

Student Sessions

Moderator: *Chance Armstrong*

People sometimes lie, but lesions seldom do

Addressing field necropsy and submissions for more useful information

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Abstract

The objectives of the presentation are to cover efficient field necropsy techniques, sample collection, and laboratory submission to address clinical situations presented. We will discuss situational evaluation of common infectious, metabolic, and toxic conditions along with lesions (or lack thereof), and how to evaluate clinical presentation along with lesions and laboratory results to generate actionable information. Specific collection of samples from field necropsies and laboratory test requests will be discussed, as well as the interpretive nuances of lab results. We will also cover sample handling and chain of custody for potential legal cases. What we are attempting to create is information that is interpretable and actionable.

Key words: necropsy, diagnostics, laboratory, sample collection

Résumé

L'objectif de cette présentation est de couvrir les techniques efficaces de nécropsie sur le terrain, la cueillette d'échantillon et la soumission au laboratoire dans les différentes situations cliniques qui se présentent. Nous discuterons de l'évaluation selon la situation de conditions infectieuses, métaboliques et toxiques communes et de leurs lésions (le cas échéant). De plus, nous discuterons comment évaluer la présentation clinique de même que les lésions et les résultats de laboratoire pour pouvoir générer de l'information exploitable. La cueillette spécialisée d'échantillon provenant de la nécropsie sur le terrain et les requêtes pour des tests en laboratoire seront aussi abordées de même que l'interprétation nuancée des résultats de laboratoire. Nous allons aussi détailler la manipulation des échantillons et la chaîne de responsabilité dans les cas d'affaires judiciaires. En bout de ligne, nous tentons de créer de l'information qui est interprétable et exploitable.

Introduction

Unfortunately time and space will not permit a comprehensive coverage of diagnostic approaches and techniques

for cattle practice, so I offer metaphylactic apologies to my pathology and diagnostic lab colleagues for what is inevitable incompleteness. I also hope the understanding is clear that geographic differences often dictate what is common and uncommon, so regional expertise in diagnostic patterns is vitally important. My intent is to bridge the gap between diagnostic lab capabilities, practitioner experience and situational observation to help facilitate the necessary communication between practitioners and diagnosticians. I find this particularly useful for veterinary students and new graduate practitioners to help them prioritize the common from the uncommon, and to help them to recognize the uncommon or cases of high consequence early in the investigation process, and to react with the appropriate diagnostic submissions in those instances.

Field necropsies and disease investigations can be approached as a funnel with a wide set of possibilities at the top, tapering down to a smaller number of possibilities with each observational disclosure. There are a limited number of diseases that will affect cattle, and patterns tend to repeat themselves. In short there are precious few truly unique things out there, and the list of usual suspects is far more common and repeatable. With that said pattern recognition is important, but approaching necropsy without bias is important. Let the animal, lesions and environment speak first before jumping to interpretation, and allow the initial integration of history, signalment, environment and lesions guide the subsequent sample submission before arriving at a final conclusion.

In reality initial clinical decisions will need to be made based off of incomplete information, (translation: that means history, signalment, environment and gross lesion) because in death loss or high morbidity situations an educated guess will have to be made regarding treatment or management changes BEFORE even preliminary lab information is available. To me this is where the skill of gross diagnostics and clinical acumen is important, but admittedly incomplete. Reality dictates that when making decisions on incomplete information you will be at least partially incorrect a fair portion of the time, but delaying intervention until one is certain may be even more costly. That does not mean you were wrong, you just adjust subsequent interventions based off the more complete information.

Generally infectious disease leaves some tracks visible on necropsy if you know how and where to look. Toxicities may leave visible lesions as well but do so less predictably. Metabolic diseases and peracute toxicity may leave no lesions or lesions so non-specific as to be useless. These generalities are useful to categorize what you are likely looking at while you are on the ground, and what samples you will likely have to submit. For example there is little need to run fecals and ocular fluid nitrates on 4 head all showing signs of acute fibrinous pneumonia. Likewise there is little need in doing viral respiratory PCR on a dozen head of mature cows dead with no evidence of pneumonia, perhaps that money is better spent on GC Mass Spec of rumen contents. In contrast multiple cattle dead with little to no visible lesions calls for comprehensive submissions to detect a wide array of metabolic, toxic, and infectious diseases. (KSS: Kitchen Sink Submissions).

