Summer 1997

FROM THE ASSISTANT DIRECTOR

The North Central Conference of Veterinary Laboratory Diagnosticians was held at Purdue University on June 10-11, 1997. We had representatives from 10 states in the Midwestern United States attend this meeting which included pathologists, virologists, bacteriologists and toxicologists working in diagnostic laboratories. Dr. Max Rodibaugh began the session by giving a talk entitled *Are Swine Practitioners "Hog Wild" About Your Services?* Dr. Rodibaugh was able to provide his insight regarding the future direction of swine production as well as veterinary services and diagnostic laboratories. His talk was well received. A total of 26 scientific presentations were given by faculty, staff and graduate students from these diagnostic laboratories. We are especially proud of Dr. Cindy Fishman, a graduate student beginning her third year of residency training here in the Animal Disease Diagnostic Laboratory, who was the winner of the Graduate Student Award. Congratulations Cindy!

Since this meeting was filled with new and exciting presentations regarding animal diseases and diagnostics, we have enclosed some abstracts. We always receive a large number of phone calls regarding aquaculture cases during this time of year, so please note the two articles regarding aquaculture. We have also received several questions regarding rabies over the last few weeks, so please check out the article entitled *Rabies Update*. Finally, the Quarterly Bacteriology Organism Occurrence Report is absent from this newsletter, but look for this report in a more condensed format in our next issue.

Rabies Update

Ohio has reported an increase in rabid raccoons in the past few months. Mahoning County, in northeastern Ohio, has reported 26 rabies positive raccoons, 1 positive cat and 1 positive bat. The issue of exposure of rabies from wildlife animals was significantly elevated when a 3 year old child, who was riding his tricycle in his parents' driveway, was bitten by a rabies positive raccoon in Mahoning County. Therefore, Ohio has launched a plan to deal with raccoon rabies by the use of an oral baiting/vaccine program, a public awareness campaign and a quarantine/ vaccination order for dogs and cats in Mahoning, Columbiana, Trumbull and Ashtabula Counties. It is postulated that this increase in raccoon rabies is due to the spillover of rabies in raccoons in Pennsylvania, since it is known that raccoon rabies has moved north and eastward beginning in the Northeastern region of the United States at rates of 25-50 miles per year.

A total of 2,151 animals in Indiana were tested for rabies at the Indiana State Department of Health Disease Control Laboratory in 1996. Dogs were the most frequently tested species with 750 (34.9%) submissions, followed by cats, 722 (33.6%); bats, 204 (9.5%); and raccoons, 169 (7.9%). The remaining 306 (14.2%) animals included 32 different animal species. In 1996, there were 8 bats positive for rabies and one skunk in Indiana. To date, in 1997, there has been 1 bat positive for rabies and 4 skunks.

Realizing the significant public health risk associated with this disease, as well as the current rabies situation in Ohio, Dr. Bret Marsh, Indiana State Veterinarian has assembled a Rabies Task Force. The purpose of this task force is to ultimately develop a plan of action for controlling rabies in Indiana. Members of this task force include individuals from the Animal Disease Diagnostic Laboratory, Indiana State Board of Health, Indiana State Board of Animal Health, Department of Natural Resources, Department of Veterinary Pathobiology at Purdue University and others. Although this task force is just beginning to evaluate the needs of Indiana with regard to this disease, public and domestic animal vaccination programs have been the hallmarks of successful strategies in other states on the eastern coast which have dealt with this disease problem.

- By M. Randy White, DVM, PhD

Aquaculture Submissions to ADDL

Aquaculture case submissions range from one fish submitted by a fish hobbyist to numerous fish from large private or zoological collections, food-fish producers, or pet-fish suppliers. With the increasing number of hobbyists as well as the emerging aquaculture industry in Indiana, the number of aquaculture case submissions is rapidly increasing.

It is imperative to note that the condition of the fish sample submitted dictates the outcome of our diagnostic investigation in many instances. Care must be taken in collecting an adequate sample and transporting this sample to our lab.

The best sample for submission is an acutely affected, live fish exhibiting clinical signs or having gross lesions of disease. Clinical signs of disease are usually limited to anorexia, lethargy, abnormal swimming or position in the water column, "flashing" (rubbing of the body against a substrate in the aquatic environment), and the loss of fright response. Gross lesions include exophthalmia (unilateral or bilateral), ascites, skin erosions or ulcers, missing scales, frayed fins, or hemorrhage of the skin, eyes or fins. If small fish are involved in the disease outbreak (less than 4 inches, head to tail length) are involved, 6-8 fish should be submitted. If these fish represent a single disease process, as determined by the pathologist, then tissues are commonly pooled, and a single case accession fee is assessed.

