



The primary mission of the Kansas State Veterinary Diagnostic Laboratory (KSVDL) is to develop and deliver accurate, innovative and timely diagnostic and consultative services to the veterinary and animal health community in Kansas and the nation. The KSVDL is a full-service, AAVLD-accredited laboratory, offering a complete range of diagnostic services for all species.

# Diagnostic INSIGHTS

Volume 2, Number 1



Winter 2008

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**Visit our website at:**

Web: <http://www.ksvdl.org>

Diagnostic Insights welcomes your suggestions for future articles or comments about current articles.

Send your ideas to Barbara Barkdoll at [bbarkdol@vet.k-state.edu](mailto:bbarkdol@vet.k-state.edu).



Janice Muller, Medical Technologist, Clinical Pathology Lab



Gary Anderson, DVM, MS, PhD

Presidents Day, Columbus Day and Veterans Day.

I want to take this opportunity to wish you and yours the very best throughout 2008. We have greatly appreciated your business and intend to do our very best in partnering with you. Our goal is to serve you and your clients in ways that will enhance your practice in the coming year. Please do not hesitate to contact me with your ideas, questions or concerns. I can be reached at 785-532-4454 or [ganders@vet.k-state.edu](mailto:ganders@vet.k-state.edu). Again, we appreciate your business!

We have recently made some minor adjustments to our histopathology fees. This includes necropsies performed by pathologists on-site and by our clients in the field (necropsy-in-a-bottle), and we also initiated minimal animal disposal fees.

When a necropsy is performed on a **single animal**, the charges are \$40 for 1-4 tissues and \$50 for five or more tissues submitted. **Multiple animals submitted as one accession** will be charged \$50 for up to three animals. The "multiple animal policy" applies to disease outbreaks in groups of animals suspected of having the same problem (group of neonates with diarrhea, litter of aborted fetuses, poultry die-off, etc.). This policy will apply whether or not animals have been identified separately on the submission form.

Disposal fees have been initiated because of the increasing costs we incur disposing of carcasses, animal waste and hazardous material. The charges apply to all species and are based on weight: <50 lbs is \$5, 50-100 lbs is \$10, and >100 lbs is \$20. The charge is \$0.30 per lb when the alkaline digester is used for disposal.

The KSVDL holiday schedule this year will be similar to last year. We generally follow the Federal holiday schedule, but there are a couple differences. The KSVDL is closed New Years Day, Martin Luther King birthday, Memorial Day, Independence Day, Labor Day, Thanksgiving and the day after, and Christmas, but we are open

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## Poisonous Plants – A New Web ID Service

by Deon van der Merwe, BVSc, PhD, Assistant Professor, Toxicology Department



As part of our ongoing efforts to make KSVDL a center of excellence for the diagnosis of plant poisoning in the region, we have added a web application to compliment our macroscopic poisonous plant identification service, GI tract content microscopy service and forage assays for cyanogenic glycosides and nitrates. The web application is aimed at assisting veterinarians, producers and others to create a short list of plants to consider or to look for in the field when plant poisoning is suspected, based on where the problem occurred and the major organ system affected. A link to the web application can be found on the KSVDL home page ([www.ksvdl.org](http://www.ksvdl.org)) or it can be accessed directly by going to [www.ksvdl.org/toxicology/ppk.htm](http://www.ksvdl.org/toxicology/ppk.htm). While

this is currently targeted for Kansas, it will have value for the surrounding Region as well, since many of the plants are in other areas. The website facilitates a link to USDA resources to additional distribution and information.

If you have any questions please contact Dr. Deon van der Merwe at the Toxicology Section of the KSVDL by phone (785 532 4333) or email ([dmerwe@vet.ksu.edu](mailto:dmerwe@vet.ksu.edu)). I also welcome your comments on how to make this service even more valuable to you as practitioners.



