Field Necropsy of Cattle and Diagnostic Sample Submission

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KEYWORDS
- Bovine • Necropsy • Diagnostic • Sharpening knives • Shipping specimens • Data collection

KEY POINTS
- Field necropsies can provide a wealth of information that can help guide production management decisions.
- The outlined procedures emphasize not detaching organs from the carcass unless necessary, thereby making carcass removal by rendering companies more efficient and minimizing clean-up on the production unit premise.
- An observation and history collection system using form templates and photographs improves efficiency of recording necropsy results.
- One key to necropsy efficiency, speed, and enjoyment is having sharp knives. The first part of the article includes tips for sharpening knives.

FIELD NECROPSY AND DIAGNOSTIC SAMPLE SUBMISSION

This section includes:
- A brief overview of knife-sharpening skills and sharpening tools
- A step-by-step field necropsy technique for ruminants
- Review of simplified observations collection
- Techniques for handling laboratory samples including proper packaging and shipping of samples to diagnostic facilities

The principal purpose of field necropsies is to gain information that may be used to evaluate production or influence production management decisions. Many production management issues relate to making a complete and thorough assessment of organ systems, including their associated lymph nodes.

Safety is paramount, so always have protective clothing, gloves, and boots that allow for disinfection. Remind all bystanders and observers of the importance of being careful to minimize their personal contamination. Have water, soap, disinfectant, and cleaning brushes readily available.

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KNIFE-SHARPENING SKILLS

A quick alternative to using a knife-sharpening abrasive is to use a “V” carbide blade knife sharpener. These put a very coarse, crudely shaped, yet usable edge on a knife blade.

Sharpening Abrasives

Although 3-sided oil stones work well, these are generally expensive and do not travel well when needed for field necropsies. Solid abrasives such as diamond-coated steel slabs are more durable than stone abrasives. They are easily cleaned and come in an assortment of grits. Most knives can be sharpened nicely with any abrasive that is finer than 300 grit (medium or fine).

Angle consistency, or the angle at which the blade is held as the abrasive is stroked, is by far the most important key to developing a sharp cutting edge. There are several diamond-coated sharpening abrasives designed to maintain a consistent sharpening angle. Most feature a clamp to hold the knife blade, and the abrasive is connected to a rod that slides through angle slots above and below the knife’s cutting edge.

A consistent edge can be maintained on a flat abrasive if the back of the knife blade is pushed into the palm side of the thumb and the side of the thumb is laid flat against the abrasive as it is stroked (Fig. 1). It is difficult to maintain a consistent angle when using a diamond-coated sharpening rod or steel, and for this reason, these sharpening tools are not recommended.

The best knife-sharpening abrasives for veterinary practitioners are motorized. The author strongly encourages having a high-quality diamond-coated, set-angle disk sharpener in clinics. The better diamond hone machines have 3 sharpening stages, meaning that there are 3 slightly different angles for the abrasives. The final stage in these usually is 5° wider, which provides increased durability to the cutting edge. Diamond honing knife sharpeners can be found at most large department stores in the kitchen appliance area. Buy lots of knives and keep several sharp knives in the practice vehicle. Purchase a good-quality motorized knife sharpener and delegate the sharpening to a technician.

Angles of a Sharp Cutting Edge

There is no perfect angle for a cutting edge … but, instead, knives with a cutting edge angle not suitable for the intended job. The steeper the angle, the thinner is the blade near the cutting edge and the less durable is the cutting edge. Although durability is lost with steeper angles, the resistance caused by the knife sliding through the tissue is less. Examples include slicing knives. Similarly, the flatter the sharpening angle, the thicker is the steel to support the cutting edge and therefore the more durable. The kind of angle targeted for axes and shears is shown (Fig. 2). A flat file works well for

Consistent Angle Is Key to Sharpening

To maintain a consistent cutting edge angle, keep your thumb flat against the abrasive and always replace the blade back into the indent created in your thumb while stroking the abrasive after each cutting edge check.

Flat Diamond Coated Abrasive

Fig. 1. A consistent angle can be maintained while developing a cutting edge by holding the back of the knife blade against the thumb and resting the thumb on the flat sharpening abrasive.
sharpening the soft metal found in axes and shears. Necropsy knives seem to work well if the angle is $15^\circ$ to $25^\circ$. Again, the key is keeping the angle constant when developing a cutting edge.

