Selection of Specimens for Bovine

Animals selected for laboratory analysis should be free from antibiotic therapy and in an early or acute disease stage. Selected tissues should be collected as aseptically as possible. A meaningful history of the disease outbreak and a tentative diagnosis, based upon clinical evaluation, should be included. Laboratory test results are directly affected by the selection, preparation, handling, and shipment of selected specimens.

Preparation & Collection of Tissues/Samples:

1. Tissues - Fresh
   1.1 Samples only from freshly dead (or sacrificed) cattle free from antibiotic therapy and in early or acute stage.
   1.2 Samples for isolate collection: bacterial culture, virus isolation, PCR, or intended for autogenous production should be fresh (refrigerated).
      • Fresh tissue samples should be 2x4 inch in size.
      • Place separately in Ziploc® or Whirl-pak® bags.
      These samples should not be fixed in formalin.
   1.3 Intestinal samples should be at least two inches in length. These samples should be kept separate from other samples to avoid contamination in double bagged and sealed Whirl-pak bags. Do not cut the loops of intestines open. The sample should be refrigerated and cooled thoroughly prior to shipping.
   1.4 Avoid shipping all samples over a weekend.
   1.5 Samples of the affected tissue itself are typically preferable to swabs.

2. Swabs
   Use swabs if more appropriate for joint (ante-mortem or post-mortem) or exudate from an ear. It is important to use the correct swab and transport media, such as a Culturette® (or similar), as indicated below:
   2.1 Aerobic bacterial culture: Amies or Stuart’s media
   2.2 Anaerobic bacterial culture: Amies media with charcoal.
   2.3 For abscesses or exudates use a capped syringe with needle removed.
   2.4 For Fusobacterium necrophorum (footrot), use Cary Blair media
   2.5 Mycoplasma culture: viral swab and transport fluid
   2.6 Virus isolation or PCR: viral swab and transport fluid

Note: Use of the incorrect swab and media may jeopardize the ability to detect or culture the offending pathogen. For bacterial isolation, avoid using Mycoplasma or viral media which contain antimicrobials and may inhibit growth of the desired pathogen. Avoid using bacterial culture media to isolate viruses or Mycoplasma organisms.

2.7 All nasal swabs – the external nares and internal nostrils are cleaned with a moist towel to remove common contaminants. (Use swabs with transport media such as Amies or Stuart’s). Insert swab into the pre-cleaned nasal cavity and rotate.

*Identify all swabs with the following:
   Building, pen, pasture or site
   Animal ID number or tattoo
   Fluids, exudates/aspirates, tracheal washes, or urine

3. Histopathology
   3.1 Preparation of Tissue for Fixation
   Multiple sites or types of lesions, to include both normal and diseased tissue and a sample at the line of demarcation, should be taken. The sections should be less than 2 cm thick. The small size of the tissue results in rapid and complete penetration of the fixative.

   Selected tissues should be cut with a sharp scalpel since the squeezing action of scissors crushes and tears tissue. The tissue should be rinsed briefly with 0.85% NaCl to remove adhering blood, since blood will retard fixation. Autolysis or freezing will make samples unsuitable for proper evaluation. Place tissues in double Whirl-paks, identify bags if multiple animals are submitted. Do not use narrow mouth bottles to submit fixed tissues.

3.2 Volume of Fixative
   All samples for histopathology exam should be fixed in 10% neutral buffered formalin. The volume of formalin should be at least equal to 10 times the volume of the sample itself to assure good perfusion of the sample and to maintain the tissue architecture.

3.3 Formula of Fixative:
   37-40% formaldehyde 100 mL
   Distilled water 900 mL
   Sodium phosphate, monobasic monohydrate 4.0 g
   Sodium phosphate, disbasic anhydrous 6.5 g
3.4 Tissue Selection for Histopathology
Check the recommended samples in the guideline table. If the cause of death is unknown, submit samples exhibiting gross lesions, including heart, liver, lung, kidney, spleen, various levels of the gastrointestinal tract, mesenteric lymph nodes and brain.

Note: All hollow organs (intestine or uterus) should be gently flushed with 10% formalin without disturbing the mucosal lining before placing in the formalin bag. Be sure to take proper precautions when handling formalin. Formalin-fixed samples should be kept separate from fresh samples.

Identification and Handling of Blood Samples:

4. Collection of Blood Samples
4.1 Collect in sterile tubes. Serum separator tubes work well. Follow the manufacturer’s directions. Based on the number of tests requested, 1 ml – 3 ml of nonhemolyzed serum is required.
4.2 Fill vacutainer tubes 1/4 full and allow to stand at room temperature for an hour to permit a solid clot to form and retract.
4.3 Pipette the serum into sterile tubes with snap caps (3 ml plastic tubes with snap caps, Falcon #2054, are recommended). Make sure caps are securely closed.
4.4 Use permanent markers and underline the I.D. numbers (e.g. 16 vs. 91).
4.5 Do not freeze whole blood or samples with the clot remaining.
4.6 Contaminated or toxic samples cannot be used in virus isolation tests. Do not use brucellosis test tubes!

5. Identification of Samples on the Request Form
5.1 Identify the tubes on the request form according to the different age groups. Remember to write the age. Start with the youngest group.
5.2 Clearly specify the test(s) requested on the submission form.
5.3 When sending paired sera, identify the acute samples from the convalescent samples on the tube and on the request form.

6. Packing Specimens
To avoid leaking in transit, double bag ALL samples. Whirl-pak bags or equivalent are recommended. Wrap sample bags and 2-4 ice packs in absorbent paper (e.g. newspaper). Place the package into a styrofoam container. Completed submission forms should be inserted in a bag in case of leaking. Avoid mixing intestinal samples with other tissues. If you need more information about shipping specimens to Newport’s Diagnostic Laboratory, please call us at 800-220-2522.

7. Mailing
Newport Laboratories provides free returnable cardboard cartons with a styrofoam lining designed for sample submission. Submission forms are available for microbiology and serology testing. Call us at 800-220-2522 if you would like submission form(s) or shipper containers. Submission forms are also available online at www.newportlabs.com under the “Diagnostic Services” tab. Samples should be submitted by the fastest means possible to avoid deterioration of specimens. Next day or overnight delivery is preferred. The most reliable mailing services that we have found are listed below:
- United Parcel Service (UPS)
- Airborne Express
- Fed Ex
- DHL
- Spee-Dee
- U.S. Parcel Post (only as a final option)

8. Laboratory Hours
The Newport Diagnostic Laboratory is open for service from 8:00 A.M. to 5:00 P.M. (CST) Monday through Friday, with the exception of holidays. Mail is received on Saturdays and those holidays when the postal service and/or private delivery companies are operating.

9. Diagnostic Shipping Address

![Newport Laboratories Logo]

Newport Diagnostic Laboratory
1524 Prairie Drive • PO Box 938
Worthington, MN 56187

Submission forms available online at www.newportlabs.com under the “Diagnostic Services” tab.