Whirl-Pac® bag or any other type of plastic bag. CAUTION: The ear notch must be free of contaminating dirt, feces, tattoo ink or BVD vaccine. Do not vaccinate or tattoo at the same time samples are taken.
  - Send samples to the laboratory with sufficient cold packs to chill the specimen during shipment. Notify the laboratory prior to shipment regarding the number of samples and when they will be submitted. This allows us to have adequate supplies for testing and to arrange staffing to do the testing.

**Equine Infectious Anemia (EIA or Coggins)** serology requires at least two (2) mL of clear, fresh, non-hemolyzed serum and takes at least 24 hours to complete when the laboratory is fully staffed and sample numbers are not excessive. Each tube of serum should be identified with the **vet code** and a **tube number** that corresponds to the tube number on the submission form. If additional tests other than EIA are requested, please submit two samples. The submission form (VS form 10-11) must be completely and correctly completed and signed by the submitting veterinarian.

#### Please note the following:

- KAHDL cannot make any changes to the form or fill in any missing information.
- Forms that are not properly completed will cause delay in testing.
- The AGID test used for EIA testing requires substantial preparation time and a 24-hour incubation period. Next-day results cannot be guaranteed. Results will be available within 4 business days.
- Submit serum tubes in slotted blood boxes. These are available at no cost from the laboratory (615-837-5231). Leave indicated information on the recording.

**Export testing:** The submitting veterinarian is responsible for informing the laboratory of any special requirements (e.g., specific dilutions or type of test). If this information is not supplied, there may be a delay in receiving test results. For information on test regulations, call:

- Federal – USDA, APHIS, VS, Veterinarian in charge (615.781.5310)
- State – Office of the State Veterinarian (615.837.5120)

**When calling for results,** please have the following information exactly as provided on the submission form:

- Submitting veterinarian's name
- Owner's name
- Animal's name or ID
- Sample number or ID
- Submission date
- Bleeding date

**Advance notice** (7-10 days) should be provided when submitting a large number of samples.

**Serodiagnosis:** In an individual animal, paired sera are recommended to test for specific antibody: the first sample taken when the animal is initially examined (acute-phase serum) and the second sample 2-4 weeks later (convalescent-phase serum). An increase in antibody titer between the paired samples is a basis for a serologic diagnosis of a particular disease. The paired serum samples should be submitted together in order to obtain a better understanding of the diagnostic significance of titers as they relate to the clinic and/or vaccination status of the animal.

**If only a single serum sample** can be obtained, it should be taken from a convalescent animal. An acutely ill animal is typically devoid of antibody against the particular disease-causing agent.

**If only acutely ill animals** are present, swabs or tissues for identification or culture of the causative agent should be submitted.

## **SAMPLE SUBMISSION FAILURES**

Examples of problems in submission of serum and tissue specimens are:

- Improper sample or incomplete submission form
- Serum not separated from blood clot
- Chemical (e.g. detergent and disinfectants) contamination of ear notches, which adversely affects BVD testing
- Hemolysis
- Insufficient sample quantity
- Overheating or freezing of blood before serum separation
- Leaky stopper or container
- Broken or improper container
- Decomposed tissue specimens
- Frozen fresh tissues

## USPS MAILING/PACKAGING REQUIREMENTS FOR BIOLOGICAL LAB SPECIMENS

Sending items by the United States Postal Service may result in significant delays – in some cases, up to a week’s increase in turn-around time can occur. All biological specimens sent via the USPS must be properly packaged so the contents do not leak during shipment. Reference: Domestic Mail Manual, CO23.10.0-10.7.

- The specimen must be packaged in a securely sealed watertight primary container (test tube, vial, etc) which is then placed in a watertight, secondary plastic container with a tightly sealed closure that cannot open during shipment.
- **CAUTION: Do not use sealable food bags except for submission forms**
- The specimen container and the secondary container must then be placed in a sturdy cardboard box protected with additional absorbent packing material. The submission form should be submitted in a leak-proof sealed bag.
- Etiologic agents and biohazard materials must be sent by first-class Mail, Priority Mail, Express Mail, UPS® or FED-EX.

### SUBCONTRACTORS

NOTE: Any test not listed in this guide is subject to subcontracting. Subcontracting of tests may also occur if a test or procedure normally conducted at the KAHDL is not currently available. Submission of samples to the KAHDL implies authorization to use a subcontractor for testing. Fee-based testing requires client preapproval. Certain tests (toxicology testing, somatic cell counts on milk, plant identification, and other tests indicated by the Tennessee Department of Agriculture) are mandated by the Tennessee Department of Agriculture to be subcontracted to the Technical Services Laboratory of the Division of Consumer and Industry Services and Animal Health. Client reports will include the identity of subcontractor if used.

Subcontractor	Address	Phone	Tests	Certification
NVSL	USDA, APHIS, 1800 Dayton Ave, Ames, IA 50010-9674	515-663-7212	Non-KAHDL diagnostic testing	ISO 17025
Plum Island Animal Disease Center	P.O. Box 848 Greeneport, NY 11944	631-323-3200	Foreign Animal Disease confirmation	ISO 17025
Tennessee Dept of Health	630 Hart Lane, Nashville, TN	615-262-6351	Rabies, <i>Salmonella</i> typing, bacterial ID	External proficiency testing by Wisconsin Dept. of Hygiene
Technical Services Lab, Tennessee Dept of Agriculture	Ellington Agricultural Ctr, 436 Hogan Rd Nashville, TN 37220	Toxicology: 615-837-5414 Somatic cell counts: 615-837-5267	Toxicology tests, milk somatic cell counts, plant identification	FDA Laboratory Proficiency Evaluation Team (somatic cell counts)

### ABORTION

Diagnosis of the cause of abortion is often difficult and complex. Tips to increase your diagnostic yield include:

- Submit tissues from multiple fetuses and placentas if available.
- For serum titers, collect and submit the first of paired serum samples from the aborting animal and follow in 2-3 weeks with the second sample.
- If a toxic condition is suspected, submit samples of the aborting animal’s feed and water along with aqueous humor or an intact eye.
- If the entire fetus and placenta cannot be submitted, submit the following:
  - Fresh tissue for bacterial culture or other tests may include stomach contents, placenta, liver, lung, spleen, kidney, and brain.
  - Fixed tissue for histopathological examination should include placenta, lung, liver, heart, adrenal, kidney, thymus, and brain.
  - Ocular fluid (best) or eye for nitrates.