The best and most assured method for transporting fish to the lab is hand delivery with the fish in a clean bucket, plastic or styrofoam cooler with water from the environment from which the fish originated. If transportation time is greater than 1-2 hours, it is recommended that a small battery operated aerator be used for supplemental oxygen. For shipment of fish, place fish in a large thick transparent plastic bag filled approximately 1/3 full with water. An "air cap" or oxygen should be present immediately above the water surface, occupying at least 1/3 to 1/2 of the plastic bag. The bag should be sufficiently tied and placed inside another bag to prevent leakage. This bag should be placed within a thick, wax-coated cardboard box for shipping. This box, along with the submission form, can be shipped via UPS or another appropriate overnight carrier company.

Since many aquaculture cases are associated with water quality problems, it is a good practice to submit a water sample with the fish submission. We are able to perform all of the standard water quality tests on water samples except for the dissolved oxygen

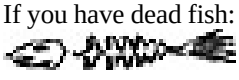
(DO) concentration. This water quality parameter is best evaluated "on site" since it changes rapidly in a sample container. THIS WATER SAMPLE MUST BE SUBMITTED SEPARATELY, i.e., without fish. This water sample should be submitted in a clean one quart glass jar with a screw top lid (i.e. canning jar) with a layer of aluminum foil placed between the water sample and the lid. Water samples shipped in this manner are also satisfactory for pesticide or herbicide analysis as well as testing water quality parameters.

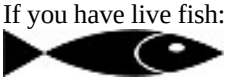
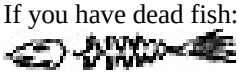
If recently deceased fish are the only sample available to the veterinarian for diagnostic evaluation, the best results are obtained by having the veterinarian take tissue samples for submission. Culturette swabs, with transport media, of the liver and spleen are the best bacteriologic samples for submission. The internal organs including gills, liver, spleen, gastrointestinal tract, integument, skeletal muscle, etc. can be submitted for histopathology in 10% formalin solution. Alternatively, if the fish is less than 3 inches in length, it may be submitted "whole" in formalin after opening the abdominal cavity to expose the internal organs to fixative.

If herbicides or pesticides are suspected, it is imperative to take a water sample from the pond or lake IMMEDIATELY. Detection of certain herbicides and pesticides can be made from frozen fish fillets, so filleting of the fish and freezing before shipment is the best way to prepare samples for shipment. The table below emphasizes the important points of this article.

A short history should be included with each submission. Husbandry, water quality problems, stocking densities, size of the tank/pond/lake, approximate mortality and morbidity, algal bloom history, origin of the fish, date and time of onset of current problem are all very pertinent issues to remember when completing the history portion of the submission form. Please feel free to contact us if you have questions regarding aquaculture submissions.

-By: Tim Muench, DVM, MS
-Edited By: M. Randy White, DVM, PhD

Suspected Problem	
If you have live fish: 	If you have dead fish: 
Water Quality	Water Quality
Parasitism	Parasitism
Bacterial Agents	Bacterial Agents
Viral Agents	Viral Agents
Environmental Contaminants	Environmental Contaminants

Best Sample To Submit	
If you have live fish: 	If you have dead fish: 
Water sample in clean glass quart jar	Water sample in clean glass quart jar
Acutely affected, live, ill, non-treated fish	Culturettes in transport media
* if < 3" long - submit 12-15 fish	Tissue sections of pertinent organs in 10% formalin solution
* if > 3" long - submit 6-8 fish	Frozen fillets (muscle and skin only)

PATHOGENIC *E. COLI* OF PIGS AND CALVES

Some strains of *E. coli* are intestinal pathogens, but other strains are part of the normal flora, making the diagnosis and interpretation of intestinal colibacillosis difficult. Consequently, a number of laboratories test *E. coli* isolates for the presence of virulence factors. Pathogenic *E. coli* can cause one or more of the following disease syndromes, depending upon which virulence factors they possess.

