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IN THIS ISSUE

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LLOYD'S

Commentary

This issue of the Equine Disease Quarterly contains an update of nocardioform placentitis cases in central Kentucky. In an effort to better understand this emerging condition, a survey and experimental study were recently conducted. While useful information was generated, the results reported herein underscore the inherent difficulties often encountered in experimental studies.

The basic goal of any experimental study is to test the validity of a theory (hypothesis) regarding a particular disease or condition. The hypothesis is generated following careful observation of naturally occurring cases of the disease and then posing questions regarding causes, interactions with body systems, treatments, and communicability.

The hypothesis is tested by reproducing the disease in experimental subjects under controlled conditions and measuring the effects or responses. Sufficient numbers of subjects to be statistically significant must be used and careful control of variables that could influence the outcome is essential to produce valid information.

Like pieces of a puzzle, research often builds upon itself with experimental findings generating other questions and hypotheses, ultimately leading to a thorough understanding of a disease condition and ways to treat or prevent.

Unfortunately, the path to unraveling the mysteries of a disease is often fraught with pitfalls. The interaction of a disease-causing agent with the host is complex with many exogenous as well as endogenous variables affecting susceptibility and severity. Factors such as virulence of the microorganism in question, infective dose, animal age, breed, sex, and individual susceptibility can affect the outcome. Many of the variables are unknown and, therefore, are difficult or impossible to account for and control during experimentation.

There are numerous examples of failed attempts to experimentally reproduce an equine disease. Equine protozoal myeloencephalitis (EPM), a common neurologic disease, could not be reproduced experimentally by injecting protozoa obtained from horses with EPM, even though large numbers of protozoa and immunosuppressive agents were used. It was later learned that only the stage present in the intestine of the definitive host (opossum) was infectious for horses. Even then only mild disease was produced.

Potomac horse fever (PHF), a severe alimentary disease, was believed to be spread by blood-feeding insects, based on its seasonal occurrence and the ability to transmit the disease by injecting blood from a sick horse into a normal horse. However, many attempts, over almost 20 years, to experimentally transmit PHF using biting insects were unsuccessful. It was not until the discovery that the intermediate host was actually a terrestrial snail infested with flukes that successful transmission was achieved.

Equine herpesvirus-1 infection causes abortion in late gestation mares. However, in experimental studies to test vaccine effectiveness, difficulties have been encountered in producing abortions in infected control mares. In another example, equine influenza is a highly contagious respiratory condition; however, experimental intranasal instillation of high doses of virus was ineffective in producing similar disease. It was not until a nebulizer was utilized to deliver the virus that accurate reproduction of clinical disease was accomplished.

To effectively study and understand a disease it is necessary to be able to experimentally reproduce the disease. From these examples it is obvious that this is not always a simple process. ■

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International

Second Quarter 2000

The International Collating Centre, Newmarket and other sources reported the following disease outbreaks.

A single case of contagious equine metritis was confirmed in Japan and in California, USA a single case of eastern equine encephalitis was diagnosed in a yearling show horse during May. Abortions attributable to equine herpes virus (EHV) were reported from Germany: 4 cases on 3 premises; Ireland: 2 cases on 2 premises; Japan: 3 cases on 2 premises; Netherlands: single cases on a small number of farms; Sweden and Switzerland: 2 and 3 notifications, respectively; and in the United Kingdom: single cases on 3 premises.

Twenty-seven cases among Thoroughbred mares were confirmed in central Kentucky on 16 farms. Multiple abortions occurred on 3 farms (9, 3 and 2) among vaccinated mares. Respiratory disease caused by EHV was extensively reported in France. Cases of coital exanthema (EHV-3) among non-Thoroughbreds were diagnosed in Switzerland and the United Kingdom and a Thoroughbred stallion in Turkey.

Further to the report of equine infectious anemia (EIA) among non-Thoroughbreds at an equestrian center in France during the first quarter, tracing has identified 7 additional cases on 5 other premises. Respiratory disease and abortion attributable to equine arteritis virus (EAV) was diagnosed on 2 premises in Denmark.

Influenza was reported among horses in France, Sweden, Switzerland, Turkey (on 2 racetracks during January and February) and the United Kingdom. Rotavirus infection among foals was reported on several premises in Ireland. Strangles was reported from Australia, Denmark, Ireland, Sweden, Switzerland, United Arab Emirates, United Kingdom and USA.

