



EQUINE DISEASE QUARTERLY

FUNDED BY UNDERWRITERS AT LLOYD'S, LONDON

JULY 2019
Volume 28, Number 3



COMMENTARY

IN THIS ISSUE

Commentary

International

First Quarter
2019..... 2

Changing Trends
in Equine
Reproduction..... 3

National

Clostridial Myositis... 5

In an article contained in this issue of *EDQ*, Dr. Troedsson nicely details advances in the science and practice of equine reproduction over the past 50 years that have contributed to improved fertility and breeding efficiency in the horse. It is important to point out, however, that research conducted at the Kentucky Agricultural Experiment Station at the University of Kentucky beginning over 100 years ago laid much of the groundwork for understanding important diseases impacting reproduction as well as the clinical evaluation of stallions and mares for infertility. Beginning in 1911, Good and others worked to identify the causative bacterium for outbreaks of infectious abortion (*Salmonella abortusequi*). Subsequent development of a bacterin and control procedures led to the eventual eradication of this important cause of infectious abortions in the U.S. Research by Dimock, later joined by Edwards and Bruner, led to descriptions of techniques for the clinical examination of mares for pregnancy as well as a detailed description of clinical, bacteriologic and pathologic changes in the reproductive tract of infertile mares. This work provided the foundation for recommendations related to breeding hygiene, uterine infections, infertility as well as pregnancy detection in mares. In 1933, research by this group provided early characterizations of equine (herpes) virus abortion. They also conducted large studies that characterized both infectious and noninfec-

tious causes of abortion in mares in Central Kentucky. Subsequent work by Doll, Bryans, and later Allen in the Department of Animal Pathology at UK lead to the development of the first live and inactivated virus vaccines for the control of equine herpesvirus abortion in mares. In the mid-1950s, Doll, Bryan, McCollum, and Crowe described the viral agent causing equine viral arteritis and abortion in mares with subsequent development of vaccination and control strategies based upon the work of Timoney and McCollum in the mid-1980s. Research by Loy and others led to a better understanding of postpartum reproduction in mares, together with the use of artificial lighting regimes and endocrine manipulation for control of the estrous cycle in mares. Although this chronology is incomplete and omits other significant research, I think that this historical perspective adds to the work characterized in Dr. Troedsson's article. In particular, it demonstrates the need for research expertise and interest focused on reproduction in the horse to address current problems as well as unforeseen future problems related to equids.

CONTACT:

Barry A. Ball, D.V.M., Ph.D., D.A.C.T.
b.a.ball@uky.edu
(859) 218-1141
Maxwell H. Gluck Equine Research Center
University of Kentucky
Lexington, KY

 University of
Kentucky
College of Agriculture,
Food and Environment
Department of
Veterinary Science

LLOYD'S



First Quarter 2019

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

For the period under view, African horse sickness (AHS) was reported by Cameroon, Chad, the Republic of South Africa (RSA), and Swaziland. Swaziland reported four outbreaks of AHS, each involving very limited numbers of horses. Two outbreaks of AHS were confirmed in Cameroon and Chad. Both outbreaks in Cameroon involved working horses and donkeys, and horses were primarily affected in Chad. Incidence of the disease was above average in the RSA, especially in Gauteng Province. Sporadic cases were recorded in the remaining provinces in the AHS infected area. No cases were reported to date in the Western Cape Province, either in the infected or disease-free areas.

Equine influenza (EI) was confirmed in numerous countries in Europe (Belgium, Denmark, France, Germany, Ireland, the Netherlands and the UK), West Africa (Burkina Faso, Ghana, Mali, Nigeria, Niger, and Senegal), and the USA. In Europe, the outbreaks primarily involved unvaccinated horses, but the disease also was observed in vaccinated horses. Strains of clade 1, Florida sub-lineage of H3N8 EI virus were implicated. The major impact of EI in West Africa was seen in donkeys, in which thousands of cases were reported with very high fatality rates. Equine influenza is endemic in the USA; 57 outbreaks were confirmed in eight states during the first quarter of 2019.