## BACTERIOLOGY SUBMISSIONS

KAHDL does not conduct any testing for food safety purposes or make any recommendations regarding safety of items intended for human consumption.

- **AEROBIC SPECIMENS FROM NECROPSIED ANIMALS**

- Collect all specimens as aseptically as possible. A golf ball-sized portion of each organ should be collected.
- If the outside of the specimen is accidentally contaminated, wash the specimen with clean tap water.
- Refrigerate specimens as soon as possible.
- For neonatal diarrhea, submit tied off 4-5 cm segments of jejunum, ileum, and colon with the accompanying lymph nodes for culture. Fecal parasite identification is not performed at KAHDL.
- Tissue specimens should be placed in individual leak-proof plastic bags and identified (use water-proof ink). Ideally each tissue should be separate - at a minimum, separate gastrointestinal tract from other organs.

- **MASTITIS MILK SPECIMENS**

- Wash udder to remove dirt and allow to dry.
- Scrub teat end with alcohol soaked cotton and allow to dry.
- Collect milk (4 mL) in a sterile container immediately prior to regular milking without discarding any streams of milk and submit immediately. Use a master list for sample numbers and animal IDs and send a copy with the samples.

- **SWAB SUBMISSIONS** - Collect samples aseptically and submit in commercial transport media that is not expired.

- **ANAEROBIC AND MICROAEROPHILIC SPECIMENS**

- Note: Anaerobic and microaerophilic organism culture is heavily dependent on sample selection and proper shipment.
- Samples should be taken from a living animal or a fresh carcass. Intestinal loops should be tied off.
- Specimens should be submitted in a transport medium that limits or excludes air from the sample. Use a commercial anaerobic transport media swab.

## MYCOLOGY (FUNGAL CULTURE) SUBMISSIONS

- **COLLECTION AND CARE OF SPECIMENS** - Only dermatophytes are cultured; systemic zoonotic fungi such as *Blastomyces* will not be cultured. Submit skin scrapings from the outer edges of a lesion and plucked (not cut) hairs. Skin, hair, and nails should be shipped to the laboratory without refrigeration.
- **RESULTS** – Isolation of fungi normally takes longer than isolation of bacteria and may require 14-21 days.
- **DIMORPHIC FUNGI** – *Blastomyces* and *Histoplasma* are among the highly pathogenic dimorphic fungi that pose a significant risk to laboratory personnel; the KAHDL does not culture these fungi. These diseases are best diagnosed by serological methods, cytology, or histopathology, which provide a more rapid diagnosis than fungal culture.

## MILK SOMATIC CELL COUNTS

Somatic cell counts on ruminant milk samples are performed in a pilot program by a subcontractor, the Food and Dairy Microbiology Section of the Technical Services Laboratory, Division of Consumer and Industry Services, Tennessee Department of Agriculture, on Mondays and Fridays. This subcontractor is mandated by the Tennessee Department of Agriculture. Samples are submitted through the Kord Laboratory. Bulk milk tank samples and samples from individual animals are acceptable. For samples from individual animals, follow the collection guidelines for mastitis milk samples in the Bacteriology Submissions section of this laboratory guide. Samples should be kept cold but not frozen. If submitting > 6 samples, please contact the lab at least 3 days in advance.

- Each submission may include a maximum of 20 samples; use a master list for sample numbers and animal IDs and send a copy with the samples
- Each producer may submit routine samples once every 3 months
- Problem herds may be tested more frequently after consultation with the Kord Laboratory
- Samples with somatic cell counts > 400,000 cells/mL may be cultured bacteriologically in the Kord Laboratory if desired; a maximum of 10 samples/submission will be cultured; antimicrobial susceptibility testing will be performed if known mammary gland pathogens are isolated

## TURNAROUND TIME

KAHDL staff make every effort to provide prompt and accurate results. Clients will be notified if a significant delay in reporting of results is expected. Some procedures may take several weeks or more to complete; if there is a question regarding results, feel free to call for an estimated finalization date.

## COMMON MICROBIOLOGY TESTS

The Microbiology Section of the KAHDL provides culture and susceptibility testing for a wide variety of infectious organisms. Some of the most common are listed below along with the preferred sample.

TEST	SAMPLE
Acid fast stain	Feces or intestine (for <i>Cryptosporidia</i> )
Aerobic culture	Fresh chilled tissue, urine, exudate, transtracheal wash
Anaerobic culture	Fresh tissue, anaerobic culturettes, exudate
Antimicrobial susceptibility	Performed on isolates recovered from specimens
Calf scours	Feces or affected intestine
<i>Candida</i>	Lesion, milk
Dermatomycosis	Lesion, hair, scales, fungal slants or trays
Dermatophilosis	Hair and scabs (please submit a generous sample)
Diarrhea/enteritis	Feces, affected intestine
Lumpy jaw ( <i>Actinomyces</i> )	Exudate, lesion, sulfur granules
Mastitis (milk) culture	Milk (2-4 mL) submitted in a sterile tube. Please notify the lab in advance if submitting more than 40 samples. <i>Staphylococcus</i> -only culture is available if desired – other organisms will not be identified.
<i>Mycoplasma</i>	Fresh chilled tissue, transtracheal wash, swab. Submission in Universal Viral Transport Medium (Fisher 22-349-981 [BD 220220]) is recommended. May require 10-14 days for culture. Submit duplicate swab if other tests are required.
Pinkeye ( <i>Moraxella bovis</i> )	Culturette of affected eye. Do not submit in Universal Viral Transport Medium.
Pneumonia	Lung (indicate if <i>Mycoplasma</i> , <i>Hemophilus</i> , or <i>Rhodococcus</i> is suspected)
<i>Salmonella</i>	Feces
<i>Strep. equi</i> (strangles)	Exudate from non-draining lesion

## TRICHOMONAS – SAMPLE COLLECTION & SUBMISSION

Note that the preferred method for diagnosing trichomoniasis in cattle is now PCR assay; the KAHDL no longer performs routine *Trichomonas* culture. All samples must be submitted in an InPouch™ TF pouch, which may be purchased from Biomed Diagnostisics.<sup>1</sup> Accurate diagnosis is dependent on sample collection, handling, and processing.