**Finishing the Cutting Edge**

A ceramic sharpening rod is one of the better tools for honing a fine edge on a properly sharpened knife blade. When using a ceramic rod or metal steel, stroke the blade gently, feeling for defects in the cutting edge as the blade slides down the tool.

**Determining When the Edge is Sharp**

A sharp cutting edge should be as smooth as glass. The best and safest way to test the edge is to hold a plastic ink pen at a $45^\circ$ angle and see if the knife blade will sit on the pen without sliding down the pen barrel. If it holds onto the plastic barrel, the cutting edge is sharp. Additionally, no defects should be felt when the plastic barrel of the ink pen is lightly slid down the cutting edge (Fig. 3).

**The Keys to Having Sharp Necropsy Knives**

Do not use the necropsy knife for jobs that will damage its cutting edge. For example, use a disposable bladed box cutter for skin incisions, thereby not damaging the cutting edge of the necropsy knife on hide, hair, and dirt. Do not use a necropsy knife for cutting rib cartilage unless it is of a very young animal. A shear or axe works well.

**TESTING THE CUTTING EDGE**

**FIRST:** See if the edge will slip

**SECOND:** See if the edge is smooth

Fig. 3. Testing the sharpness of a cutting edge is easily done using a plastic ink pen or the thumbnail. Sharp cutting edges will grab the barrel or the thumbnail when held at a $45^\circ$ angle and not slip down. Additionally, a sharp cutting edge will feel smooth as a plastic pen or thumbnail is slid down the edge.

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**Cutting Edge Angles Relate To Use Needs**

- **Common Usage Angles**
  - 35 - 40° Tough ➝ Axe
  - 25 - 30° Durable ➝ Utility
  - 15 - 20° Fine ➝ Meat
  - 10 - 15° Ultra-fine ➝ Shave

- **Angle Degrees and Uses**
  - increasing the final honed angle 5 degrees increases edge durability

Fig. 2. The angle of the knife’s cutting edge determines its durability and the ease with which a sharp cutting edge slides through tissues. Select the edge best suited for the job.
and saves the cutting edge of the knife. Use a ceramic rod to repeatedly touch up the cutting edge during the necropsy.

FIELD NECROPSY PROCEDURE WITH MINIMAL LOOSE PARTS

Important Note

Animals that will be rendered must not contain chemical residues that could be harmful to other animals that would consume rendered products.¹

Start with the Ruminant on its Left Side

Think about what is observed. Collect histopathologic and culture specimens while working. Histopathologic specimens should not be thicker than 5 to 7 mm. Try to connect observations into a unifying diagnosis or production management observation.

The procedure outlined is designed to make it easier for animals to be picked up by rendering trucks and to minimize hide damage, thereby improving the hide value to renderers. Detaching any organs that are not required for examination is not being considerate of the people working for the rendering company and is more likely to create a mess at the farm, ranch, or feedlot on which the necropsy examination is being performed.

Review Anatomy and Gross Pathology

Knowledge of the structure and function of the organ tissues being examined can be key to linking observations to a meaningful diagnosis.²,³ Be slow to jump to diagnostic conclusion based on the first observations. The “lift a leg and look” or “peek-a-boo” necropsies generally leave important production management observation undiscovered and minimize the value of the observations that could have contributed to better animal care and management.

Accessing the Brain

It is important to check with the rendering company serving the animal facility about the acceptability of examining the brain, because some companies will not pick up carcasses that have had the cranium opened. Also remember that rabies should be on the differential list for all central nervous system cases, so take all appropriate precautions.

Fig. 4 demonstrates the appropriate lines for removal of the calvaria. The cut needs to be approximately as deep as the distance from the front of the skull to the lateral

Fig. 4. The steps for opening the skull with an axe to expose the brain. (A) Cut across the face just dorsal to the lateral canthus then cut from the lateral canthus dorsal in front of the ear over to the poll, across the poll to the level of the opposite ear. (B) Using the blunt side of the single-bit axe, strike the edge of the cut bone between the lateral canthus and the ear at a 45° angle. (C) This will break the skull away from the brain.
canthus. Make sure the axe cuts are completely through the cranium. Using the blunt or hammer side of the single-bit axe, strike the cut edge of the cranium along the frontal crest at a 45° angle (see Fig. 4).