Turkey reported that commencing in the year 2000 a national equine disease testing and eradication program for glanders would be undertaken as well as surveillance for African horse sickness, EIA, dourine and equine viral arteritis.

Hong Kong reported a high incidence of horses at Sha Tin Racecourse and several riding schools showing signs of inappetence and mild pyrexia during April and May. Investigations are continuing to identify the cause.

A 3-year-old Thoroughbred colt imported to Hong Kong from the USA in 1999 was diagnosed as a clinical case of equine protozoal myeloencephalitis (EPM) and euthanized, December 1999. Testing of the entire equine population of Hong Kong for equine piroplasmosis by the indirect fluorescent antibody test (IFAT) found no positive samples. ■

National

West Nile Update

Information for this update was derived from the National Atlas of the United States at www.nationalatlas.gov/virusmap.html, the State of New York Department of Health at www.health.state.ny.us, ProMed at www.promedmail.org and USDA at www.aphis.usda.gov.

As a result of intensive, active surveillance, a considerable amount of information regarding the distribution of West Nile Virus (WNV) has accumulated over the summer months. Based on the most recent reports as of September 18, 2000 six states in the northeast have reported the presence of the virus: New York, New Jersey, Connecticut, Massachusetts, Rhode Island and New Hampshire (see **Figure 1**).

The virus has been reported most frequently in dead birds and mosquitoes particularly since August 1, with 12 reported human cases and 8 equine cases. The state of New York has identified 680 positive dead birds of 47 different varieties, 448 of which were crows, distributed over 51 counties, including boroughs of New York City. Additionally, in the same state the virus has been identified in 6 healthy live birds, 9 bats, and 1 raccoon.

In New Jersey 726 dead birds have been positively identified in 15 counties, all crows except 1 cockatiel; Connecticut, 280 birds in 7 counties; Massachusetts, 103 birds in 8 counties; Rhode Island, 4 birds in 3 counties; and New Hampshire, 1 bird.

While flocks of sentinel chickens have been monitored extensively, only 1 chicken in Westchester County, New York has so far sero-converted.

WNV has been identified in mosquito pools in New York (269), New Jersey (12), Connecticut (4), and Massachusetts (3). The vast majority of positive mosquito pools were clustered around and in the city of New York.

Several different species have been found to be infected, the majority being *Culex pipiens* but also *C. restuans* and *salinarus* as well as *Aedes species, A. japonicus, A. triseriatus, A. vexans, Anopheles*

punctipennis, and *Psorophora ferox*.

Of the 12 human cases, 11 were in New York City and 1 in New Jersey; all are recovering. Eight equine cases have been reported: Connecticut (3), New Jersey (2), New York, Rhode Island, and Massachusetts, of which 6 were fatal.

Equine fatalities attributable to WNV have recently been reported in the south of France and human cases have been confirmed in Israel. ■

Drugs and Show Horses

The American Horse Shows Association (AHSA) is the National Equestrian Federation for the United States, enforcing rules for 26 disciplines from Shetland pony to hunter/jumper to endurance riding divisions.

In 1970 the AHSA Drugs and Medications Program began testing samples from equine athletes in order to protect the fairness and safety of AHSA recognized events, and is now the largest private equine drug testing program in the world.

Veterinarians and technicians obtain blood and urine samples from randomly selected horses at competitions. These samples are sent to the AHSA drug-testing laboratory for thorough testing for drugs which may alter the horse's performance.

Drugs and medications are classified by the AHSA as being permitted, restricted, or forbidden. Permitted drugs are those assumed to be only therapeutic which do not affect the horse's performance, such as many antibiotics, dewormers, some hormones, vitamins, and electrolytes.

Restricted drugs include methocarbamol (muscle relaxant) and five non-steroidal anti-inflammatory drugs (NSAIDs): phenylbutazone, flunixin, ketoprofen, meclofenamic acid and naproxen. These restricted drugs must only be given therapeutically to horses in very defined doses at specific time intervals prior to competition.

AHSA Rules permit the use of not more than two of the NSAIDs. Administration of these restricted drugs in excess of published amounts is forbidden.

Forbidden drugs include those which affect the performance of the horse, or mask the presence of a forbidden substance. Examples include any stimulant, tranquilizer, local anesthetic (*e.g.*, the procaine in procaine penicillin), mood-altering drugs, etc. Many drugs are forbidden, including substances as obvious as cocaine, and ones as simple as caffeine found in chocolate or cola.