- **MATERIALS REQUIRED:**
  - InPouch™ TF pouch
  - Disposable gloves
  - Infusion pipette
  - 20 mL syringe
  - Wooden applicator stick or sterile cotton-tipped swab
- **SAMPLES:**
  - Smegma or preputial flush of bulls
  - Uterine or vaginal fluid from cows
  - **SAMPLE COLLECTION:** To view an online training course, Trichomoniasis Testing Course for Bovine Practitioners, go to: <http://extension.wsu.edu/vetextension/Beef/trich/Pages/default.aspx>. Contact Dr. Sara Clariday, Assistant State Veterinarian, at 615.837.5120 if you have questions about certificates of completion for this training course.
- **INOCULATION OF InPouch™:** For details, see the InPouch™ TF manufacturer's instructions.
- **SUBMISSION and SHIPPING REQUIREMENTS:**
  - Samples must be collected into InPouch™ TF pouches (see vendor information below)
  - Pouches must be kept at ambient (room) temperature (65-80°F or 18-27°C) prior to shipping to the lab
  - Avoid overheating or cooling the samples
  - Do not use expired InPouch™ TF pouches (dated on side of each pouch). Such samples will be rejected.
  - Do not submit samples in other containers or media.
  - Do not refrigerate or freeze the inoculated InPouch™ sample, or an un-used InPouch™.
  - Send the inoculated InPouch™ samples in an **INSULATED CONTAINER with NO ICE PACKS** by overnight express or 1-day delivery (not USPS). The Kord Laboratory should receive the samples within 24-48 hours after collection.
  - Samples in transit for more than forty-eight (48) hours after collection will NOT be accepted for testing.
  - When planning to collect ten (10) or more samples, please schedule the submission with the laboratory prior to collecting the samples.
  - Number the pouches with consecutive numbers (1, 2, 3, etc.). Keep a master list of the numbers and the corresponding animal ID or tattoo numbers and send a copy to the lab along with the regular lab submission form.
  - Schedule shipments to avoid weekend or holiday delivery to the laboratory. Samples are accepted on Friday if they are delivered in person by 4:00 P.M.

- **TURNAROUND TIME:**
  - PCR: Two (2) to five (5) business days after samples are received.

<sup>1</sup>**BioMed Diagnostics, Inc.**, 1388 Antelope Road, PO Box 2366, White City, Oregon 97503 - (800) 964-6466

- [www.biomeddiagnostics.com](http://www.biomeddiagnostics.com)
- InPouch™TF Test – Bovine                      Cat # 11-1003      100 tests
- InPouch™TF Test – Bovine                      Cat # 11-1010      10 tests

## **NECROPSY SUBMISSIONS & CARE OF REMAINS**

- Livestock that do not pose a threat to laboratory personnel will be euthanized for necropsy (not for disposal); fractious or dangerous livestock, such as ambulatory cattle loose in a trailer, must be euthanized by a private veterinarian before submission
- Cool dead animals as soon as possible after death.
  - Large animals should be thoroughly hosed down with cold water.
    - Birds, rabbits, and fur-bearing animals: soak in cold, soapy water; place in a plastic bag; refrigerate
  - **NOTE:** Do not place animals in a plastic bag without prior cooling.
- A cursory confirmatory examination or refusal of an animal for necropsy will be at the discretion of the diagnostician assigned to the case and will be based on the following:
  - An animal is deemed too decomposed for further diagnostic testing
  - An animal has signs that are consistent with a recent laboratory diagnosis in the same group or herd
  - An animal has already had a diagnosis confirmed by veterinarian or owner (e.g., fractured limb, uterine prolapse, or chronic laminitis)
- In all cases, the KAHDL diagnostician will determine the suitability of submitted animals, tissues, or other materials for testing and determine which tests are performed.
- When multiple animals are submitted representing a herd or flock problem, the KAHDL diagnostician will determine the number of animals to be necropsied.
- Legal and cruelty cases - Animal cruelty and legal cases will be handled in the same manner as a routine necropsy submission.
  - Additional veterinary forensic testing such as determining the time of death, forensic entomology, or determining types of accelerants used in burn cases will not be performed.
  - Please notify the laboratory at the time of submission that the case may involve cruelty or a legal issue.
- Care of remains and cremation services – KAHDL does not perform private cremations. All remains are incinerated without return of individual animal ashes. If the owner wishes to use a private crematory service, remains are saved and packaged (fee, \$50.00) for transfer to one of the local cremation services.

## **HISTOPATHOLOGY - COLLECTION AND PRESERVATION OF SPECIMENS**

- Diagnostic accuracy is directly proportional to the collector's ability to select the specimen that represents the lesion or disease process. Poor selection can result in inaccurate interpretation. Improper samples or samples deemed non-diagnostic will not be processed. Specimens should include grossly observable lesions with a small amount of adjacent normal tissue.
- Tissue specimens should include the surface and all anatomical features; i.e., specimens of the kidney should include the cortex, medulla, and pelvis.
- Entire brain should be removed and cut longitudinally on the midline into two equal portions; 1/2 should be submitted in neutral-buffered 10% formalin for histopathology and 1/2 submitted fresh for other test procedures, as indicated. Gross examination by an experienced pathologist is often necessary to locate focal lesions for further sectioning - random sampling may miss important lesions.
- Specimens (except the brain) should be 0.5 cm to 1 cm thick. Specimens that are too thin cannot be properly trimmed for sectioning and those that are too thick decompose before they are fixed (formalin penetrates approximately 3 mm on each side of the section per day at room temperature).
- Fixation must begin as soon as possible after a specimen is obtained. The volume of formalin used should be ten times the volume of the tissue specimen. Samples may be shipped in a smaller volume of formalin after fixation.
- Intestinal specimens requiring examination of villi (i.e. rotavirus and coronavirus infection) require special handling. The preferred method is to tie off approximately 3-cm-long segments of intestine and gently fill the segments with neutral-buffered 10% formalin, using a syringe and hypodermic needle.
- Skin, gastrointestinal endoscopic biopsy specimens, and uterine biopsy specimens should be placed on a piece of tongue depressor, sliced cucumber (GI biopsies), or smooth cardboard (do not use paper). Subcutis, submucosa or serosa, or cut surface, respectively, should be in contact with the wood or cardboard.
- To provide meaningful information for tumor margin evaluation, margins should either be inked with a commercial inking system or have sutures placed and accompanied by a clear indication of specimen orientation.

- The mouth of specimen containers should be wide enough to allow the tissue to drop into the bottle without touching the sides of the opening. Note that unfixed tissue can be easily forced into a jar that has an opening too narrow to allow removal following fixation without breaking the container. Do not use glass containers.