Suggested Diagnostic Approach

Samples to collect and potential test lab will run

As always the preferred method is to call the lab prior to submission to make sure we have the right samples and are chasing the right things. The following is a generic approach that addresses some of the most common presentations.

Respiratory Disease - lesions diseases and clinical correlates

Any tissue has a limited way that it can react to injury. The lung is basically a capillary network of air spaces connected to the outside environment by a variably expandable tube to conduct air. Movement is created by a negative pressure. Gas exchange is created by diffusion across small spaces between the blood and air space in the alveoli. That seems to be a simplistic review but any disruption in any of those anatomical structures (macroscopic or microscopic, or biochemical) will result in failure of gas exchange. The most common causes of respiratory dysfunction are edema, inflammation, collapse of air spaces, or filling of air spaces by inflammatory exudate or fluid. Less commonly alveolar disease resulting in increased distance from blood to gas exchange interface occur, as will alveolar or airway disease that changes the lungs' ability to expand or contract. Lung disease in cattle is mostly represented by these changes. Those changes may have clinically recognizable consequences or help to establish patterns of death that provide clues to potential causes. For example acute to peracute pneumonias that produce lots of vascular damage, fibrin, and edema will be more likely to cause acute onset of disease or rapid progression to death (often found dead) than etiologic agents that cause slower more indolent pneumonias resulting in less edema and fibrin.

Similarly those agents (toxic and viral) that cause diffuse acute pneumocyte damage can result in what appears clinically as acute onset of profound dyspnea. Said another way disease conditions that cause even moderate acute vas-

cular or pneumocyte damage affecting the entire lung can appear clinically as more severe respiratory disease than severe damage that affects a small portion of the lung.

A simple classification of pneumonias into fibrin rich and fibrin poor pneumonias is useful clinically. Those fibrin rich pneumonias are those that cause significant vascular endothelial damage and result in edema and fibrin accumulation in alveolar spaces and pleural spaces. Mannheimia and Histophilus are the most obvious examples of this. Fibrin poor pneumonias are the Mycoplasmas, and *Pasteurella multocida* falls somewhere in between, as there are some strains of *P multocida* that can be pretty fibrin rich. The differences in lesion manifestations are likely due to their virulence factors and the inflammatory cascade induced by the different agents. Of course mixed infections are common and recent evidence suggests that polymicrobial bacterial infections have more adverse outcomes than monomicrobial infections, and we are all aware that previous or concurrent viral infections (BoHV-1, BVDV, BRSV especially) have substantially diminished treatment response and higher death loss.

Respiratory Disease Lookalikes

Any ancillary conditions that interfere with gas exchange, vascular perfusion of the lung, or expansion or contraction of the lung will present as dyspnea resembling respiratory disease. Profound anemia, cardiac failure, prussic acid, nitrate toxicity, acute pulmonary edema, endotoxemia, anaphylaxis, or hyperthermia can all cause issues with blood gas exchange that could be clinically mistaken for infectious respiratory disease, but in general they leave scant gross lung lesions other than occasionally edema (which can be life-threatening itself).

Respiratory Disease Sample Collection Postmortem

Fresh tissue - lung (marginal zone of pneumonia), trachea

Nice to have - spleen or liver, tracheobronchial lymph node, or colon swab

Tests - lung and/or trachea

- bacterial culture - fresh tissue or transport media.
- viral PCR - on fresh tissue, or swabs from tissue bovine herpes virus 1 (BoHV-1), bovine virus diarrhea virus (BVDV), bovine respiratory syncytial virus (BRSV), parainfluenza virus 3. (Some now including Bovine respiratory corona virus, and potentially Influenza D)
- culture for Salmonella colon swab, Liver (depends on age and disease presentation)
- Fixed tissue - In relative order of importance lung (marginal zone, anterior ventral and dorsal caudal lung; make sure there are some large caliber airways in some sections. Trachea, liver, heart, spleen, ileum, colon, rumen, abomasum, kidney,

skin.