## K-State College of Veterinary Medicine - Continuing Education Opportunities

- On-Line Continuing Education - K-State VetBytes Seminars - 24/7**
- February 10, 2008 - 16th Annual Small Animal Conference on Clinical Hematology and Hemostasis**
- February 23-24, 2008 - Artificial Insemination Course for Horse Owners**
- March 1, 2008 - Veterinary Technicians Conference**
- March 1-2, 2008 - Equine Reproduction Conference for Veterinarians**
- March 8, 2008 - Advanced Conference for Horse Owners**
- April 6, 2008 - 25th Annual Frank W Jordan Seminar on Fielding a Winning Team**
- May 28, 29, and 30, 2008 - International Symposium on Beef Cattle Welfare**
- June 1-4, 2008 - 70th Annual Conference for Veterinarians and KVMA Veterinary Trade Show**

For more information on these conferences, contact:

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Visit our website: [www.vet.k-state.edu/CE/index.htm](http://www.vet.k-state.edu/CE/index.htm)





## Diarrhea Outbreaks in Group-Housed Puppies: Problem Solving Basics

by William Fortney, DVM, Assistant Professor

Diarrhea is one of the most common and potentially serious problems encountered in group-housed puppies (kennels, pet shops, animal shelters, boarding kennels, obedience classes, doggie day care facilities and foster homes). Fortunately, most cases of diarrhea are either self-limiting or respond favorably to empirical therapies; therefore, the definitive cause is seldom confirmed nor even investigated.

The most common etiology of diarrhea outbreaks in puppies involves one or more infectious agents. However, a suppressed immunity from poor nutrition or environmental stressors such as overcrowding or the stress associated with weaning or shipping acts synergistically with an agent to increase the morbidity and/or mortality within the facility. Inadequate cleaning/ disinfection protocols combined with poor disease outbreak containment practices will facilitate the dissemination within the facility.

At the Kansas State Veterinary Diagnostic Laboratory it is not uncommon to find more than one “infectious” agent involved in the pathogenesis of a group-housed puppy presented for diarrhea. In addition to the various strains of *Canine parvovirus 2 virus* (CPV 2a, 2b, 2c.), the agents commonly found in group-housed puppies in Kansas include *Canine distemper virus*, *Canine adenovirus 1*, *Canine coronavirus*, coccidiosis, hookworms, *Giardia sp.*, enterotoxigenic *Clostridium perfringens*, *Clostridium difficile*, entero-pathogenic *Escherichia coli* and occasionally *Salmonella sp.*

While the KSVDL has the expertise and the diagnostic capabilities to identify the specific infectious agents associated in cases of puppy diarrhea, those contributory environmental and/or poor management practices cannot be detected by any laboratory methodology. Therefore, it goes without saying, the key to solving / managing diarrhea in group-housed situations begins with a thorough knowledge of the facility, including their management practices, combined with a complete diagnostic work up.

### 6 Steps to Success

1. There is no substitute for an on-site visit to the facility (animal shelter, kennel, pet shop, etc.).
2. Always investigate for possible immune system stressors.
3. Review anthelmintic and vaccination protocols.
4. Carefully evaluate the disinfection / sanitation and sick puppy

**segregation protocols.** Infected fomites (boots, cleaning carts, hoses, etc.) are common causes of disease spread throughout the facility

5. **Since endemic problems (diseases firmly established in the kennel) are difficult if not impossible to eradicate, shifting to “control” measures is a prudent practice.** *Canine parvovirus 2 virus* (CPV2), hookworms, *Giardia sp.*, and coccidia oocysts are very environmentally resistant. Asymptomatic carriers often act as the continuous or periodic sources of CPV2, *Giardia sp.*, coccidia, *Clostridium sp.*, and *E. coli* to susceptible puppies.
6. **The proper diagnostic sample and the sample submission are usually critical in confirming the exact agent(s) involved in the outbreak.**

The best possible diagnostic outcome begins with the selection and submission of the best possible representative sample(s). Communication with KSVDL laboratory personnel in advance, regarding appropriate sample collection, proper packaging and shipping methods, can be critical in arriving at a diagnosis which will most benefit the owner.