## SHIPMENT

- Use wide-mouth plastic or non-breakable bottles or vials with leak-proof lids. Avoid taping containers shut; it does not prevent leakage (Parafilm™ is acceptable).
- Refer to attached postal guidelines. Note that using the USPS instead of FedEx® or UPS® may result in significantly longer turn-around times. Unless there are special requirements (decalcification or special stains), we will do everything possible to limit turnaround time to one day to process slides and another day for them to be read by pathologists (total turn-around time from receipt of fixed tissue is typically 3 days or slightly longer).
- Pack the specimens with adequate padding to prevent breakage.
- For multiple samples from a single patient, samples should be clearly differentiated in separate containers or somehow marked (ink, suture) if submitted in the same container.

## SUBMISSION FORM

- Provide ALL the information requested on the form.
- Brief, concise, complete histories are required and aid in providing diagnoses and pertinent advice.
- Please use black indelible ink and write or print legibly. List the tissues submitted and the number of samples. This will help ensure that all submitted specimens are identified and examined. Always indicate the source or location from which the specimen was obtained.

## CYTOLOGY

- The laboratory offers cytologic examinations, and peripheral blood smear examinations for blood-borne parasites. However, we do not perform clinical chemistries, CBC's or differential blood counts. These can best be accomplished by commercial laboratories.
- Fine-needle aspiration of lesions for cytology is typically safe and easy and often yields valuable information. However, cytology does have its limitations. Material collected may not always represent the ongoing process. For example, large quantities of blood in an aspirate may represent part of the pathologic process or be due to the aspiration procedure. Aspiration of some lesions, such as dense fibrous tumors or tissues, may not yield sufficient cells. The quality of the sample strongly influences the diagnostic potential of cytology. Therefore, close attention must be paid to slide preparation and handling. Ideally, preparations should be thin enough to visualize individual cells but cellularity must be sufficient for diagnosis. Samples should be handled gently to prevent artifact. Because fresh cells make the best preparation, slides should be prepared promptly after collection.
- Fine needle aspiration
  - Use a 22-gauge needle with 12-mL syringe and pre-cleaned slides.
  - Take several vigorous aspirates from mass.
  - To avoid rupturing of cells, release suction pressure before removing the needle from mass. Often the specimen will be contained only in the hub of the needle.
  - After withdrawing the needle from the mass, remove the needle from the syringe. Then, fill the syringe with air, replace the needle and use aspirated air to force cellular material onto the slide.
  - Make a "squash" or "pull-apart" smear by covering the material on the slide with another slide, squashing the material on the slide with slight digital pressure and then pulling the slides apart. This must be done immediately because cytologic material often clots rapidly. Note that excessive pressure will destroy labile cells such as neoplastic lymphocytes.
  - Please send 3 or 4 unstained, air-dried smears.

**Note:** Lymph node aspirates must be handled gently. Lymphocytes are frequently damaged if shear force is applied to them. This is especially true in the case of malignant lymphoblasts. Slides should be squashed together with slight digital pressure and pulled apart vertically rather than horizontally to avoid shear force.
- Imprints – Imprints or touch preps can be made from solid tissue. A fresh surface should be blotted to remove the majority of surface blood. Several imprints per slide should be made. Material should not be smeared.
- Scrapings – Fibrous tissues are best sampled by scraping. A fresh surface is cut and then scraped with a clean scalpel or razor blade. The material is then gently spread across the slide.

- Body fluids and washes – Slides from turbid fluid samples can be made in the same manner as peripheral blood slides. Clear or slightly turbid fluids should be centrifuged and the sediment spread on slides immediately after collection, to avoid cellular degeneration. Cellular degeneration will occur within 2 to 3 hours after collection.
- Evaluation for blood parasites – Submit 2 unstained, air dried blood smears (fresh blood preferable to avoid artifacts caused by EDTA).
- Only slides prepared at the time of collection will be examined. Do not submit fluids or blood for microscopic evaluation.

For further information on cytology procedures see:

1. Baker R, Lumsden JH. *Color atlas of cytology of the dog and cat*. St Louis: Mosby, 2000.
2. Cowell RL, Tyler RD, Meinkoth JH, et al. *Diagnostic cytology and hematology of the dog and cat*. 3<sup>rd</sup> edition. St Louis: Mosby, 2008.
3. Rebar AH. Collection techniques in veterinary cytology. In: *Handbook of veterinary cytology*. St. Louis: Ralston Purina Company, 1978.

## PARASITOLOGY

The KAHDL no longer accepts fecal samples for parasitology. Identification of ectoparasites is available. Cryptosporidia may be identified in a fecal smear submitted to the Bacteriology Section. We recommend the University of Tennessee Diagnostic Services for parasitology:

<http://www.vet.utk.edu/diagnostic/index.php>

## SEROLOGY/IMMUNOLOGY/MOLECULAR DIAGNOSTICS

*Brucella abortus* antigen is used for bovine testing and porcine testing (anti-*B abortus* antibodies cross react with *B suis*), as is the BAPA test, which is also used for testing cervidae. Testing for *B abortus* and *B suis* requires submission of a completely filled out Brucellosis Test Record (Form 4-33). The expected turnaround times for selected analyses based on batch sample testing are:

- 24-hour turnaround is expected on agglutination tests, AGID tests
- 48-hour turnaround is expected on fluorescent antibody testing
- 4-day turnaround is expected on ELISAs
- 1-week turnaround is expected on serum neutralization tests

Note that turnaround times are calculated from the time at which the laboratory receives the sample. Shipping by overnight **UPS®** or **FedEx®** is always recommended to speed up the testing process. We do not recommend USPS shipping.