To remove the brain, cut the dura mater across the cerebral falx, the tough medial division of the dura. Extend this cut to allow the fingers to slide beneath the cerebrum. Using the necropsy knife, cut between the cerebrum and the cerebellum at the level of thepons and lift the cerebrum out of the cranium. Next, split the dura mater covering the cerebellum dorsally. Slide the tip of the necropsy knife behind the cerebellum into the spinal canal and cut across the spinal cord distal to the obex. Lift out the cerebellum and spinal cord, containing the obex.

**Opening the Hide and Reflecting the Legs**

Using a box cutter, cut along the underside of the jaw, over the larynx, and down the neck over the trachea. The incision should drift toward the animal’s right foreleg axillary space. Continue the skin incision along the ventral thorax, crossing the costal cartilages and along the abdominal wall toward the right rear inguinal area. The incision across the thorax and abdomen will be lateral to the midline 3 to 6 inches (Fig. 5).

Do not cut the hide upward toward the scapula as the foreleg axillary space is passed. The hide is worth half of the value of the carcass to the renderer, and mutilating the hide reduces its value so much that many renderers will not pick up necropsied carcasses without charging a fee if the hide is damaged.

Reflect the rear leg before attempting to reflect the foreleg. To reflect the rear leg, cut the heavy muscles (adductor, semimembranosus, pectineus, and sartorius) that hold the coxofemoral joint in place. The round ligament will be easily identified and the joint examined (see Fig. 5).

The best approach to examining the stifle and hock joints is to start with the rear leg reflected and then skin along the inside of the leg from the stifle joint past the hock joint. To examine the stifle joint, cut along the side of the femoral trochlea and cut above the patella through its attachment to the quadriceps down to the femur. The patella will rotate over, yielding a great view to the stifle joint. To examine the hock joint, slide the necropsy knife between the extensor muscles and the tibia and cut the extensors loose below the stifle joint. retracting the extensor muscles will allow the knife to be slide down to the hock joint, and then pull up the joint capsule, thus allowing one to cut open the joint capsule without invading the joint with the tip of the knife blade. This allows for cleaner joint sampling.

**Fig. 5.** Note the real leg is reflected while the foreleg remains unreflected. Working from the back side, continue to skin forward toward the foreleg. Lift the foreleg and cut through the latissimus dorsi holding the foreleg down.
Working from the back side, continue to skin the carcass toward the fore leg. When skinned to the level of the transverse processes and proximal rib attachments, the latissimus dorsi holding the foreleg down will be easily cut. Move to the sternal side of the animal and lift the foreleg, cutting the pectoral muscles. The foreleg should lay over with only minor fascia dissection.

**Examining the Oral Cavity and Neck Structures**

Incise along the side of the cheek, exposing the premolars and molars. This approach provides a good view of the oral cavity and allows for examination of molar eruption ([Fig. 6](#)). The first molar erupts in cattle at approximately 7 months of age and is in full wear at approximately 12 months. This information can be useful when examining stocker and light feeder cattle.

To examine the tongue and larynx, slide the knife on the caudal side of the hyoid bones, feeling for the bend formed between the epihyoid and the ceratohyoid bones. The knife will generally cut the cartilage connection easily in younger animals. Shears can be used if needed.

Reflect the tongue while dissecting the larynx, trachea, and esophagus. Open the esophagus, larynx, and trachea down to the level of the thoracic inlet for examination. If a “bloat-line” observation is potentially important in the necropsy, this would be a good time to separate the esophagus from the trachea to the level of the thoracic inlet. Later in the necropsy, when the pluck is reflected over the first rib, the esophagus can be retracted through the thoracic inlet and its entire length can be examined.

**Opening the Abdomen and Thorax**

There are several acceptable ways to gain entry into the abdomen. The author generally starts by incising the abdominal wall along the greater curvature of the last rib, being careful not to incise the intestine. Once a hand-size hole is made, the author reverses the grip on the necropsy knife so the tip of the handle is forward, slides the hand into the abdomen with the knife handle leading the cutting edge, and incises the abdominal wall as the hand is advanced ([Fig. 7](#)). The incision is continued until the abdominal wall can be reflected.

The greater omentum is cut away, revealing the small intestine and allowing the abdominal viscera to shift away from the diaphragm, which is examined and cut free along its costal attachment. Using shears or an axe, cut across the distal ribs close to

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*Fig. 6. The cheek has been incised, exposing the molars and oral cavity for examination.*
the costochondral junctions. The ribs may be separated and manually reflected by breaking the costovertebral joint, or one can cut across the proximal ribs close to the costovertebral joints and reflect the entire plate of ribs forward off the top of the thoracic organs (Figs. 8 and 9). Leave the first rib intact. This will hold the thoracic organs in the carcass as it is winched onto the rendering truck. It is always a good idea to be considerate of both production personnel and those who work for the renderer.