Horse Show Magazine, the official publication of the AHSA, lists all notices of penalty summarizing drug violations. Issues from February 1998 through August 2000 were reviewed to determine the types of forbidden drug usage being detected at AHSA horse shows, and the fines associated with these actions. Since cases are still pending, and samples from Summer 2000 shows are in the testing process, it should not be assumed that all drug detections through August 2000 are represented here. For the purposes of data discussion, a restricted drug in excessive amounts or a forbidden drug is termed "forbidden." In 1998 and 1999 the AHSA tested approximately 500 days of competition each year, with about 600 days being tested in 2000.

Violations in 94 horses were noted during this time span, with 76 involving detection of a single forbidden drug; 9 cases of 2 forbidden drugs; 8 cases of 3 drugs; and 1 horse with 4 drugs being detected. In the cases of horses testing positive for 3 and 4 drugs simultaneously, all drugs were NSAIDs. Of horses with 2 drugs detected, 6 involved NSAIDs and 3 involved miscellaneous drug combinations.

Of the 76 horses with one forbidden drug detected, excessive NSAIDs were the most numerous: phenylbutazone (15), flunixin (15), and ketoprofen (11). Other forbidden positives included tranquilizers/sedatives (14), antihistamines (7), local anesthetics (6) and miscellaneous drugs (8). Miscellaneous drugs included cocaine, Ritalin, and mephentermine (an amphetamine illegal even for human use), among others.

Trainers are ultimately responsible for the proper use of medications in their horses. When forbidden amounts or types of drugs are detected, the trainer usually has a monetary fine, and depending upon the offense, may be suspended from training and showing horses. All awards, ribbons and prize money earned by the positive horse are required to be returned. In the above cases, fines ranged from $50 (NSAID offense) to $7,500 (use of detomidine, a potent sedative/analgesic). For the 94 cases of illegal drug usage in horses, fines totaled $116,000; 33 trainers were fined $1,000 or more.

Trainers and owners should be acutely aware of the AHSA Drugs and Medications Rule which can be obtained by calling the Drugs and Medications Program, 1-800-MED-AHSA, or by reading it on-line at www.ahsa.org. ■

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Figure 1.
West Nile Virus Update
as of September 18, 2000

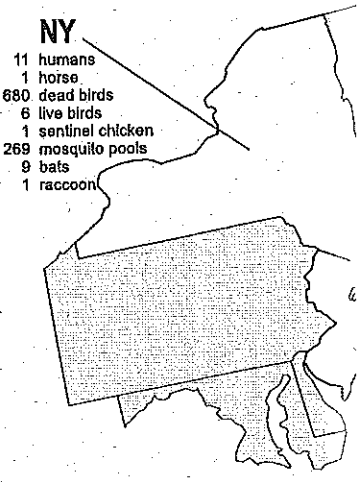
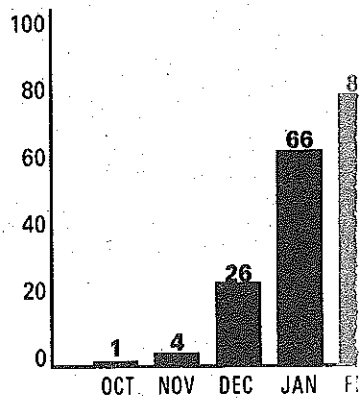
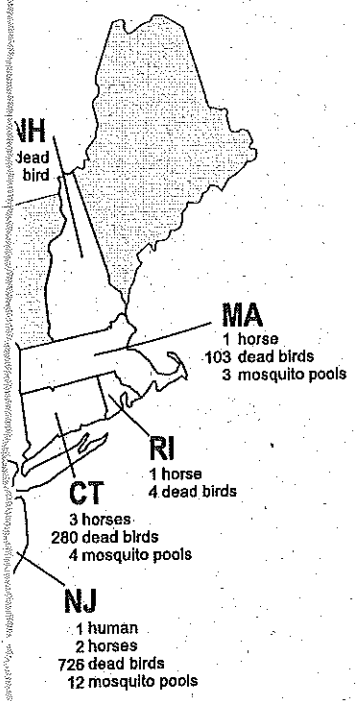
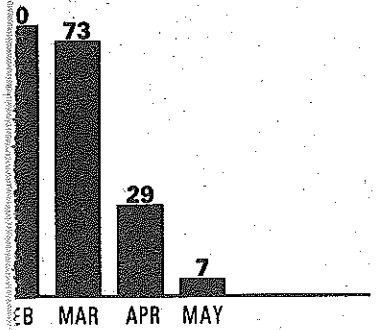


