General Information and Guidelines

Please refer to the test information on the website for specific guidelines on type of specimen submitted. The following is intended to serve as a guideline. We do request that you use collection containers or swabs that are not expired, sterile, and appropriate for the testing.

Submission of blood tubes

Note: Do not submit blood samples in syringes (especially with needles attached). Syringes that are submitted to the lab with the needles attached will not be tested, and a handling fee will be assessed!

1. Collect blood samples in clean sterile tubes (Vacutainer tubes). Fill tubes 3/4 full. If drawing blood into a purple topped tube (EDTA), or any tube that contains an anticoagulant, gently invert 5-6 times after collection to mix. For red top tubes, allow to stand at room temperature for a few hours to permit a solid clot to form and retract. Submission of blood samples in the original blood tube is preferred, unless there is the possibility of freezing. However, if weather conditions are such that the blood may become too cold or too hot, the serum should be removed, placed in a separate tube, then shipped. Do not place blood tubes in windows as direct sunlight can cause the red cells to lyse, and may impact the usability of the specimen. We recommend shipping to the laboratory in the shortest time possible, 48 hours maximum transport time.

2. Red-topped tubes are recommended for serological tests. EDTA (purple top tubes) are primarily used for hematology evaluation, PCR testing and viral isolation.

3. Identify each sample individually. Identification should be on the tube and NOT on the stopper.

4. A completed accession form must accompany each submission. If sending in more than one animal, use the continuation form and attach it to the Accession form. Please number sample tubes to match numbers from Multiple Animal ID form.

5. All regulatory charts must include complete animal identification and veterinarian signature. Sheets that are not completed properly will be sent back to the ordering veterinarian. Testing will not be done until the completed form is returned.

6. Package specimens according to the shipping guidelines and send to the lab for testing. Overnight delivery for all shipments to the lab is preferred, and no more than 48 hours transport time is recommended.

7. Note: for serological testing, 10-14 days are required for the development of demonstrable antibodies. If a blood sample is drawn during the acute phase of a disease, a second sample should be drawn 10-14 days later. If testing for export, subclinical carriers, pre-sale, pre-breeding examination, clinically normal animals, etc, contact the laboratory for the best samples to collect.
Histopathology specimens

1. Specimens should represent typical lesions, including active margins and adjacent (normal) tissue, rather than lesion cores or curetted debris. Autolysis, freezing, mutilation (forceps crushing or tearing), or removal of small samples by electrocautery will make samples unsuitable for proper evaluation.

2. Multiple specimens (from different sites or types of lesions) should be identified individually by size, suture tags, or separate containers. Samples that are < 1cm should not be sliced, but fixed and submitted for testing. Brain and eyes should be fixed whole.

3. Nearly all diagnostic histopathology can begin with tissue fixed in 10% buffered formalin.

4. The volume of fixative should be 10-20 times the specimen volume. After 12-24 hours, specimens can be transferred to just enough formalin to keep them moist during shipment. There is no need to pay for transport of excess fixative. The tissues should be submitted overnight to the lab.

5. Wide-mouth bottles are preferred containers. Narrow-mouth bottles often have to be broken or cut to release fixed tissues. Plastic bottles are better than glass; anticipate rough handling during shipment, and package accordingly. To prevent leakage tape the lid, or wrap a piece of parafilm™ around the cap. Label the container with animal ID and source of tissue.

6. Place the bottle in a leak proof secondary container. Place container and accession form into a mailing container. Mail to the lab as soon as possible.

General Bacteriologic Considerations

Used correctly, microbiologic cultures can identify etiologic agent(s) and contribute key information towards a diagnosis and treatment. However specimens that are not collected properly or shipped properly can lead to erroneous diagnoses. Etiologic agents may be missed because of improper transport medium, improper transport environment, or improper preservation techniques. We offer the following guideline to optimize these procedure for sample collection and submission.

If you have any questions please email Dr. Erdal Erol or Dr. Deborah Maples or by phone at: 859-257-8283.

1. Samples should be collected aseptically and placed in sterile plastic bags (e.g., Whirl-pak) or heat sterilized containers. Seal tightly. Do not use chemically disinfected
containers, or plastic gloves or sleeves.

2. **Label** all submissions with the location (tissue) and species of origin. The same bacterial species may be highly significant or a meaningless contaminant, depending on the tissue and/or species from which the sample was obtained. Also, depending on the tissue/species of origin, different culture conditions may be necessary to isolate and identify specific pathogens.

3. Always specify the tests you want done, and the pathogens you suspect, particularly in the case of specimens with normal bacterial flora (feces, intestinal contents, skin, or oral mucus membranes). If we don't know what you're looking for, we may not inoculate the proper media to find it.

4. It is best to collect other samples before opening the gastrointestinal tract. Tissue samples (lung, liver, spleen, kidney, etc.) should be 5 g or larger to allow surface searing in the laboratory to reduce contaminants. Fecal samples should not be submitted in stoppered tubes, as fermentation will dislodge the stoppers.