Examining the Thoracic Cavity

First examine the pericardial sac and fluid. Detach the lung by cutting between the thoracic vertebra and aorta. Then dissect the dorsal lung free from the anterior thoracic to the diaphragm (Fig. 10). Next, free the caudal right lung lobe from the diaphragm by cutting the aorta, esophagus, and mediastinal reflections (right and left) between the pericardial sac and diaphragm. Continue detaching the pluck by cutting attachments between the pericardial sac and ventral thoracic. Reflect the lungs and heart forward over the first rib (Fig. 11).

Palpate the lung for abnormalities. Examine the tracheobronchial lymph nodes and airways. Examine the thoracic esophagus. The esophagus can be pulled through the thoracic inlet if a potential bloat line is of interest.

The heart’s pericardium, myocardium, and endocardium are evaluated as the organ is opened. Start the examination with the right heart. Make an incision in the right ventricle just below the vena cava and extend the incision through the semilunar
valves. Extend the incision distally along the border of the right ventricular wall around its entire connection to the septal wall. This flaps the right ventricle and allows an excellent view of the tricuspid valve. To open the left ventricle, make an incision in the middle of the ventricle such that when opened, the 2 large papillary muscles will lay on either side of the incision. Cut across the ventricle just below the coronary groove. This forms a “T”-shaped incision, allowing the bicuspid valves to be examined. The left semilunar valves can be examined by extending the vertical incision into the aorta. These steps are illustrated from left to right in Fig. 12.

Examining the Abdominal Cavity

The small intestines can be fanned out or spread over the rumen for examination (Fig. 13). Autolysis generally makes opening the entire length of the intestine pointless.

Fig. 9. After cutting across the ribs dorsally and the ventral costochondral junctions, reflect the rib plate forward.

Fig. 10. To remove the lung and heart, start by dissecting the lung away from the thoracic vertebra. Continue dissecting the lung free from the diaphragm and the pericardial attachments from the sternum.
However, mesenteric lymph nodes will retain their architecture longer than bowel and are useful in evaluating inflammatory changes. Always examine the ileocecal valve for signs of inflammation as could be associated with salmonellosis.

Although the small intestine is spread over the rumen, examine the right kidney and liver. Flip the small intestine over the transverse processes, exposing the small colon (Fig. 14), left kidney, bladder, distal colon, and rectum (if a female, their reproductive organs) for examination.

Make a palm-size hole in the rumen behind the anterior pillars. Reach in and find the ruminoreticular fold. Pull the fold to surface and examine the side next to the cranial sack for acidosis lesions or scars.

Palpate the abomasum, reticulum, and omasum for masses and normal texture. Reach under the anteroventral edge of the abomasum next to the diaphragm and grasp the spleen. Retract the spleen for examination. Open the abomasum to examine the surface for lesions such as ulcers, parasites, or scarring.

**RECORDING OBSERVATIONS**

The principal purpose of field necropsies is to gain information that may evaluate or influence production management decisions. Necropsy reports are intended to communicate the necropsy observations to others and to serve as a record that

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Fig. 11. Reflect the lung over the first rib. It allows for complete examination of the lung, associated lymph nodes, and heart.

Fig. 12. The sequence represents the recommended steps for examining the heart. This approach allows for thorough examination of all valves and myocardial muscle structure. (A) The knife opens the right ventricle, allowing the incision (B) to extend into the outflow. (C) The right ventricle has been completely opened, allowing examination of the tricuspid valve. (D) Two incisions have been made in the left ventricle. One has been longitudinally and the other across the ventricle below the coronary groove. This exposes the bicuspid valve for examination. (E) The longitudinal incision has been extended through the aorta.
can be used in production management. The brevity that many practice in our reports creates severe deficiencies in communication. There is a better way.