Justification - histology aids in determining other contributory issues to respiratory disease, i.e. hepatopathy, enteric disease, acidosis, cardiac lesions etc. Fixed tissue is also available for immunohistochemistry (IHC) for a variety of agents should that become necessary. Most importantly, histology helps to determine the relevance of agent detection by PCR or other techniques. We struggle to discern the relevance of some agents, like BRCV or *Mycoplasma bovis* when detected on PCR, unless we can demonstrate lesions in the lungs. With the explosion of respiratory viral PCRs, it is becoming more important to relate anatomic lesions to the molecular diagnostic findings. Histology also allows more perspective in staging age of lesion and evaluating other potential contributory causes like viral pneumonia, or pre-existing lesions. These can be important in instructing caretakers on proper timely identification of sick cattle, or in staging lesions where disputes occur relative to onset of disease after purchase of feeder cattle.

Live Cattle - Respiratory Sample Collection

- Deep nasopharyngeal swab (in transport media) and for second sample for PCR (usually Dacron, moistened with saline in red top tube). Transtracheal wash or Bronchoalveolar lavage are the preferred samples. It is important to know what the vaccination status of the cattle are relative to the sampling, as vaccine virus can be detected for as long as 3 to 4 weeks post vaccination, depending on the type of vaccine administered.
- Tests run -
 - Standard culture.
 - Multiplex viral BoHV-1, BRSV, BVDV, PI3, Bovine Respiratory Corona Virus (BRCV) and bacterial PCR (including *Mycoplasma bovis* in most cases. Now more and more PCRs for bacterial agents are available. Again this becomes an interpretive exercise to interpret significance of *Histophilus*, *Pasteurella multocida*, *M. hemolytica* from nasal swabs, and their relationship to diseases. The presence of bacterial agents in BAL or TTW is probably a bit more relevant.

Enteric Disease

Enteric disease can leave either dramatic gross and microscopic lesions or have virtually unrecognizable changes. That again is due to the agent itself and how it caused disease. Many of the enteric agents, especially in neonates, cause subtle changes in the intestinal mucosa that are not very impressive grossly and often similarly subtle microscopi-

cally. Those agents often cause mechanistic changes in gut physiology resulting in diarrhea with scant lesions. In more severe cases, those agents that cause direct mucosal damage or vascular damage leave more visible lesions (salmonella, coccidiosis). The clostridial diseases usually cause disease by elaboration of potent exotoxins which may cause visible hemorrhagic lesions in the affected gut, or may be more active systemically. From a practitioner standpoint, neonatal diarrhea investigation becomes more of a tissue and sample harvesting process, and the ultimate diagnosis comes from comprehensive analysis of the histology and ancillary diagnostic tests. From a practical standpoint most will be addressed with the same management interventions

Intestinal lesions are often misinterpreted. Profound capillary congestion occurs at death in many animals without enteric disease. Grossly this is often misinterpreted as hemorrhagic enteritis, but histologically it is just pooling of blood in intestinal capillaries. There are plenty of capillaries there and they can pool an astonishing amount of blood. After death mucus continues to be released from the goblet cells. Enterocytes slough too after death. Grossly this may look like a purulent exudate. It is nothing but us usually misinterpreted as enteritis, especially if it occurs with the above congestion.

If ingesta is adherent to the mucosal surface be suspicious of a lesion there. Typically ingesta washes off, if it sticks there is probably mucosal ulceration and/or fibrin on the mucosal surface. That is a great place to take a sample. The ileum or ileocecal junction is one part that needs to be collected for culture, FA, histology etc. It is one of the most immunologically active portions of the gut, so it makes sense to take that regardless.