I. Enterotoxigenic (secretory) diarrhea.

II. Enteropathogenic diarrhea (necrotic or hemorrhagic).

III. Systemic toxemia (i.e. Edema Disease of pigs).

IV. Systemic (enteroinvasive) septicemia.

To determine whether an isolate of *E. coli* is pathogenic, it is most useful to test for functional factors that either permit the *E. coli* to colonize the gut (attachment factors such as pili), or permit the *E. coli* to cause damage (cytotoxic factors). Due to the number of virulence factors and the confusing terminology, interpretation of test results can be difficult. For your convenience, a table of commonly tested virulence factors and associated diseases are listed on the following page.

The table is intended to help you interpret results from various labs and is, therefore, redundant. For example, edema disease principle (EDP), verotoxin (VT), and shiga-like toxin II (SLT-IIv) are names used by various labs for the same toxin. In addition, bacteria

don't always read the books. For example, F18ab or F18ac pili are found on *E. coli* that colonize post-weaned pigs. They may either be associated with SLT-IIv (Edema disease), STB (enterotoxin-diarrhea) or both. When a single F18+ strain of *E. coli* secretes both SLT-IIv and STB, edema disease and post-weaning diarrhea can occur simultaneously. Also, there are undoubtedly other virulence factors that have not yet been discovered. For example, we sometimes isolate disease causing toxigenic strains that don't possess any known colonization or attachment factors.

-By: Duane Murphy, ADDL-SIPAC
Tom Hooper, ADDL-SIPAC
-Edited By: Greg Stevenson, DVM, PhD

E. COLI VIRULENCE FACTORS ASSOCIATED WITH PIGS & CALF DISEASES

FACTOR	DESCRIPTION	ASSOCIATED DISEASE
(Attachment Factors)		
*K88	Pilus antigen	Secretory diarrhea of pig.
F4	Same as K88	Secretory diarrhea of pig.
*K99	Pilus antigen	Secretory diarrhea of calf & pig.
F5	Same as K99	Secretory diarrhea of calf & pig.
*F41	Pilus antigen	Secretory diarrhea of calf.
*987P	Pilus antigen	Secretory diarrhea of nursing pig.
F6	Same as 987P	Secretory diarrhea of pig.
F(Y)	Pilus antigen	Secretory diarrhea of calf.
CS31A	Pilus antigen	Colisepticemia of calf.
F18ab	Pilus antigen	Post-weaning edema disease of pig.
F107	Old name for F18ab	Post-weaning edema disease of pig.
*F18ac	Pilus antigen	Post-weaning diarrhea of pig.
*2134P	Old name for F18ac	Post-weaning diarrhea of pig.
EAF	EPEC adhesive factor	Enteropathogenic diarrhea.
eae	Attaching-effacing gene	Enteropathogenic diarrhea.
(Cytotoxic Factors)		
LT	Labile toxin	Secretory diarrhea.
STA	Stable toxin A	Secretory diarrhea.
STB	Stable toxin B	Secretory diarrhea.
SDT	Shigella toxin	Human dysentery.
SLT-I	Shiga-like toxin I	Enteropathogenic diarrhea.
SLT-IIv	Shiga-like toxin II var	Edema disease of pig.
VT	Verotoxin	Edema disease of pig.
EDP	Edema disease principle	Edema disease of pig.
(Other Factors)		
*Hemolysis	Seen on blood agar	Diarrhea & edema disease of pigs
*0138	Cell wall antigen	Edema disease & diarrhea of pig.
*0139	Cell wall antigen	Edema disease of pig.
*0141	Cell wall antigen	Edema disease & diarrhea of pig.
08	Cell wall antigen	Diarrhea of pig.
045	Cell wall antigen	Diarrhea of pig.
0147	Cell wall antigen	Diarrhea of pig.
0149	Cell wall antigen	Diarrhea of pig.
*0157	Cell wall antigen	Diarrhea of pig (experimentally).
08	Cell wall antigen	Diarrhea of calf.
09	Cell wall antigen	Diarrhea of calf.
020	Cell wall antigen	Diarrhea of calf.
026	Cell wall antigen	Diarrhea & colisepticemia of calf.
0101	Cell wall antigen	Diarrhea of calf.
0141	Cell wall antigen	Diarrhea of calf.
02	Cell wall antigen	Colisepticemia of calf.
078	Cell wall antigen	Colisepticemia of calf.
086	Cell wall antigen	Colisepticemia of calf.
0115	Cell wall antigen	Colisepticemia of calf.
Col V	Colicin V plasmid	Colisepticemia of calf.
Vir	A virulence plasmid	Colisepticemia of calf.