### Key to abbreviations for serology tests:

AGID – Agar gel immunodiffusion	CA – Card agglutination	ELISA – Enzyme-linked immunosorbent assay
FA – Fluorescent antibody (direct)	HI – Hemagglutination inhibition	IFA – Indirect fluorescent antibody
MA – Microagglutination	SPA – Serum plate agglutination	PCR – Polymerase chain reaction
SPT – Standard plate test	BAPA – Buffered acidified plate antigen	

## SEROLOGY/IMMUNOLOGY/MOLECULAR DIAGNOSTIC TESTS BY SPECIES

AVIAN					
TEST	SPECIMEN	SHIPPING	TEST TYPE	DAYS RUN	COMMENTS
Avian influenza (AI)	Serum (at least 1 mL)	Refrigerate	ELISA	M-F	
			AGID	M-Th	Not run on Friday (24-hour test)
	Respiratory or cloacal swab-inoculated broth		PCR		Requires special request – please notify State Vet's office before submission.
	Respiratory or cloacal swab-inoculated broth	Refrigerate	Strip	M-F	Reported as positive or negative
Eastern equine encephalitis virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.
Newcastle disease virus	Respiratory or cloacal swab-inoculated broth	Refrigerate	PCR		Requires special request – please notify State Vet's office before submission.
<i>Mycoplasma gallisepticum</i> and <i>M. synoviae</i> (MS-MG)	Serum	Refrigerate	ELISA, HI	M-F	Requires at least 1 mL of serum
<i>Salmonella pullorum</i>	Serum (at least 1 mL)	Refrigerate	MAT	M-Th	
West Nile Virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.

CANINE					
TEST	SPECIMEN	SHIPPING	TEST TYPE	DAYS RUN	COMMENTS
Blastomycosis	Serum	Refrigerate	AGID	M-F	Reported as positive or negative.
<i>Brucella canis</i>	Serum	Refrigerate	IFA	M-F	Tested at 1:10, 1:50, 1:250, 1:1250 dilutions.
Canine adenovirus	Liver, lung, kidney, spleen	Refrigerate	FA	M-F	Reported as positive or negative
Canine coronavirus	Small intestine, lymph node	Refrigerate	FA	M-F	Reported as positive or negative
Canine distemper virus	Lung, kidney, spleen, bladder, brain, stomach, liver, blood smear, CSF, conjunctival swab	Refrigerate	FA	M-F	Reported as positive or negative
	Serum		IFA	M-F	IgG, IgM determination. Tested at 1:10, 1:50, 1:250, 1:1250 dilutions
Canine influenza virus	Nasal/oropharyngeal swab	Refrigerate, red top tube with drops of saline or viral transport medium	PCR	Batch	Detects subtype H3N8 (not H3N2). Reported as positive or not detected.
Canine parvovirus (CPV)	Intestine, tongue, spleen, mesenteric lymph node	Refrigerate	FA	M-F	Reported as positive or negative.
	Serum	Refrigerate	IFA	M-F	IgG, IgM determination. Tested at 1:10, 1:50, 1:250, 1:1250 dilutions.
Eastern equine encephalitis virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.
<i>Clostridium difficile</i> toxins A and B	Bacterial isolate, feces	Refrigerate	PCR	M-F	Reported as positive or not detected.
<i>Clostridium perfringens</i> toxin typing	Feces, bacterial isolate	Refrigerate	PCR	M-F	Reported as not detected or by toxin type.
<i>Ehrlichia canis</i>	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:10, 1:50, 1:250, 1:1250 dilutions.
Herpesvirus	Lung, liver, kidney, spleen, lymph node	Refrigerate	FA	M-F	Reported as positive or negative

	Serum		IFA		Tested at 1:10, 1:50, 1:250, and 1:1250 dilutions.
Histoplasmosis	Serum	Refrigerate	AGID	M-F	Reported as positive or negative
Leptospirosis	Urine, kidney, liver, placenta	Refrigerate	PCR	M-F	Reported as positive or not detected. Submit urine within 24 hours of collection.
Listeriosis	Serum	Refrigerate	Agglutination	Batch	Tested at 1:20, 1:40, 1:80, and 1:160 dilutions
Lyme disease (Borreliosis)	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:64, 1:128, 1:256, and 1:512 dilutions
<i>Neospora caninum</i>	Serum		IFA		Tested for IgG at 1:50 dilution
Rocky Mountain Spotted Fever (RMSF)	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:64, 1:128, 1:256, 1:512 dilutions
Rotavirus	Feces, intestine (colon, ileum, and jejunum)	Refrigerate	ELISA	BATCH	Reported as positive or negative.
West Nile Virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.

## EQUINE

TEST	SPECIMEN	SHIPPING	TEST	DAYS RUN	COMMENTS
<i>Brucella abortus/suis</i>	Serum	Refrigerate	BAPA	M-F	Reported as positive or negative.
<i>Clostridium difficile</i> toxins A and B	Bacterial isolate, feces	Refrigerate	PCR	M-F	Reported as positive or not detected.
<i>Clostridium perfringens</i> toxin typing	Feces, bacterial isolate	Refrigerate	PCR	M-F	Reported as not detected or by toxin type.
Eastern equine encephalitis virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.
Equine infectious anemia (EIA) Coggin's test	Serum	Refrigerate	AGID	M-F	Complete EIA form (VS Form 10-11). Submitting veterinarian's signature is required. Results available within 4 business days. Submit tubes in slotted blood boxes (available at no cost; 615-837-5231).
Equine influenza virus	Nasal swab/wash, lung	Refrigerate	PCR	Batch	Reported as positive or not detected.
Equine herpesvirus (EHV)	Serum	Refrigerate	SN	M, Th, F	Tested at 1:8, 1:16, 1:32, 1:64, 1:128, 1:256, 1:512
Equine herpesvirus-1 (EHV-1) <b>respiratory, neurologic</b>	Nasal swab/wash, brain, CSF, non-clotted blood	Refrigerate	PCR	M-F	Reported as positive or not detected.
Equine herpesvirus-1 (EHV-1) <b>respiratory, abortion</b>	Mare: nasal swab/wash, non-clotted blood (10 mL), uterine swab, placenta. Fetus: stomach fluid, liver, lung.	Refrigerate	PCR	M-F	Reported as positive or not detected.
Equine herpesvirus-2 (EHV-2) <b>conjunctivitis, rhinitis, pharyngitis</b>	Non-clotted blood (10 mL), nasal swab/wash, conjunctival swab, lung	Refrigerate	PCR	M-F	Reported as positive or not detected.
Equine herpesvirus-3 (EHV-3) <b>equine coital exanthema</b>	Reproductive tract swab (penis, vulva, vagina)	Refrigerate	PCR	M-F	Reported as positive or not detected.
Equine herpesvirus-4 (EHV-4) <b>respiratory</b>	Swab/wash from upper resp. tract, lung, non-clotted blood (10 mL)	Refrigerate	PCR	M-F	Reported as positive or not detected.
Equine herpesvirus-5 (EHV-5) <b>multinodular pulmonary fibrosis, lymphoma</b>	Non-clotted blood (10 mL), nasal swab/wash, lung	Refrigerate	PCR	M-F	Reported as positive or not detected.
Leptospirosis	Urine, kidney, liver, placenta	Refrigerate	PCR	M-F	Reported as positive or not detected. Submit urine within 24 hours of collection.
Listeriosis	Serum	Refrigerate	Agglutination	Batch	Tested at 1:20, 1:40, 1:80, and 1:160 dilutions
Lyme disease (Borreliosis)	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:64, 1:128, 1:256, and 1:512 dilutions