5. Place each sample in a separate container. If the intestine is to be cultured, tie off both ends of a segment and place it in a separate container.

6. Except in the case of abortions, please separate samples that are to be examined by different laboratory sections. If a specimen is to be examined by both the virology and bacteriology sections, the specimen should be divided, each piece placed in a separate container, and labeled with the source of the tissue and the desired laboratory service.

7. Fluids for culture (e.g. body cavity fluids, pericardial fluid, joint aspirates) should be submitted in a sealed sterile tube, in as large a volume as is available (up to ~10 ml), since the concentration of organisms may be very low in these samples. Fluids may be submitted in blood culture bottles or Isolator tubes for highest sensitivity. Differentiating contaminants from etiologic agents may be difficult from blood culture bottles, due to loss of quantitation information. Fetal fluids (thoracic or peritoneal fluids, or heart blood) to be examined for *Leptospira* sp. by FA test should be submitted in a sealed sterile tube to which 10% buffered formalin is added at a rate of 1.5 ml per 20 ml fluid. **Never submit fluid in syringes**, because they tend to leak in transit and contaminate packaging. **Never submit fluids or other specimens for bacteriologic culture in EDTA (purple top) evacuated tubes**, as EDTA is highly toxic to many bacterial species.
8. Milk samples should be submitted in screw top tubes frozen or placed on ice packs. Less than 1 ml is required, and larger volumes are undesirable, especially if the samples are frozen.

9. Specimens for isolation of anaerobic pathogens require special care. Anaerobic bacteria die in the presence of oxygen and should be shipped in an anaerobic container, such as anaerobic swab or Port-a-Cul system. Cultures for Clostridium sp. in parenchymatous organs and intestines ordinarily provide no significant information concerning the cause of death if the samples are taken more than one hour after death.

10. Some specimens, such as porcine nasal swabs for Bordetella sp. isolation must be delivered to the laboratory within 12 hours of collection. Fastidious organisms such as Campylobacter spp. require special media for transport to the laboratory. Where there is any doubt as to what samples to collect and how to transport them -- CALL THE LABORATORY FIRST at 859-257-8283!

11. When collecting large numbers of samples (e.g. >30 milk samples for mastitis diagnosis or fecal samples for Johne's disease diagnosis), call the laboratory for scheduling. This permits the laboratory to have personnel and media available for prompt processing.

12. Keep specimens cold from the time they are collected until they arrive at the laboratory. Specimens should be shipped in insulated containers with a sufficient number of ice packs to last 48 hours. Specimens arriving in the laboratory in a decomposed state will not be processed, since processing and culture of these tissues lead to meaningless or erroneous results.

13. For cases where bacteremia is suspected and blood culturing is requested, blood culture systems should be inoculated with the proper amount of blood collected aseptically. Single bottle blood culture systems are recommended.

14. Different bacteriologic procedures take different times to complete, turnaround times can be found here:

**Parasitology**

Fecal Specimens
Specimens should be submitted in individual sealed containers labeled with owner's name, animal species, and animal number/name. Whirl Pak bags, Ziplock bags (double bagged), or plastic containers are recommended.

Feces must be fresh for accurate results. Refrigerate fecal sample immediately after collection but do not freeze. Submit sample to the laboratory as soon as possible in a container with blue ice via any of the 24 to 48 hour transport services.

**Mycology**

Sample collection for evaluation for mycotic diseases. (Note: Isolation and identification of mycotic agents may require 30 days.)
1. The affected area of skin should be washed with 70% alcohol, scrapings taken from the active border area and placed in a sterile container. The basal portion of the hair or hair stubble should be plucked out with forceps and submitted as well. Hair and skin scrapings for dermatophyte isolation should be shipped dry at room temperature, between glass slides or in a paper envelope.

2. Exudates or tissues for culture should be collected aseptically, refrigerated, and sent to the laboratory on ice packs. It is strongly recommended that tissues be submitted for histopathology rather than mycology in the initial evaluation of these cases. Please contact the laboratory at: 859-257-8283 for additional information.

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**Virology**

If a viral/ disease is suspected, it is very important to collect specimens as soon as possible following onset of clinical symptoms or death of the animal. If testing for export, subclinical carriers, pre-sale, pre-breeding examination, clinically normal animals, etc, contact the laboratory for the best samples to collect.

Tissue specimens should be placed in sterile sealed containers or plastic bags, (i.e., Whirl-Pak bags) and kept refrigerated. Swab samples (conjunctival, nasal, oral, fecal, vaginal) can best be collected using a universal transport media such as BHI broth. If no media is not available, 2 ml of sterile saline solution (contact lens solution) may be substituted.

Chilled specimens and swabs are preferred. If the time period between collection and receipt at the Laboratory is longer than 24 hours, add an extra ice pack or two.