*Tests available at the ADDL-SIPAC lab are indicated by a star. More complete testing can be done by request at other labs.

Catastrophic Oxygen Depletion and How to Avoid It

During the warm spring and summer months, we receive an increased number of phone calls regarding sudden fish kills in ponds. The typical history includes observing a very large number of fish dead in an otherwise normal pond following a rainstorm or summer thunderstorm. Usually the owner is very concerned that the fish may have died due to "run-off" of farm chemicals into the pond. Most of the time, these fish kills are a result of a phenomenon known as "pond stratification". Pond stratification is somewhat of a misnomer, since the stratification can also occur in lakes, creeks and some rivers. The stratification leads to a catastrophic depletion of oxygen which almost always results in a very high mortality of aquatic animal life within 24-48 hours following the "de-stratification".

The scientific reasoning behind this phenomenon of pond stratification relates to the temperature of the pond. In the early spring, while the temperature of the pond is still relatively low, the dissolved oxygen is uniformly distributed throughout the pond. As the atmospheric temperature increases, the pond begins to stratify, that is, become layered, with the surface water becoming warmer and lighter while the cooler and denser water forms a layer underneath. Circulation of the colder bottom water is prevented because of the difference in densities between the two layers of water. Dissolved oxygen levels decrease in the bottom layer since photosynthesis and contact with the air is reduced. The already low oxygen levels are further reduced through the decomposition of waste products, which settle to the pond bottom. After a rain, or any other event which disrupts the two layers, a "de-stratification" or "turn-over" of the pond occurs. This has the effect of releasing all of the dissolved oxygen from the upper layer of the pond into the atmosphere, hence, a catastrophic oxygen depletion.

Once stratification of a pond occurs, there is nothing that can be done to alleviate the situation. However, pond stratification can be very easily prevented by the use of supplemental aeration. Aerators come in all sizes and shapes as well as

different power sources, i.e., tractor p-t-o, electrical, mechanical, etc. It is important to aerate the pond properly, i.e. match the size of the aerator to the pond, since over-aeration is wasted and may even lead to oxygen supersaturation, known as "gas-bubble" disease and under-aeration will not prevent stratification.

In those cases where we suspect catastrophic oxygen depletion, all other possible pathogens including bacterial, viral, parasitic agents are eleminated from the differential diagnosis list. However, the history of several days to weeks of warm weather followed by a sudden rainstorm are highly suggestive of this condition. If you suspect that you are dealing with a pond turnover situation following pond stratification, it is imperative to have the pond owner take a water sample and have the dissolved oxygen (DO) concentration evaluated immediately. This water sample should be collected in a clean glass jar or bottle with a screw-top lid and should be completely filled by completely submersing the sample and container and placing the lid on the container while it is still under water.

By: Tim Muench, DVM, MS
Edited By: Randy White, DVM, PhD

Illustration of seasonal changes of water temperatures which occurs in fish ponds. In spring, temperatures and dissolved oxygen are uniform throughout the pond. During the summer, stratification may occur which creates an upper layer of water with high dissolved oxygen and a lower layer with low dissolved oxygen. After a rain or when a phytoplankton die-off occurs the water may turnover.

NCCVLD ABSTRACT

Case Report: Atypical PRRS Outbreak in Illinois Swine Herd

Dale M. Webb[1*]

A well-managed, 2,200 sow herd began experiencing illness characterized by inappetence, lethargy, fever, abortion, stillbirths, and sow death in early June, 1996. The average parity in the herd at the time of the outbreak was 3.2. Illness did not correlate with parity nor was it correlated with stage of gestation. Moving or treating sick animals resulted in exacerbation of clinical signs and, in some cases, death of the sow. The herd had prior natural exposure to porcine reproductive and respiratory syndrome (PRRS), and was routinely vaccinated with RespPRRS® (NOBL Laboratories). Abortions occurred in 8-10% of the affected sows and sows that did not abort had a high occurrence of stillbirths and mummified fetuses. Live-born piglets were often small and weak. Abortions and stillbirths also did not appear to correlate with parity.

Transmission of the disease was slow with neighboring sows becoming ill as long as several weeks apart. Affected animals generally returned to feeding and normal activity within 72 hours of first showing signs of illness. A few sows did not return to normal feeding and activity quickly. These animals were often found not to be pregnant even though they did not appear to cycle. The referring veterinarians[2] indicated the disease appeared to have affected 95% of the sows in the facility by the end of July (8-9 weeks from the onset of disease in the first animals).