Equine protozoal myeloencephalitis (EPM) <i>Sarcocystis neurona</i>	Spinal cord, fresh or formalin-fixed	Frozen if fresh	PCR	M-F	Reported as positive or not detected.
Potomac Horse Fever ( <i>Neorickettsia</i> )	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:80 and 1:160 dilutions.
Rotavirus	Feces, intestine (colon, ileum, and jejunum)	Refrigerate	ELISA	Batch	Reported as positive or negative.
West Nile Virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.
	Serum	Refrigerate	ELISA	M-F	Reported as positive or negative.

FELINE					
TEST	SPECIMEN	SHIPPING	TEST TYPE	DAYS RUN	COMMENTS
<i>Clostridium difficile</i> toxins A and B	Bacterial isolate, feces	Refrigerate	PCR	M-F	Reported as positive or not detected.
Eastern equine encephalitis virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.
Feline calicivirus	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:10, 1:50, 1:250 and 1:1250 dilutions.
Feline herpes virus	Nasal swab, conjunctival swab, tonsil, trachea, lung	Refrigerate	FA	M-F	Reported as positive or negative.
	Serum		IFA		Tested for IgG at 1:10, 1:50, 1:250 and 1:1250 dilutions.
Feline infectious peritonitis (FIP)	Affected tissues (kidney, liver, spleen, lymph nodes)	Refrigerate	FA	M-F	Reported as positive or negative.
	Serum, pleural or peritoneal fluid		IFA	M-F	Tested for IgG at 1:6400 dilution.
	Note: PCR testing for FIP virus in circulating monocytes is currently the diagnostic test of choice for FIP.				
Feline panleukopenia (FPL)	Small intestine, lymph node, liver, kidney, spleen, fetal tissues	Refrigerate	FA	M-F	Reported as positive or negative.
	Serum		IFA		Tested for IgG at 1:10, 1:50, 1:250, 1:1250 dilutions.
Leptospirosis	Urine, kidney, liver, placenta	Refrigerate	PCR	M-F	Reported as positive or not detected. Submit urine within 24 hours of collection.
Listeriosis	Serum	Refrigerate	Agglutination	Batch	Tested at 1:20, 1:40, 1:80, and 1:160 dilutions
Toxoplasmosis	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:16, 1:32, 1:64, 1:128 dilutions.
West Nile Virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.

PORCINE					
TEST	SPECIMEN	SHIPPING	TEST TYPE	DAYS RUN	COMMENTS
<i>Brucella abortus/suis</i>	Serum	Refrigerate	BAPA	M-F	Reported as positive or negative. Requires completed Form 4-33 (Brucellosis Test Record) and official animal ID.
Circovirus	Lung, spleen, liver, kidney, lymph node, heart, intestine	Refrigerate	FA	M-F	Results reported as positive or negative.
	Serum		IFA		Tested at 1:16, 1:32, 1:64, & 1:128 dilutions.
Classical swine fever (CSF)	Nasal swab, tonsil, tonsil scraping	Refrigerate	PCR	Batch	Requires special request – please notify State Vet's office before

					submission.
<i>Clostridium difficile</i> toxins A and B	Bacterial isolate, feces	Refrigerate	PCR	M-F	Reported as positive or not detected.
<i>Clostridium perfringens</i> toxin typing	Feces, bacterial isolate	Refrigerate	PCR	M-F	Reported as not detected or by toxin type.
Eastern equine encephalitis virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.
Foot and mouth disease (FMD)	Swab or tissue from lesion	Refrigerate	PCR	Batch	Requires special request – please notify State Veterinarian's office before submission.
Leptospirosis	Urine, kidney, liver, placenta	Refrigerate	PCR	M-F	Reported as positive or not detected. Submit urine within 24 hours of collection.
Listeriosis	Serum	Refrigerate	Agglutination	Batch	Tested at 1:20, 1:40, 1:80, and 1:160 dilutions
Porcine parvovirus	Fetal tissues	Refrigerate	FA	M-F	Reported as positive or negative.
	Fetal serum or fetal fluid		IFA		Tested for IgG at 1:10, 1:50, 1:250, 1:1250 dilutions. A single serum sample from the dam is of little value because the breeding herd is often seropositive.
Porcine respiratory and reproductive syndrome (PRRS)	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:20 dilution.
Pseudorabies	Brain, lung, tonsil, kidney	Refrigerate	FA	M-F	Reported as positive, negative or suspect.
	Serum		ELISA	Batch	
Rotavirus	Feces, intestine (spiral colon, ileum, and jejunum)	Refrigerate	ELISA	BATCH	Reported as positive or negative.
Seneca Valley virus	Swab or tissue from lesion	Refrigerate	PCR	M-F	Reported as positive or not detected.
Transmissible gastroenteritis (TGE) virus	Intestine	Refrigerate	FA	M-F	Reported as positive or negative.
West Nile Virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.

## RUMINANT (CATTLE, SHEEP, AND GOATS)

TEST	SPECIMEN	SHIPPING	TEST TYPE	DAYS RUN	COMMENTS
<i>Anaplasmosis (cattle only)</i>	Serum	Refrigerate	ELISA	BATCH	For export, advance notice (> 1 week) required
<i>Bluetongue (BT)</i>	Serum	Refrigerate	AGID	M-F	Reported as positive or negative.
<i>Bovine coronavirus</i>	Intestine, trachea	Refrigerate	FA	M-F	Reported as positive or negative.
<i>Bovine leukosis virus (BLV)</i>	Serum	Refrigerate	AGID	M-F	Reported as positive or negative.
<i>Bovine respiratory syncytial virus (BRSV)</i>	Lung, bronchial lymph node, trachea	Refrigerate	FA	M-F	Reported as positive or negative.
	Serum	Refrigerate	IFA		Tested for IgG at 1:50 dilution.
<i>Bovine trichomoniasis</i>	Preputial wash, vaginal wash	Avoid temperature extremes	PCR	BATCH	Sample must be submitted in InPouch™ TF pouch. Results are reported as positive or negative.
<i>Bovine viral diarrhea (BVD)</i>	Lung, intestine, trachea, lesion swabs, liver, spleen, fetal tissue, placenta	Refrigerate	FA	M-F	Reported as positive or not detected.
	Serum		SN	BATCH	Tested at: 1:8, 1:16, 1:32, 1:64, 1:128, 1:256, 1:512 dilutions.
<i>BVD persistent Infection (PI)</i>	Serum, ear notch	Refrigerate	ELISA	BATCH	Follow instructions for ear notch submission. Reported as positive or negative. Analyzed in batches as dictated by demand. For serum, calf must be > 3 months old.