The “best” sample(s) to submit for diagnostic evaluation will vary with the particular intestinal disease(s) suspected, transit time to the laboratory, and the level of interest (financial commitment) on the owner’s part. Diagnostic specimens should be collected from:

1. **Acutely affected animals**
2. **Animals with symptoms characteristic of the problem**
3. **Animals that have not been treated with antibiotics which may inhibit bacterial cultures**
4. **All of the samples submitted as fresh as possible**

In most circumstances, where mortality is commonplace or where a specific diagnosis is essential in the disease management strategy, submission of a live puppy / puppies would be considered the best possible sample for histopathology; plus would provide sufficient fresh feces for parasite evaluation, bacterial cultures, molecular diagnostics, and/or electron microscopy. In other situations where sacrificing a puppy is not an option, then submission of fecal culture swab, and/or 10+ grams of fresh feces, and/or sera may provide adequate diagnostic sampling for the problem. For a complete fecal parasitic evaluation including examination for *Giardia*, a minimum of 5 grams of fresh feces (shipped on ice) is required.

## Bovine Abortions - Achieving Better Diagnostic Results

by Casey Hackett, DVM and Jerome Nietfeld, DVM, PhD, DACVP

Abortions can be a significant loss of revenue for cattle producers. Finding the cause of abortions can be frustrating for the client, practitioner and diagnostician with less than 35% of cases submitted to diagnostic laboratories diagnosed. Of the abortions that are diagnosed, the cause is usually an infectious agent(s).

### Submission of specimens

The quality of specimens received by the diagnostic lab directly affects the results of the laboratory examination. Because submission requirements vary between labs, call KSVDL prior to shipment of specimens to find out what is required and/or recommended. Every effort must be made to prevent leakage of blood, fluids or water from the package.

### Two options are available:

1. Submit the entire fetus and part of the placenta (cotyledon + intercotyledonary areas) to KSVDL (preferred method) with a detailed history. Keep cool, not frozen.
2. Perform a detailed necropsy examination on the aborted fetus and send fresh specimens on ice, but not frozen, to KSVDL with a detailed history and gross findings.

### Abortion Specimens

In Separate Whirl Packs (Fresh tissue on ice)	Bacteriology	Lung, liver, and any affected organs
	Fluorescent Antibody	Lung, kidney and placenta
	Virus Isolation	Pooled lung, kidney, heart & spleen. Placenta in separate bag
	Toxicology	Entire eye or thoracic fluid
	Mycology	Placenta
	Ureaplasma	Lung, Placenta in separate bag
In Separate Red Top Tubes (Fresh on ice)	Bacteriology & Mycology	Abomasal contents, placenta in separate bag
	Serology	Fetal blood from heart or thoracic fluid
In Formalin	Histopathology	Brain, heart, lung, liver, spleen, kidney, skeletal muscle, thyroid, placenta and any affected organs
Fresh tissue on ice	Chlamydia	Cotyledon (including impression smear) and liver



## Haemophilus parasuis in Swine

by Steve Henry, DVM, Diplomate APVP, Swine Practitioner/Consultant

*Haemophilus parasuis* has emerged as one of the significant bacterial diseases of swine and can be a “proving ground” for swine clinicians. When the diagnosis is made and intervention is timely, success occurs often. When the early stages of illness are missed or the incorrect diagnosis is made, many pigs die and more are permanently damaged. Because this disease has so many appearances and presentations, an astute and careful observation technique for every case is needed; less effort or diligence and *H. parasuis* will make a fool of the clinician. Critical points to have in mind in the clinical approach are:

- Always expect *H. parasuis* to make some appearance in newly weaned pigs, those less than 14 days post weaning, if any viral respiratory pathogens are active.
- Remember always the “five blind alleys” rule for serositis – meninges, pericardium, pleura, peritoneum and synovium. If any one or any combination of these tissues is not examined, *H. parasuis* may be missed.
- Know that the most common expression of *H. parasuis* is in the immediate post-weaning phase, but be alert to older animal outbreaks.
- Identification of likely *H. parasuis* outbreaks calls for immediate and vigorous intervention. This is an emergency situation and antibiotic needs to be on board for all in the group – timing is critical.
- Water or feed-based prophylactic therapies are effective only to the point of the first clinical case. Oral tetracycline or potassium penicillin may be most effective when begun at weaning and continued for 5-7 days post-weaning in afflicted, endemic herds. The cost of this approach needs to be weighed against the parenteral intervention at first clinical signs.