Early in the course of the outbreak, two sows were submitted alive to the Illinois Department of Agriculture Animal Disease Laboratory in Galesburg, IL, for necropsy. The sows were in excellent physical condition and pregnant with near-term fetuses. There were no significant grossly evident findings. No bacterial growth was obtained from aerobic cultures of brain, lung, and pooled liver, kidney, and spleen. Salmonellae were not isolated from culture following enrichment techniques (tetrathionate broth). Both sows were leukopenic and had slightly increased hematocrits and total proteins, suggesting mild dehydration. Fibrinogen was less than 400 mg/dl in both animals, and total protein/fibrinogen ratios were 21 and 27, respectively. No antibody titers were detected to the routinely pathogenic serovars of *Leptospira interrogans*, except one sow ha a 1:100 titer against the *bratislava* serovar. One sow was seropositive for exposure to encephalomyocarditis virus at 1:32. One sow had a PRRS ELISA S/P ratio of 0.62; the other sow was seronegative.

Significant histologic alterations were limited to the lung and liver. The lungs had mild diffuse interstitial pneumonia. The liver had randomly distributed, disseminated foci of hepatic necrosis involving small groups of hepatocytes with variable (generally minimal), accompanying inflammatory infiltrates consisting of macrophages, lymphocytes, and neutrophils.

Attempts at virus isolation were unsuccessful at our laboratory (MARC 145 cells, swine testicular cells, and baby hamster kidney cells), the National Veterinary Services Laboratory (NVSL; fetal porcine kidney cells, swine testicular cells, and MARC 145 cells), and Iowa State University (ISU). Subsequent passage of liver homogenate into cesarean-derived , colostrum deprived piglets resulted in all piglets producing PRRS virus antibodies and PRRS virus was isolated from all piglets.[3]

Based on these findings, this diagnostically challenging case was eventually believed to be an

atypical PRRS-virus infection. A novel feature of the disease in this herd was the presence of necrotizing hepatitis, which had not been described previously, but has been seen subsequently in other outbreaks of atypical PRRS. Why this (and other) well-vaccinated herd broke with clinical disease is currently under further investigation at ISU and NVSL.

*Presenter

[1]Illinois Department of Agriculture, Animal Disease Laboratory, Galesburg, IL.
[2]Terry L. Bolton, DVM and James R. Lehman, DVM, Bolton and Lehman, Ltd., Atlanta, IL.
[3]Thanks to Pat Halbur, Veterinary Diagnostic Laboratory, Iowa State University, and Kelly Lager, National Veterinary Services Laboratory, Ames, IA.

NCCVLD ABSTRACT

A Diagnostic Dilemma: Turkey Coronaviral Enteritis in Southern Indiana

T.A. Bryan[1*], D. Hermes[2], T.A. Hooper[1], C. L. Kanitz[3], T.L. Lin[3], D.A. Murphy[1],
R.E. Porter[3], Jr., D.L. Schrader[3], H. L. Thacker[3], M.M. Woodruff[3]

Enteritis in southern Indiana turkeys had been a serious problem in Dubois County. Summer after summer affected flocks were shrill and uneasy, marching the house, refusing feed and water, acting chilled, but managing to impact their gizzards with litter. Dead birds were dehydrated, light in weight, with dark beaks and shanks. Grossly the cecae were distended with pale, syringable fluid. Usually young flocks were affected. The size uniformity in the flock quickly disappeared. It was noted by Dr. Hermes that flocks on crumbles had much lower mortality but similar morbidity than those fed mash. In the fall and winter mortality was not a problem but weight gain and feed conversion were economically unacceptable.

For a number of years, enteric virus detection has depended upon electron microscopy. In our experience, electron microscopy yielded varying results. There was no consistency within a flock. Electron microscopy results improved when naïve, test poults were exposed to field cecal material, followed by harvest of cecal contents after 2-3 days. Poult inoculation definitely improved electron microscopy results.

A literature search of Minnesota's TCE experience suggested an avenue for FA work that Dr. Kanitz had initiated at ADDL-SIPAC for other species. Dr. Y.M. Saif of OSU provided TCE antisera produced in SPF turkeys. Tom Hooper adapted the test procedure for use in several trials with inoculated and uninoculated poults of 5-7 days of age. In the direct immunoflorescence procedure composite sections of fresh frozen intestines taken from the yolk stalk to cecal pouch were stained with conjugated antisera. TCE FA+ diagnosed flocks continued to provide FA+'s for about 4 weeks after the first FA+ result.