Neonates - very young calves may produce very subtle lesions in most organs, especially in the digestive tract. I tend to encourage practitioners to collect a complete set of tissue in neonatal cases, as lung and liver lesions are often missed by gross examination alone, and intestinal lesions are often misinterpreted. Intestine should be open for histology, (ileum, jejunum, cecum and colon). Scraping the mucosal surface to remove ingesta basically ruins the sample.

Postmortem samples - neonatal diarrhea

- Fresh tissues- Intestine (Ileum, jejunum or any section with suspected lesions), Cecum and Colon intestinal contents (refrigerate or freeze as soon as possible). Mesenteric Lymph node (ileal or from area of suspected gross lesions), liver, spleen.
- Serum from live presented calves. Tests available-bacterial culture, PCR enteric viruses Corona Virus, Rotavirus(s), PCR for virulence genes of E. coli or Clostridium. Clostridium toxin detection on intestinal contents. Colon content smears for cryptosporidium, BVD PCR or IHC. Total protein on serum.
- Fixed tissues- Intestine (ileum, jejunum), cecum, co-

lon, lung, heart, liver, spleen, abomasum, mesenteric lymph node, umbilicus, synovial tissue (usually cut out knee cap or cut into tibiotarsal joint) Skin.

- Evaluate bone marrow grossly for evidence of serous atrophy of fat

Older cattle - mainly a BVD corona virus and/or salmonella. In adult cattle Johnes' feces

- Feces for Culture, PCR. Necropsy fresh and fixed ileum, colon, and caecum along with complete set of tissues. Usually run acid fast stain on ileum and colon for Johnes, although gross lesions can be very predictive. In small ruminants mesenteric lymph node is often needed for Johnes' diagnosis.

Abortion Diagnostics - It is important to consider that cattle are single birth species, and any metabolic or genetic abnormality that is incompatible with life will result in an abortion, and usually without gross or microscopic lesions. As well cows that have uterine insufficiency will lose pregnancies when there are insufficient placentomes to support the pregnancy to term. Usually those types of abortions will occur without fetal lesions or subtle non-specific lesions. Cows will abort from systemic disease unrelated to the fetus, and those will occur without specific fetal lesions generally. If an abortion occurs due to a demonstrable infectious cause it usually incubates in a 101F, 100% humidity incubator for hours to days, resulting in substantial post mortem changes. Finally, usually the diagnosis is made off of placenta, which unfortunately gets into the hands of the diagnostician a fraction of the time. So based on those disclaimers, often the best we can do is identify a limited number of agents that we have good diagnostic tests for, and as a second tier we might be able to determine if pregnancy loss is due to an infectious agent or not. For those reasons simply the ability to differentiate between herd problems and individual abortions is important. The list of abortion causes for individual abortions is nearly endless, because so many opportunistic bacteria and fungi can cause pregnancy loss. The list of those causing herd problems is more manageable, and generally is more discoverable when multiple submissions are made available. The important distinction is that with those herd related reproductive problems, management and control decisions are made specific to each identified pathogen, so accurate identification of which infectious agents are present is relevant to the intervention.

Abortion diagnostics is again largely a tissue harvesting process. While pictures of gross lesion in fetuses exist, they are not that common in practice. About the best you can hope for are either skin lesions that may be present in fungal abortions, or placental lesions that are present in a variety of bacterial or viral placentitis. They are often similar in multiple etiologic agents, and thus require careful tissue collection for diagnosis. Identification of agents usually occurs through culture, PCR, Histology or Immunohistochemistry.

Fresh Tissues

- Placenta, lung, liver, kidney, abomasal contents, skin, spleen. Ocular fluid, EDTA blood from dam, serum from dam, thoracic fluid, uterine fluid from aborted cows.