Figure 2.
Nocardioform placenti
(1998-2000 foaling seasons).





is cases by month



NAHMS Equine '98 Study

Highlights Industry's Lack of Awareness of EVA

Equine viral arteritis (EVA) was one of the equine diseases selected for inclusion in the USDA's National Animal Health Monitoring System (NAHMS) Equine '98 Study, the primary goal of which was to gather data on equine management and determine the seroprevalence of antibodies to equine arteritis virus (EAV) from a statistically valid and representative sampling of horses on operations in the U.S.

A total of 7,027 blood samples were obtained from horses residing on 837 (81.8% of 1,023) operations in 25 states that participated in the EAV serological study. Data on individual horses, including vaccination status for EVA, was recorded at the time of blood collection. Blood samples were tested for neutralizing antibodies to EAV at the USDA's National Veterinary Services Laboratory (NVSL), Ames, Iowa.

Based on responses to a questionnaire, some 59.4% of horse operation personnel/owners had never heard of EVA and an additional 27.6%, though familiar with the name, knew little else about the disease. Only 13.0% knew some basics or were knowledgeable of the disease.

The larger the operation, the greater the likelihood the operator had some knowledge about the disease. Familiarity with EVA was higher for operations where at least 50% of the resident horse herd was Thoroughbreds and/or Standardbreds and where the horses were used primarily for racing or breeding.

Only 1.6% of the operations surveyed had vaccinated at least one resident horse against EVA and only 0.9% had tested any horses for EVA in the previous 12 months. The percentage increased with size of operation, which was based on the horse inventory.

The overall prevalence of neutralizing antibodies to EAV was 25.3% in horses with a history of vaccination and 2.0% in non-vaccinated horses. The percentage of operations with at least one seropositive horse increased with vaccination use. The highest percentage of seropositivity in non-vaccinated horses was in Standardbreds. The seroprevalence of EAV antibodies in non-vaccinated horses increased with age.

In light of the findings of the NAHMS Equine '98 Study, a vigorous educational program on EVA reaching out to all sectors of the horse industry is urgently needed to help dispel the widespread lack of knowledge about the disease. In spite of the availability of a safe and effective vaccine against EVA (ARVAC®, Ft. Dodge Animal Health) for the past 15 years, few horse operations report vaccination of their horses against this disease. Operations with breeding stallions as well as mare owners are evidently unaware of the significance that the carrier stallion plays in the epidemiology of EVA, *e.g.*, the carrier stallion is the primary reservoir for the virus between outbreaks of EVA.

A concerted effort should, therefore, be made by equine practitioners and the American Horse Council to educate owners/managers of the need to screen their breeding stallions for evidence of EAV infection and inform them of the risks, legal and otherwise, associated with failing to determine the EVA status of their stallions.

The findings of the serological survey confirm only small differences in the prevalence of EAV infection between different regions in the U.S.A. The seropositive percentage among Standardbreds (23.9%), though the highest of the breeds represented in the survey, is considerably less than that reported in several previous studies.

The survey results confirm that the great majority of the horse population is seronegative and, therefore, highly susceptible to infection should they become exposed to EAV. The unrestricted importation of carrier stallions and virus-infective semen enhances the risk of dissemination of EAV throughout the country and the likelihood of an increase in the number of outbreaks of EVA.

Measures to identify carrier animals/infective semen at the time of importation would help greatly in reducing the risks involved. Implementation of a strategic EVA vaccination policy of all stallions of all breeds and colts between 6 and 12 months of age would further limit the number of carrier stallions and in time, eliminate the primary reservoir of the virus.

More detailed information on the findings of the NAHMS Equine '98 Study on EVA (publication N315.0400) is available from the Centers for Epidemiology and Animal Health, USDA: APHIS: VS, Attn. NAHMS, 555 South Howes, Ft. Collins, CO 80521; E-mail: NAHMSweb@usda.gov; Website: http://www.aphis.usda.gov/vs/ceah/cahm. ■

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Kentucky

Nocardioform Placentitis

In the July 1998 issue of the *Equine Disease Quarterly* we reported a dramatic increase in cases of nocardioform placentitis in central Kentucky. In this issue we provide an update on cases occurring during the past two foaling seasons.