- Often penicillin or tetracycline cost only 1/10th as much as newer products; this is important leverage of good diagnostic information toward the benefit of pigs and owners.
- Diagnostic diligence can produce both a best therapeutic outcome and economic outcome.
- Vaccines may work but my experience has been that this effort is a waste of effort and money. Lesions of *H. parasuis* infections are diverse, yet they are recognizable and logically understandable within the clinical setting with a bit of observation and thought.

Generally, the diagnosis depends on the history, clinical signs and necropsy. Bacterial isolation for confirmation is not always successful as the organism has fastidious growth requirements and is fragile. Enhanced diagnostic techniques such as PCR methodology circumvent this problem, and proper transport swabs in Amies media greatly improve recovery of the organism. Recent understanding of the heterogeneity among *H. parasuis* strains has begun to explain the variances in virulence. Call the KSVDL 866-512-5650, for specific assistance in efforts to diagnose polyserositis in pigs – other agents may also be causative.



## Diagnosis of Neonatal Diarrhea in Calves

by Jerome Nietfeld, DVM, PhD, DACVP

Soon it will be calving season and with it that age-old question, what are the best samples to determine the cause(s)? An entire calf for necropsy is a good specimen, but feces from acutely affected calves are almost as good. In fact, feces from an acutely affected calf are better than a whole calf that has been scouring for a week, because peak shedding of most agents begins to decrease after 2-3 days of illness. In the early 1990s, Dr. David Zeman, of South Dakota State University, looked at over 1,400 cases of calf diarrhea and found that the diagnostic success rate was 78.5% when an entire calf was submitted. When only feces were submitted the success rate was 71.5% and when the calf was necropsied in the field and tissues shipped to the lab the success rate was 67.1%.

### Summary of Samples to Submit

#### Antemortem:

- 10 to 20 ml of fresh feces. Refrigerate and ship on ice. Can be used for bacterial culture, ELISA for rotavirus and coronavirus, and parasitological examination.

- Pool feces from several animals to increase the likelihood of success without increasing costs.

#### Postmortem:

- 10 to 20 ml of feces from the distal large intestine.
- Fresh small intestine and spiral colon, mesenteric lymph nodes, and liver. Fresh, refrigerated tissue is critical.
- Formalin-fixed small intestine, spiral colon, lymph nodes, lung, kidney, and liver for histopathology (if it is desirable to hold costs down, fixed tissues are the first thing to omit for diarrhea cases).
- *Do not randomly collect intestines for submission.* Your success rate will increase if you collect fresh samples from the distal one-third of the small intestine and the colon. Bacteria, such as *Escherichia coli* and *Salmonella*, do not like the acid pH in the duodenum and the best sample for coronavirus is the spiral colon. For histopathology, multiple sections of small and large intestine are much better than one, including abnormal tissues and adjoining lymph nodes.

## EIA (Coggins) Testing

In an attempt to meet needs of all clients, to capture laboratory order/efficiency, and to maintain quality results, the KSVDL Serology Lab has established the following sample submission guidelines for equine infectious anemia testing:

#### ELISA Tests:

- Samples will be tested at 2:00 PM Monday-Friday and will be resulted by 3:30 the same day
- Samples received after 2:00 PM will be resulted by 3:30 the next business day

- Individual testing may be run priority Monday-Friday for an extra \$20 per animal and results will be available within 2 hours. Samples must be received before 2:00 PM.

#### AGID Tests:

- Samples received by 2:00 PM Monday-Friday will be resulted by 3:30 the following day
- Samples received after 2:00 PM Monday-Friday will be set up the following day and resulted 24 hours later

We hope these guidelines will be helpful as we endeavor to meet client needs and expectations.