Strangles remains endemic in most countries: France (33 outbreaks), New Zealand (one outbreak), Sweden (three outbreaks), the UK (one outbreak), and the USA in which the disease is endemic. Eighty-four outbreaks were diagnosed in 18 states, with multiple outbreaks in three states.

Equine herpesvirus 1 (EHV-1) related diseases were reported by Belgium, Canada, France, Germany, Japan, Norway, Sweden, the UK, and the USA. The number of confirmed outbreaks of respiratory disease ranged from one (Norway and Sweden), two (Belgium, the UK and the USA), and four (France). Cases of EHV-1 abortion were diagnosed in Belgium (one), Canada (two), France (five), Germany (one), Japan (eight), Sweden (two), the UK (four), and the USA (eleven).

Equine herpesvirus 1 neurologic disease was reported by France (three outbreaks), Germany (three outbreaks), Sweden (three outbreaks, each involving multiple cases), and the USA (13 outbreaks in eight states, three involving multiple cases).

Equine herpesvirus 4 respiratory disease was recorded in Argentina, Belgium, and the RSA (one outbreak apiece), Sweden and the UK (each with seven outbreaks), France (36 outbreaks), and the USA, in which the disease is endemic.

Equine herpesvirus 2 infection was reported by the USA, in some instances associated with respiratory disease.

Bulgaria, Canada, Peru, and the USA confirmed outbreaks of equine infectious anemia. The number of cases ranged from one (Bulgaria, Peru) to five in Canada. The USA recorded the disease in five states, with the number of cases by state ranging from one to nine.

Equine piroplasmiasis was reported by Ireland (a case of *Babesia caballi* infection in an imported mare and her aborted foal) and the RSA in which the disease is endemic.

Cases of leptospiral abortion were confirmed by Belgium (one) and the USA (two). The USA diagnosed four cases of nocardioform placentitis, three associated with *Amycolatopsis* spp and one with *Crossiella equi* infection, and four other cases of mucoid placentitis.

Two cases of equine coital exanthema were confirmed in the USA.

Clostridial enterocolitis was diagnosed in neonatal foals in the USA, five of which were associated with *Clostridium perfringens* infection. Additionally, four cases of *C. difficile* infection were confirmed.

Of neurological diseases reported during the first quarter of 2019, one case of rabies and nine cases of Eastern equine encephalomyelitis were reported by the USA, all but one in Florida. During the same period, the RSA confirmed one case of West Nile virus infection and 60 cases of equine encephalosis that occurred in a total of five provinces. Mexico reported 41 outbreaks of Western equine encephalomyelitis in the state of Nayarit. Fifty-three cases of the disease were recorded in these outbreaks, including seven fatalities.



Equine Disease Quarterly

Editors

Peter Timoney
Alan Loynachan

Staff

Diane Furry
Tawana Brown
Dennis Duross
Anita Fleming

Correspondence should be addressed to the editors, Department of Veterinary Science, Maxwell H. Gluck Equine Research Center, University of Kentucky, Lexington, Kentucky USA, 40546-0099
Telephone (859) 257-4757
Fax (859) 257-8542

Internet address:
<http://gluck.ca.uky.edu/equine-disease-quarterly>

Material published in the Quarterly is not subject to copyright. Permission is therefore granted to reproduce articles, although acknowledgment of the source and author is requested.

The University of Kentucky is an Equal Opportunity Organization.



Printed on recycled paper

Changing Trends in Equine Reproduction

Research advances in equine reproductive biology and health over the past 50 years have contributed to improved fertility and breeding efficiency in horses. The development of endocrine and ultrasonography diagnostics aids the clinician in determining potential fertility of mares and stallions. Follicular dysfunction and ovulation failure can be diagnosed with high precision in mares, embryo and fetal development can be monitored from 10 days after ovulation throughout gestation, and fetal/placental abnormalities diagnosed to identify high-risk pregnancies. In stallions, biochemical and genetic sperm abnormalities have been identified that previously remained undetected based on traditional breeding soundness examinations. Furthermore, testicular abnormalities and blood flow can be determined with high accuracy by Doppler flow ultrasonography.