Nocardioform placentitis is a distinct form of placentitis in mares. It is diagnosed commonly in central Kentucky with a few unconfirmed reports of occurrence in other areas. All ages and breeds of horses are affected.

Mares with this type of placentitis often show no outward signs of infection but may have premature mammary gland development and lactation. The mare will abort in late gestation, have a stillborn or weak foal prematurely or at term, or produce an apparently normal foal. Following delivery the mare usually clears the infection rapidly, and typically will breed back normally with no increased risk for repeat infection.

The placenta in nocardioform placentitis cases is characterized by an area of placentitis located on the cranial portion of the body of the placenta, often extending onto the horns. This area is often large, but solitary, and does not communicate with the cervical portion of the placenta. The surface of the placenta is covered with thick brown exudate containing sloughed chorionic cells, leukocytes, and bacteria.

The bacteria are gram-positive branching filamentous organisms that are unclassified and appear to represent at least 3 genera based on differences in morphology, antibiotic sensitivity, and nucleic acid sequencing.

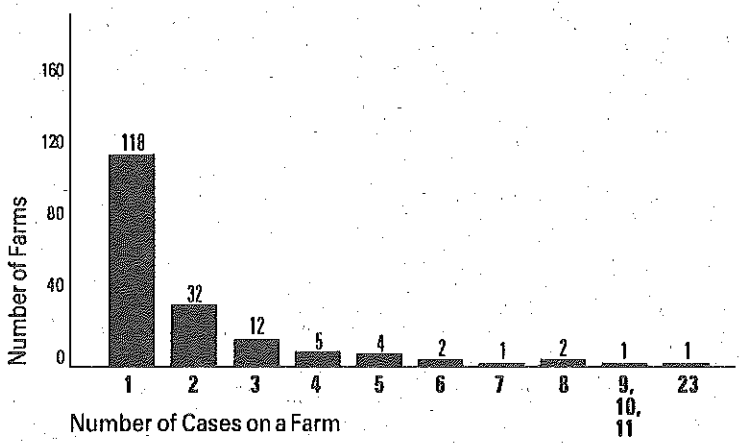
Testing has revealed that approximately 66% of the isolates appear to belong to one species. Placentitis cases associated with this species tend to have high fetal/foal mortality, while an estimated 85% of mares infected with the other species produced foals that survived. The bacteria invade the chorionic epithelium but do not spread to the fetal fluids and organs; therefore injury to the fetus appears to be the result of placental insufficiency.

Prior to 1998 the Livestock Disease Diagnostic Center diagnosed an average of 20 cases of nocardioform placentitis each foaling season. In the 1998 foaling season 94 cases were diagnosed, followed by 144 cases in the 1999 foaling season. The 2000 foaling season brought a reduction in diagnoses with 48 total.

The cause of these increases over the prior average is not known and there was not a concomitant increase in other types of equine placentitis cases. Nocardioform placentitis was typically seen in late gestation and the monthly occurrence of cases corresponds to the normal foaling season **(Figure 2).**

Nocardioform placentitis occurs sporadically, and on farms where it was diagnosed there were usually only one or two cases in a given year. Review of nocardioform placentitis cases over a 9-year period revealed that of the farms that had cases, 83% had 2 or fewer over the 9 years, and that 66% of the farms had only a single case over the 9-year period **(Figure 3).**

Figure 3. *Number of farms with one or more nocardioform placentitis cases (1991-1999).*



No farm management or veterinary practices have been associated with the development of nocardioform placentitis.

In 1998 and 1999, questionnaires were sent to veterinarians and farms submitting a case of nocardioform placentitis. Information was obtained regarding prior reproductive health of the mare, treatments associated with breeding, breeding practices, stallion used, and the mare's management and environment. The results did not identify any common factor or practice in mares with nocardioform placentitis and, consequently, no risk factors were identified.

Limited studies were undertaken in an attempt to reproduce nocardioform placentitis in mares and to establish how long the bacteria could be recovered from the reproductive tract following inoculation. Although large numbers of viable bacteria were used, both studies were unsuccessful in establishing infection. This suggests that other unknown factors play a role in the development of nocardioform placentitis. ■

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