For handling large numbers of serological tests, Tom Hooper propagated known antigen into 24 day old turkey embryos, harvested intestines 2 days later, and mounted the frozen intestine sections on glass slides. By testing convalescent sera from 6 poults within a suspect flock, incubating with previously prepared intestinal slides, washing, and staining with conjugated goat anti-turkey globulin, lab time was reduced as well as providing a greater window of opportunity for testing convalescent, suspect flocks.

In conclusion, our experiences suggest that the IFA test for turkey coronavirus has advantages over the EM and FA tests for diagnosing TCE because it produces more consistent results and is less labor intensive.

*Presenter

[1]Animal Disease Diagnostic Laboratory, Purdue University, Southern Indiana Purdue Agricultural Center, Dubois, Indiana.
[2]Perdue Farms, Inc., Washington, IN 47501.
[3]Animal Disease Diagnostic Laboratory, Purdue University, West Lafayette, IN 47907.

NCCVLD ABSTRACT

Pneumonic Pasteurellosis Associated with *Pasteurella hemolytica* Biotype A6 in American Bison (*Bison bison*)

N.W. Dyer[1*] and Alton C.S. Ward[2]

Three buffalo bulls (*Bison bison*) with a clinical history of respiratory distress and sudden death were submitted to the North Dakota State University Veterinary Diagnostic Laboratory (NDSU-VDL) for post mortem examination. The owner maintained a herd of forty-eight animals to which he had recently added twenty, sale-purchased, eight month old bull calves. The first calf died on December 25, 1996 followed by a second on December 27. Formalin-fixed and fresh tissues from the second calf reached the NDSU-VDL on December 30. In addition, the

carcasses of a calf which developed signs of respiratory disease on December 29, and a calf which died acutely on January 1, 1997, were submitted on January 3. Grossly, lung tissue showed variable amounts of bilateral, cranioventral hemorrhage and consolidation, pleural adhesions, and diffuse fibrinous pleuritis and pericarditis. Histologically, the pleura and interlobular septa were markedly expanded by focally intense clusters of neutrophils, fibrin strands and extravasated red blood cells. A similar inflammatory exudate was diffusely present within alveoli, bronchioles and bronchi. Curvilinear bands of streaming leukocytes frequently outlined these areas of fibrinosuppurative inflammation. Numerous pulmonary vessels showed vasculitis and thrombosis. Fluorescent antibody examination of lung tissue from all three animals was negative for IBR, BVD, BRSV and PI3. Culture of lung tissue from all three animals yielded a moderate to heavy growth of *Pasteurella hemolytica* biotype A serotype 6.

*Presenter

[1]Departments of Veterinary and Microbiological Sciences, North Dakota State University, Fargo, ND.
[2]Caine Veterinary Teaching and Research Center, University of Idaho, Caldwell, ID.

NCCVLD ABSTRACT

Equine Protozoal Myelitis: Current Knowledge About Etiologic Agent Life Cycle, Prevalence, Diagnosis and Treatment

M. Levy[1*]

EPM is a debilitating neurologic disease of horses. It can affect the brain, brainstem, spinal cord or any combination of these three areas of the central nervous system. Clinical signs may suggest focal or multifocal disease, which means the disease may affect a very small (focal) part of the central nervous system (CNS) or many parts of the CNS (multifocal). Therefore, the disease may present itself with a variety of different clinical signs, dependent on the location of the damage caused by the organism within the CNS.

Although EPM has been recognized since the 1970's, it was not until 1991 that the organism (protozoan parasite) was cultured from a horse and given the name *Sarcocystis neurona*. Opossum feces (definitive host) are the source of the infection for horses. Opossums acquire the infection by eating infected birds (intermediate host).

EPM occurs in much of North America. Surveys conducted in central Kentucky, one county in Pennsylvania and the entire states of Ohio and Oregon have revealed that approximately 50% of the horses have been exposed to this parasite. We know that a positive serum test indicates exposure to the parasite not necessarily the presence of disease.

EPM can affect a horse of any age, breed, or sex. The youngest horse reported affected was 2 months of age, and the eldest in its 30's. Clinical signs may be triggered or worsened by physiologic stress or the administration of corticosteroids. In most cases, affected horses are bright and alert with a normal appetite although some horses are dysphagic and may act as if they are choked.