**Necropsy Observations Check-Off Form**

A form can be created that allows rapid highlighting of the circumstance and health management history, noting and checking off body systems involved, highlighting observations within each system examined, and summarizing tentative causes or diagnosis (Fig. 15). This is particularly beneficial when necropsies are performed by trained personnel instead of the veterinarian. A consistent set of digital photographs of each necropsy that includes the *animal's identification tag in each photograph* can be valuable when communicating with the off-site veterinarian, pathologist, or lawyer. The photograph taken may include the surface and opened view of the lung with the tracheobronchial lymph node, heart, kidney, and the small intestine with an associated mesenteric lymph node. A necropsy form for cattle can be divided into 3 parts: history, observations, and cause or diagnosis.

The history portion of the form should include the date, animal identification and description, environmental stress information, and health information. It may or may not include a vaccination history, but this could easily be added.

The observation portion of the form includes each body system and several observations that can be made in each body system. Users will likely be uncomfortable trying to limit their observations to the number available on a form. The quality and

**Fig. 13.** Fan the small intestine over the rumen for examination and cut across the mesenteric lymph nodes.

**Fig. 14.** Flip the small intestine over the transverse processes to expose the small colon and allow access to the distal colon, left kidney, bladder, and reproductive system.
The quantity of information gathered will far exceed what is typically found in most practitioners' brief necropsy reports.

The cause and diagnostic section of the form may include a series of boxes that allows one to numerically rank the observer's opinion of the importance of the body system involved and rank the suspected cause involved. The final portion of the form has a place for comments and suspected diagnosis.

The highlights of each necropsy can be kept in a simple Microsoft Excel spreadsheet or Microsoft Access (Redmond, WA, USA) database. This allows necropsy report forms to be easily searched for relationships between cases and production management decisions.

The form described in a Microsoft Word format can be downloaded from the University of Nebraska – Lincoln, Great Plains Veterinary Educational Center's Internet site (http://GPVEC.UNL.EDU) under “Griffin’s Teaching Files.”

HANDLING, PACKAGING, AND SHIPPING DIAGNOSTIC SPECIMENS

Tissue specimen degradation is a serious issue that can severely handicap a diagnostic laboratory's ability to provide usable information. Histopathology specimens should be sliced thin, less than 7 mm, at the time they are collected. Intestine tissue samples are especially sensitive to crushing damage, and it is important to ensure the 10% formalin (3.7% formaldehyde) is in contact with the intestinal lining. If formalin is not available at the time of necropsy, keep the samples cold and separated. Most pathologists suggest taking samples from all major body systems with associated lymph nodes if changes are noted. More than 1 sample would be appropriate from the body system(s) that exhibited significant pathologic changes, including sections along the boundaries between normal and abnormal tissue.

Tissue samples collected for microbiology pose unique challenges. These specimens generally are not sliced as thin as are specimens for histopathologic examination and therefore are more prone to heat degradation. The bacteria in the specimen, targeted for both culture and contaminants, continue to grow. Contaminant bacteria may replicate faster and/or they may produce substances as they grow that inhibit the replication and subsequent recovery of the targeted bacterium. For this reason, the author frequently collects a needle aspirate of the tissue to culture and collects tissue specimens for laboratory microbiologist.

Needle aspirate collection for microbiology is a simple procedure that allows submission of an inoculated blood agar plate to the laboratory along with the other specimens. Starting cultures in the field improves the turnaround time and can improve the accuracy of diagnosis of some diseases. The author takes aspirates using a 10-mL syringe and a 20-gauge 1.5-inch needle. The author flames the needle using...
a butane lighter until it is red hot, then inserts it into the target tissue to be cultured. The hot needle should sear the surface and prevent contamination of the aspirate from surface contaminants. Aspirate fluid and tissue into the needle and syringe. After necropsy, the author sprays collected aspirates on blood agar plates. Next, bend the 1.5-inch needle in a 45° angle and flame the angle formed until sterile. Use the bent needle to streak the agar plate (Fig. 16). Tape the edges of the agar plates. Double bag each plate, and it is ready for shipping to diagnostic laboratory.

![Fig. 16. Starting bacteriology culture shortly after the necropsy provides an additional opportunity for the diagnostic laboratory to make a better evaluation. In this illustration, a needle aspirate has been inoculated on a blood agar plate and the aspirating needle, after bending and heat sterilization, is used to streak the agar inoculum.](image-url)
Important packaging and shipping definitions

- Biologic substances, category B means any human or animal material being shipped for diagnostic purposes. *These specimens must have both a “Biologic Substance” label and a UN3373 diamond logo (Fig. 17) on the shipping container.* As noted, the “Infectious Substances” designation does not apply to diagnostic specimens that would be shipped to diagnostic laboratories. Appropriate labels are available online or from a state diagnostic laboratory.