Test likely to be run - Lepto FA and/or PCR. BVD, IBR PCR or Immunohistochemistry, Culture (bacterial and fungal culture of abomasal contents or placenta, lung liver), Neospora PCR or IHC on liver, heart brain, ocular fluid nitrates. Anaplasma smear or PCR on EDTA blood, Brucella serology on serum. Serology may be run on thoracic fluid for BVD, Leptospirosis, and Blue tongue. PCR for Tritrichomonas on uterine fluid.

- Fixed tissues - placenta, heart, lung (anterior ventral), liver, kidney, spleen, brain, skin, lymph node, eyelid (including conjunctiva). Some histologic lesions are specific for infectious agents, like BoHV-1 abortions with necrotizing lesions and inclusions. Other less specific lesions give hints but no specific etiologies. Fetal pneumonias can be seen with a variety of bacterial or fungal placentitis, fetal enteritis with others. Fetal lymphoid hyperplasia evidences that some infectious agent has been delivered to the fetus and clues the diagnostician into a heightened possibility of an infectious event. Unfortunately sometimes the best we can do is categorize abortions into an infectious event or not, and hope future submissions get us closer to the offending cause(s). Important considerations- Brain and heart for Neospora. Complete set of tissues helps to ascertain if there is an infectious component to the abortion.

CNS Disease - of all the conditions covered CNS disease is the most unlikely to have any visible gross lesions. Occasionally one might be able to discern a faint opacity to the meninges with encephalitis, or subjectively more fluid in the submeningeal spaces, and of course the lesions of well-developed polioencephalomalacia are obvious, but they are often subtle enough in the early stages to be missed grossly. Infectious thrombotic meningoencephalitis (TME) when obvious is an easy gross diagnosis, but there are varying stages of disease and it too can be missed grossly in milder events. Because brain lesions are subtle they usually become a microscopic or toxicological diagnosis. Clinical history is important in pattern recognition diagnosis. In Rabies endemic areas half the brain needs to be preserved fresh for rabies examination.

Important anatomical sites need to be considered in CNS diagnostics. With Polioencephalomalacia the lesions tend to begin in the caudal 1/3 of the cerebrum along the dorsolateral aspect of the cerebral cortex, so this is the preferred site to select for histology. Listeria tends to cause microabscesses in the brain stem below the cerebellum, so that is a preferred site for culture or histology. Rabies inclusions tend to be more prominent in the cerebellum of

herbivores, especially cattle, so that is a site along with brain stem that needs to go for rabies examination and in formalin for histology. The whole brain or a whole sagittal half is preferable as the pathologist needs to be able to select the appropriate anatomical site for a variety of potential causes. Of course brain stem (specifically Obex) is the preferred site for Spongiform encephalopathy. There are plenty of poorly characterized viral encephalitis of cattle which are usually sorted out by PCR, Immunohistochemistry, or remain obscure and diagnosed as non-suppurative encephalitis. Lead and sulfates are a cause of polioencephalomalacia in cattle, so it is prudent to submit liver, and kidney for lead levels, and feed and water for sulfate levels with polio cases. Don't forget brain sodium levels for water deprivation.

Common causes of CNS disturbances

- Polioencephalomalacia- sulfates, thiamine deficiency or agonists, lead
- Viral encephalitis- Herpes virus, rabies, plus a variety of more obscure non-suppurative encephalitis
- Bacterial- Histophilus, Listeria, Other septicemias (including E. coli in neonates)
Toxic- lead, pesticides, paspalum, sulfate, regional forage toxicoses

Fresh tissue to collect -

Brain, EDTA blood (if alive) Serum, Urine, Kidney, Liver, Rumen contents ocular fluid. Feed Sulfate levels.

Test run- rabies, tox screen, bacterial culture, viral and bacterial PCR, cholinesterase inhibitors

Fixed tissue- Brain (complete brainstem, save appropriate section for rabies, fresh), and sagittal section of rest of brain), heart, lung, liver, kidney, spleen, colon, cecum ileum.