## RUMINANT (CATTLE, SHEEP, AND GOATS)

TEST	SPECIMEN	SHIPPING	TEST TYPE	DAYS RUN	COMMENTS
<i>Brucella abortus/suis</i>	Serum	Refrigerate	Card, BAPA, SPT	M-F	Card test used for small ruminants, BAPA used for all other testing. Reported as positive or negative. Requires completed Form 4-33 (Brucellosis Test Record)
Calf diarrhea panel – Bacterial ( <i>Salmonella</i> , <i>E. coli</i> K99, <i>Cryptosporidium</i> )	Feces, scraping of small intestinal mucosa, bacterial isolate	Refrigerate	PCR	M-F	Reported as positive or not detected.
Calf diarrhea panel – Viral ( <i>Coronavirus</i> , <i>Rotavirus</i> )	Feces, scraping of small intestinal mucosa, viral isolate	Refrigerate	PCR	M-F	Reported as positive or not detected.
Caprine arthritis-encephalitis (CAE)	Serum	Refrigerate	AGID	M-F	Reported as positive or negative.
<i>Clostridium difficile</i> toxins A and B	Bacterial isolate, feces	Refrigerate	PCR	M-F	Reported as positive or not detected.
<i>Clostridium perfringens</i> toxin typing	Feces, bacterial isolate	Refrigerate	PCR	M-F	Reported as not detected or by toxin type.
<i>Clostridium</i> spp ( <i>chauvoei</i> , <i>novyi</i> , <i>septicum</i> , <i>sordellii</i> )	Gangrenous muscle, liver, lung, spleen, heart	Refrigerate	FA	M-F	Reported as positive or negative.
Eastern equine encephalitis virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.
Epizootic hemorrhagic disease (EHD)	Serum	Refrigerate	AGID	BATCH	Reported as positive or negative.
Foot and mouth disease (FMD)	Swab or tissue from lesion	Refrigerate	PCR	Batch	Requires special request – please notify State Veterinarian's office before submission.
Infectious bovine rhinotracheitis (IBR)	Lung, trachea, turbinate, fetal tissue, lymph node, eye swabs	Refrigerate	FA	M-F	Reported as positive or negative. Submit duplicate eye swabs.
Calf diarrhea panel – Bacterial ( <i>Salmonella</i> , <i>E. coli</i> K99, <i>Cryptosporidium</i> )	Serum	Refrigerate	SN	BATCH	Tested at 1:8, 1:16, 1:32, 1:64, 1:128, 1:256, 1:512 dilutions.
Leptospirosis	Urine, kidney, liver, placenta	Refrigerate	PCR	M-F	Reported as positive or not detected. Submit urine within 24 hours of collection.
Listeriosis	Serum	Refrigerate	Agglutination	Batch	Tested at 1:20, 1:40, 1:80, and 1:160 dilutions
Lyme disease (Borreliosis)	Serum	Refrigerate	IFA	M-F	Tested for IgG at 1:64, 1:128, 1:256, and 1:512 dilutions
<i>Mycobacterium avium</i> subsp <i>paratuberculosis</i> (Johne's disease)	Serum	Refrigerate	ELISA	BATCH	Results reported as positive or negative.
<i>Clostridium</i> spp ( <i>chauvoei</i> , <i>novyi</i> , <i>septicum</i> , <i>sordellii</i> )	Feces	Refrigerate	PCR	BATCH	Reported as positive or not detected.
<i>Mycoplasma bovis</i>	Bacterial isolate, nasal swab, conjunctival swab, milk, lung	Refrigerate	PCR	M-F	Cattle only. Reported as positive or not detected.
<i>Neospora caninum</i> (cattle only)	Serum	Refrigerate	IFA	M-F	Tested at 1:200 dilution.
Ovine progressive pneumonia (OPP)	Serum	Refrigerate	AGID	M-F	Reported as positive or negative.
Q fever ( <i>Coxiella</i> ) (goats only)	Serum	Refrigerate	IFA	M-F	Tested at 1:16, 1:32, 1:64, 1:128 dilutions.
Rotavirus	Feces, intestine (spiral colon, ileum, and jejunum)	Refrigerate	ELISA	BATCH	Reported as positive or negative.
Toxoplasmosis	Serum	Refrigerate	IFA	M-F	Tested at 1:16, 1:32, 1:64, 1:128 dilutions.
West Nile Virus	Brain	Refrigerate	PCR	Batch	Reported as positive or not detected.

## TOXICOLOGY

For routine toxicology testing, samples received by the KAHDL are analyzed by a subcontractor, the Toxicology Section, Division of Consumer and Industry Services, Tennessee Department of Agriculture. Submission to KAHDL implies authorization for subcontracting to this laboratory. When poisoning is suspected, please notify the KAHDL at the time of submission. Please specify on submission forms which test should be run on which tissue (e.g., “Test liver for anticoagulant rodenticides”) – just indicating “toxicology” is not sufficient. When submitting serum, the serum should be removed from the blood clot or a serum separator tube should be used.