- The 10% formalin used for animal diagnostic samples is a volume-per-volume mixture of 1 part 37% formaldehyde added to 9 parts water (with or without additional buffers). This dilution does not meet the US Department of Transportation’s definition for a hazardous material under the Hazardous Materials Regulation; 49 CFR Parts 171–180 and is not regulated. Air transportation requirements additionally must meet the International Air Transport Association requirements. Currently, individual fixed specimen sample containers are kept to less than 30 mL or 1 ounce of 10% formalin; up to 33 of these individual containers, or less than 1 L total formalin, can be shipped by air transportation.

- Regulatory agencies
  - US Department of Health and Human Services regulates the interstate shipment of etiologic agents.
  - The US Department of Transportation regulates ground and air transportation of diagnostic specimens, infectious substances, medical waste, and chemical and radioactive materials (www.hazmat.com).
  - Samples shipped by ground (courier, bus, postal service, etc.) follow the US Department of Transportation Code of Federal Regulations (49CFR).
  - The International Air Transport Association, although not an agency, writes the requirements for all air transportation.

Common carriers include Federal Express (fedex.com) and the US Postal Service (usps.com).

Shipping Good Management Practice

- Use a sturdy reinforced container; Styrofoam (Dow Chemical, Midlan, MI, USA) boxes should be inside a cardboard box.
- Place coolant packs in Ziploc (S.C. Johnson & Son, Racine, WI, USA) bags in case of leakage or rupture.

**Fig. 17.** This is an example of a properly labeled biologic substance “Category B” shipping container that contains dry ice. Note the weight of the dry ice is included on the label.
Avoid overfilling liquid containers, don’t exceed one-half of the container capacity.

- Whirl-Pak (Uline, Pleasant Prairie, WI, USA) bags are superior to Ziploc bags; twist-tie the metal strip after closing.
- Tape all rubber-stoppered tubes.
- Double check for potential leakage of all containers.
- Place all specimens inside a large plastic bag that contains sufficient absorbent (cat litter or paper towels) for all the fluid in the containers to be shipped should they be damaged during shipment.
- Avoid breakable specimen containers ... if used, pad and double bag the container.
- Place paperwork in waterproof bag; Ziploc bags work best.
- If there are shipment questions, contact the carrier and/or laboratory.

Packaging Diagnostic Specimens (Biologic Substances, Category B)

Diagnostic specimens must be triple packed as follows:

- **Primary container** should be a screw cap tube, taped red top blood tube, or Whirl-Pak bag wrapped with the tie ends twisted together. Ziploc bags are not suitable for liquid primary containers.9–12
- **Secondary container** must be water-tight and have sufficient absorbent, such as paper towels, should the primary container leak or rupture.
- **Outer package** (third layer of the specimen shipment container) should be at least as durable as sturdy cardboard. Although Styrofoam is an excellent container; it should never be used as a shipping container without residing in a sturdy cardboard box. Styrofoam coolers are not acceptable as the exclusive outer container because of the potential for rupture if dropped or impacted in a transportation accident.
- USPS limits less than 1 L (1 L) liquid per primary container with total of less than 4 L or less than 4 kg solid per shipment. USPS requires a biohazard logo. Some carriers limit shipment to less than 0.5 L or less than 0.5 kg solids.
- Shipper’s and consignee’s contact information, including 24-hour telephone number, should be on the label (Fig. 17).
- Both a “Biologic Substance” label and a UN3373 diamond logo (see Fig. 16) should be on the outer shipping container.

**BOTTOM LINE**

Diagnostic specimens must be packaged in triple packaging consisting of: (1) a primary container, such as a screw-cap tube or plastic bag; (2) a secondary container that must be watertight and contain sufficient absorbent to capture any leakage; (3) outer packaging that is of sturdy cardboard (do not use Styrofoam coolers as the outer container); and (4) a properly labeled shipping container (see Fig. 17).

Assume that specimens will travel in part by air, so meet International Air Transport Association shipping requirements (eg, 1 L of 10% formalin per properly packaged sample container, or 30 mL per sample container if it includes formaldehyde of >10% concentration). Shipping with dry ice has a few additional requirements (see Fig. 17).12 The outer shipping container must be marked with “Carbon Dioxide, Solid or Dry Ice” and the UN Identification Number “UN1845” and a Class 9 label.

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