Liver - the liver has a great deal of variation in color, and consistency, depending on the amount of perfusion at the time of death, the amount of fat, or glycogen, or the amount of depletion of either of those. I caution veterinarians about interpretation of liver color and consistency unless it is obvious. It is always best to rely on histology for evaluation of liver changes. I have often been surprised at the level of microscopic changes in a liver that appeared grossly normal.

Outside of toxicity there are not many true liver diseases in range cattle, and those hepatotoxicities are often generic in their changes, thus it is important to have toxicological confirmation of the agent. As much as anything liver lesions are usually supportive of other systemic diagnoses. Some of the more common examples I have come across are feed related sub-clinical mycotoxicoses that contribute to other infectious diseases or performance losses, and septicemic salmonellosis complicating respiratory disease treatments.

If toxicity is suspected I recommend fresh (or frozen) liver coupled with rumen contents, fresh kidney, and of course liver for histology. Toxicologists will differ on how much is

an adequate sample, but for brevity sake a fist sized sample is usually adequate, if you want to be detailed 50 grams should be fine.

Kidney - the kidney is not a tissue we usually see a lot of lesions in on necropsies. Other than the direct nephrotoxins (oak, pigweed, etc.) we do not see much in the way of kidney changes. The kidney does catch a significant percentage of blood flow from the heart, so when there is evidence of septicemia we can see acute or chronic septic infarcts in the kidney. Drug induced toxicities can be detected microscopically as tubular damage, but in my experience they are rarely dramatic grossly. The kidney is another organ that seems to have a lot of variation in color and consistency dependent on post-mortem interval. Best to confirm subtle changes by microscopy.

Urinary obstruction (urolithiasis) is common and renal changes there can be pretty impressive, and lead you to examine patency of the distal urinary tract. Kidney is a good organ for confirmation of suspected lead toxicity

Spleen - the spleen can be a difficult organ to interpret grossly. Basically it is a cavernous channel of venous sinusoids filled with macrophages and lymphoid structures. The spleen can vary substantially in size based on venous pooling or whether the spleen has reacted over time to bacteremia. I try not to over interpret spleen changes. There seems to be a wide range of normal in spleen size and venous pooling, and cattle dying from septicemia/toxemia will have some splenic enlargement. On the polar ends of the spectrum the spleen can be meaty and big or bloody and big. A meaty spleen indicates increased density of phagocytic cells which is a response to a circulating antigen inducing that change. We see that more in chronic conditions. Large blood spleens can be a consequence of increased pooling of blood from septicemia/toxemia, but it can also be coupled with Reticulo-endothelial hyperplasia so now you have both a meaty and congested spleen. Again this is a great opportunity for histology to clear up the picture. Since the spleen is essentially the oil filter of the blood, coupled with a large lymphoid component it is one of the best organs to culture for bacteremic or viremic diseases.

Splenomegaly is pretty dramatic with Anaplasmosis and Anthrax. Generally cattle with Anaplasmosis are icteric and have thin watery blood. They may have few if any blood clots which may raise awareness of anthrax which also often has impressive splenomegaly. Anthrax cattle usually show more evidence of sepsis, but some anaplasma infected cattle die with agonal death causing petechial hemorrhages as well so the differentiation between the two may not be obvious grossly in the field. Some cases of Clostridial toxemias will show noticeable splenomegaly as well.

Acute death (with or without observable lesions)

First it is important to determine if it really is an acute

death, or if it was a death that occurred during a 3-4 day interval when cattle were not observed. Like abortions it is important to determine if this is an individual animal or a herd problem.

In true acute deaths, the list of causes is a manageable list of rule outs. Oxygen delivery (Nitrates, Prussic acid), Clostridial toxins, Anthrax, and occasionally Botulism, and a limited number of other toxins are potent enough to kill cattle in a matter of hours without any clinical signs. Metabolic derangements such as Ca and Mg can also present as acute deaths. A few infectious diseases fall into that category, but more often we see infectious disease appearing as acute deaths when in reality they are cattle that are observed infrequently. When the death losses occur cattle are observed more closely and we can generally begin to pick up clinical signs before cattle are just found dead. (Anaplasmosis, Mannheimia, are two of the more common causes I have encountered in this category).