Test	Minimum Sample Req'd	Comments
Aflatoxin	20 g feed (corn, grain)	No hay or silage.
Anticoagulants	5 mL serum, blood, plasma 10 g bait 10 g stomach contents 10 g liver (best)	Includes 12 common anticoagulant rodenticides. Avoid submitting samples in medicine bottles. Do not freeze sample.
Arsenic	5 g liver or kidney 5 mL urine 5 mL blood 5 g stomach contents 5 mL water 5 g feed	Liver or kidney is the preferred sample from a dead animal. Feed, blood, and stomach contents are best from a live animal.
BUN	2 mL ocular fluid 2 mL serum or plasma	
Calcium	2 mL ocular fluid 2 mL serum 2 mL CSF	
Carbamates (Pesticide Screen)	5 g stomach contents 5 g rumen contents 5 g bait 10 g feed	Avoid submitting samples in medicine bottles or plastic containers. Glass is preferred.
Copper	5 mL serum 5 g liver or kidney 20 g feed	Liver is preferred over kidney.
Cyanide (prussic acid)	1 lb. dry plants or 5 lb. wet plants 10 mL blood 50 g muscle (heart) Rumen contents	Used for plants with cyanogenetic potential (eg., sorghums, Sudan grass, corn). Samples should be frozen as soon as possible for shipment to the laboratory.
Fumonisin	20 g feed	No hay or silage – only corn.
Iron	10 mL serum, liver, kidney	No hemolyzed samples. Non-routine test, please call lab before submission.
Lasalocid	20 g feed	
Lead	5 mL blood, (EDTA, heparin) 5 g liver and kidney 5 mL water 5 g stomach contents	Heparin is preferred. Submit both liver and kidney.
Magnesium	2 mL ocular fluid or 2 mL serum	
Monensin	20 g feed	
Nitrate (qualitative)	2 mL ocular fluid	
Nitrate (quantitative)	1 lb. dry forage 5 lb. wet forage 1 pint water 1 mL serum or urine	
Ochratoxin	20 g feed (grain)	No hay or silage.
Organochlorines (pesticide screen)	5 g stomach contents, rumen contents, feed, or bait	Avoid submitting samples in medicine bottles or plastic containers. Glass is preferred.
Organophosphates (pesticide screen)	5 g stomach contents, rumen contents, feed, or bait	Avoid submitting samples in medicine bottles or plastic containers. Glass is preferred.
Phosphorus	5 mL serum	
Potassium	2 mL ocular fluid or 2 mL serum	
Selenium	10 g liver or kidney 10 mL whole blood or serum	Whole blood is preferred over serum.
Sodium	2 mL ocular fluid, serum, or urine	
Strychnine	5 g stomach contents 5 mL urine, serum, or blood 10 g bait	Avoid submitting samples in medicine bottles.
Vomitoxin	20 g feed (grain)	No hay or silage.
Zearalenone	20 g feed (corn and small grains)	No hay or silage.
Zinc	5 g liver, kidney, or stomach contents 5 mL serum 10 g feed	Serum sample should be in Royal blue top Vacutainer® tube for trace metal analysis.

### OTHER SOURCES OF HELP IN TENNESSEE

State Veterinarian, Dr. Charles Hatcher – regulatory, interstate shipping, health certificates	615.837.5120
West Tennessee Animal Disease Diagnostic Lab, UT-Martin	731.881.7952
University of Tennessee College of Veterinary Medicine	423.974.8387
Necropsy, days	865.974.5673
Necropsy, after-hours	865.974.5701
Tennessee Wildlife Resources Agency (TWRA)	615.781.6500
Tennessee Department of Health: <a href="http://health.state.tn.us/contact.htm">http://health.state.tn.us/contact.htm</a>	615.741.7247
Dr. Mary-Margaret Fill, Medical Epidemiologist (human exposure rabies information)	615.741.7247
USDA-APHIS Veterinary Services (Federal services)	615.781.5310
State/Federal Brucellosis Records (Brucellosis results)	615.837.5120

**Rabies Laboratories:**

Nashville:

Tennessee Dept of Health Lab Services, 630 Ben Allen Rd, Nashville, TN 37247  
 Mail: PO Box 305130, Nashville, TN 37230-5130 615.262.6350

Knoxville:

Knoxville Branch Rabies Laboratory (East Tennessee Regional Office):  
 1522 Cherokee Trail, PO Box 59019, Knoxville, TN 37950-9019 865.549.5201

**ASPCA ANIMAL POISON CONTROL CENTER (APCC) AND PET POISON HELPLINE**

For any animal poison-related emergency, 24 hours a day, 365 days a year. The APCC charges a consultation fee of \$65.00; call 888-426-4435. The Pet Poison Helpline charges a consultation fee of \$59.00 that covers the initial consultation and follow-up calls associated with the case; call 800-213-6680.

**TENNESSEE POISON CONTROL CENTER (HUMAN POISONINGS)**

The Tennessee Poison Center (TPC) provides immediate treatment advice for poisoning emergencies at 800-222-1222. They are available 24 hours a day, 365 days a year. TPC also provides information about poisons and poison prevention. TPC can provide information about household products, chemicals at work or in the environment, drugs (prescription, over-the-counter, herbal and illegal), snake and spider bites, and chemical terrorism. A specially trained nurse, pharmacist, or doctor will help. Calls are free and confidential.

## REPORTABLE DISEASES

The reporting of evidence of certain animal diseases is a requirement under Standards for Accreditation of Veterinarians in Tennessee and other states. Such reporting is also required by State law in Tennessee. It is the veterinarian's professional responsibility to report these diseases properly to ensure that appropriate control measures are instituted. Reportable diseases in general include all diseases for which control or eradication programs are in effect, and all foreign diseases (not known to exist in this country). Reportable diseases currently include but are not limited to the following:

Species	Avian	Equine	Bovine	Sheep, Goat	Porcine	Dog, Cat	Farm Elk, Deer
Avian influenza (fowl plague)	X						
<i>Salmonella gallinarum</i> (fowl typhoid)	X						
<i>Salmonella pullorum</i> (pullorum disease)	X						
Velogenic viscerotropic Newcastle disease	X						
Anthrax		X	X	X	X		X
Brucellosis			X		X		X
Psoroptic scabies			X				
Screw worm		X	X	X	X	X	X
Tuberculosis			X				X
Vesicular diseases		X	X	X	X		
Bovine spongiform encephalopathy			X				
Bluetongue				X			
Rabies		X	X	X	X	X	X
Scrapie				X			
Scabies				X			
Equine infectious anemia		X					
Piroplasmiasis		X					
Viral encephalitis		X					
African swine fever					X		
Hog cholera					X		
Pseudorabies					X		
Pox or lumpy skin conditions							
Chronic wasting disease							X

Rabies is reportable in all species. Suspected or known rabies infection should also be reported to local public health authorities. Reporting should include telephone or written notice to the State Veterinarian's office and submission of samples to the appropriate laboratory.