Ante mortem diagnosis of EPM is based on clinical signs and on testing of the horse's cerebrospinal fluid (CSF) by the western blot test. If blood contaminates the CSF sample, a false positive test may result.

Treatment of horses with EPM is expensive. The average range of treatment is 90 to 120 days, and may exceed 6 months in some instances. The current approaches to treatment for EPM includes pyrimethamine in combination with a sulfonamide antimicrobial with or without trimethoprim. It would appear that early detection and therapy increases the chance of successful treatment.

*Presenter

[1]Department of Veterinary Clinical Sciences, School of Veterinary Medicine, Purdue University, West Lafayette, IN.

NCCVLD ABSTRACT

Defining Proliferative Enteritis As A Cause of Protein Losing Enteropathy in Foals

C. Fishman[1*] and C. Gebhart[2]

Lawsonia intracellularis (LI) is an obligate intracellular, bacillary bacterium which has been established as the cause of proliferative enteritis (PE) in swine. PE associated with LI infection has been documented as an uncommon to rare disease in the hamster, guinea pig, rabbit, ferret, dog, fox, horse, deer, ostrich and non-human primate. There are currently two published reports describing the pathology of PE in the foal. There are no reports describing the clinical aspects of PE in foals, and reviews of causes of enteritis in foals do not include

this disease. PE as the cause of protein losing enteropathy was diagnosed in a six month old, quarterhorse colt presented to the Animal Disease Diagnostic Laboratory at Purdue University, West Lafayette, IN. The animal had been admitted to the Large Animal Teaching Hospital, School of Veterinary Medicine at Purdue University with the clinical history of sudden onset of diarrhea and anorexia. The animal was febrile and dehydrated. Clinical laboratory data revealed hypoproteinemia with comparable decreases in albumin and globulin, mild normocytic, normochromic anemia, mild leukocytosis with neutrophilia, left shift and toxic neutrophils and lymphocytosis with reactive lymphocytes, hyperfibrinogenemia, mild azotemia, hypocalcemia, hypomagnesemia, hyponatremia and metabolic acidosis with compensatory respiratory alkalosis. The animal failed to respond to treatment, which included plasma transfusions, intravenous fluids, corticosteroids, bismuth subsalicylate and cimetidine, and over a three week period became increasingly cachectic, developed subcutaneous, dependent edema and was euthanized. Gross necropsy revealed diffuse thickening of the jejunum and ileum with a variably corrugated or multinodular expansion of the mucosa, submucosal edema and muscular hypertrophy. The affected mucosa was multifocally ulcerated, and in one focus, a transmural ulcer resulted in intestinal perforation and septic peritonitis. The microscopic lesions in the affected small intestine closely resembled that of porcine PE, including villous blunting and deepening of crypts which were tortuous with complex branching patterns. Crypts were lined by crowded, multilayered enterocytes with vesicular nuclei containing prominent nucleoli and a moderate amount of cytoplasm with increased basophilia and a lack of mucus vacuoles. A Warthin-Starry stain revealed myriads of curved bacilli in the apical cytoplasm of the hyperplastic enterocytes. The mitotic index in crypt enterocytes was increased. Crypts were "herniated" into the submucosa, particularly in the area of Peyer's patches. In ulcerated foci, the intestinal wall was partially to fully replaced by granulation tissue. Immunohistochemical stains of paraffin-fixed tissue were positive for LI using a specific monoclonal antibody. The DNA of the bacterium in this case was shown to be homologous to that of swine isolates using extraction, PCR amplification and gel electrophoresis. Transmission electron microscopy revealed bacillary bacteria, measuring 2-4 x 0.2 microns, with a wavy trilaminar cell wall, free within the apical cytoplasm of enterocytes and often adjacent to mitochondria. The clinical signs and response to therapy of this foal were inconsistent with other commonly reported causes of enteritis. If an index of suspicion of PE exists, LI can be detected using PCR on feces and by serum antibody tests. Ante mortem identification of PE cases in foals will allow for estimates of prevalence and will provide epidemiologic information. This is the first case report to correlate clinical and clinical pathologic findings with gross and microscopic morphology in a horse with LI infection.

*Presenter/Graduate Student

[1]Animal Disease Diagnostic Laboratory, Purdue University, West Lafayette, IN 47907.
[2]Department of Veterinary Pathobiology, University of Minnesota, St. Paul, MN 55108.