Man-made toxins are an entirely different category and they becomes it becomes a toxicologic exercise to determine if this is true accidental exposure, or inadvertent overdose of a product intended to be administered to cattle. That is where identification of the toxin and semi-quantitation becomes valuable.

- Fresh tissues for suspect toxicities. Brain, lung, liver, kidney, spleen, rumen contents, intestinal contents (refrigerate or freeze expediently) urine, ocular fluid (aqueous preferably), Cerebrospinal fluid, Rumen contents, feed, water. Blood smear, blood.
- Fixed tissues for suspect toxicities- Brain, heart, lung, liver, kidney, spleen, rumen abomasum, intestine, colon, skeletal muscle (weight bearing, diaphragm, intercostal), and bone marrow.

It is important to remember that some toxicities are best determined by feed in lieu of rumen contents and vice versa. Nitrates and ionophores are soluble enough in solution that rumen contents may not give an accurately reflective level of what was fed. It is always a good idea to talk to a diagnostic toxicologist to make sure the proper samples are collected for the proper assays.

Similarly in anthrax endemic areas be aware of anthrax submission protocols.

Tests run- Nitrates (ocular fluid), anaplasma blood smear, anthrax-blood smear or gauze, Tox screen, rumen plant ID, Ionophore screen, Blue green algae screen, GC Mass Spec or other suitable tox screen for common compounds. Botulism on frozen intestinal contents. Culture of spleen, liver, lung etc. to rule out unrecognized septicemia or other acute bacterial diseases

Can also run Ca, K, Mg, Nitrates and urea on ocular contents, standard levels may vary from serum levels so clear communication on estimated post mortem levels is important.

- Fixed tissue- of course depending on lesions observed- Heart, lung, liver, kidney, spleen, skeletal muscle (diaphragm intercostals and weight bearing), brain, intestine, rumen, and abomasum.

Other things to note- ambient temperature, lightning strike, condition of the ground around the animal (did the animal struggle?), water availability, amount of autolysis and or bloating.

Chain of Custody

At some point you will probably be involved with a diagnostic event that is contentious or has some potential for legal consequences. Diagnostic Laboratory personnel are aware of this and have procedures in place for documentation that is adequate for most legal cases, but it is always valuable for them to know about potential litigation early in the process as it may have consequences for sample retention beyond their normal retention time. At times it is necessary for the practitioner to document samples collected and submitted in a manner that would withstand scrutiny from an attorney for the defense. In most cases samples collected routinely from necropsies will be fine, but proper identification of animal(s) ID, site of death, photographs, and description of findings is important. In potential feed related toxicities, feed or commodity samples need to be collected, dated, sealed and initialed by the collector. It may be wise to have a third party collect feed of mineral samples and sign date and seal those samples, depending on the gravity of the case. If you know from the outset this has legal implications instruct the Diagnostic laboratory to record the seal and integrity of the seals of the submissions on the accession form and in subsequent reports.

Conclusions

Diagnostic submissions do not need to be a mystery. Communication with your chosen diagnostic laboratory or diagnostician helps to focus efforts and helps to disperse the mystery of test availability and test interpretations. It is important to understand what common rule outs are, how those agents cause disease, and what organs are the best to sample to detect the common diseases. Understand the diagnostic tests are designed to find infectious agents, and simply finding an agent that is a commensal or variable pathogen does not necessarily ascribe causation. We also have to acknowledge that part of the equation of causing clinical disease is failure of the host animal to effectively deal with the pathogen. Usually that is a consequence of naiveté or overwhelming exposure, but occasionally it is a result of comorbid conditions that make the animal more susceptible to disease. Simply detection of the same list of suspect infectious agents by more and more refined techniques does not advance our understanding of predisposing conditions leading to clinical disease.

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