The current trend in horse breeding is characterized by a decline in mares that are bred and a new focus on wellbeing of established pregnancies as well as enhanced genetic selection with regards to health and future performance of the foal. In addition, great advances in the area of assisted reproductive technologies offer numerous possibilities that were not available in the past.

Monitoring Healthy Pregnancy

Biomarkers for high-risk pregnancies and efficient treatments to prevent late pregnancy losses are being investigated. As new potential markers for unhealthy conditions are discovered, effective treatment options need to be developed. A similar trend is seen with regards to endometritis as well as the development of new diagnostics for dormant bacteria and uterine biofilms. While new therapeutic regimens for endometritis often include non-antibiotic alternatives or supplements, most treatment options for high-risk pregnant mares include long-term use of broad-spectrum systemic antibiotics without the possibility of performing bacterial sensitivity tests. There is a global awareness of consequences when antibiotics are overused in human as well as veterinary medicine—and equine reproduction is not an exception. An overuse of antibiotics can lead to bacterial resistance and contribute to one of the most urgent public health issues today. New trends in equine reproduction reflect these challenges to

veterinary medicine, public health and the global equine breeding industry, and additional non-antibiotic treatment options need to be developed.

Improved Genetic Selection

The art of breeding horses is based on genetic selection. Horsemen evaluate visible characteristics (phenotypes), such as conformation, coat color, speed, endurance as well as family history (blood lines) and make breeding decisions based on these evaluations. As equine breeding has shifted from quantity to a greater focus on quality, genetics and genomic research in reproduction has the potential to improve the accuracy of breeding selection criteria. Research leading to sequencing of the equine genome has resulted in the identification of a growing number of genes that have been linked to positive characteristics (speed, coat color, etc.) as well as heritable diseases. These scientific advances are currently used to test potential carriers before making breeding decisions. If allowed by the breed registry, pre-implantation genetic testing of embryos provides a more effective method to select for (or against) certain genetics. Recent and ongoing research allows us to recover embryos non-surgically from the uterus a week after conception and test the conceptus for the presence or absence of specific genes. The embryo can be cryopreserved while molecular genetic testing is performed in the laboratory and if test results are favorable, it can be transferred into a recipient mare or even into its biological mother at a later time. The potential benefit of this technology to horse breeding is obvious. For example, unless genetic carriers of a debilitating disease (and potentially also carriers of some desirable genes) are completely removed from breeding, selection of healthy embryos based on pre-implantation genetic testing is the only way to eradicate the disease. Because of its practical benefit to horse breeders, the technology of pre-implantation genetic testing in horses can be expected to be further developed and gain popularity as research identifies additional genes that influence a variety of characteristic of horses, and tests become available. A related area of expected research intensity in the near future is investigations on the importance of environmental influence on gene expression during fetal development (epigenetics).



STEVE PA

Assisted Reproduction

Assisted reproduction is not suitable to all breeders. However, current knowledge and expected future research advances in this area offer exciting possibilities for selective high end breeding operations. Various assisted reproduction techniques carry some important advantages with regards to management of both fertile and subfertile stallions, allowing for treatment of the semen. For example, frozen and preserved semen can be shipped anywhere in the world, making valuable genetics available globally, and preserving genetics from stallions long after their death. Researchers in Australia have recently developed a semen extender that allows for storage of equine semen for an extended period of time at ambient temperature. This is without doubt a major breakthrough that can simplify breeding with shipped semen and reduce some hurdles associated with timing of the shipment in relation to expected ovulation. Research advances on semen biology has also resulted in improved management of stallions with specific and selective fertility problems. Stallions that produce ejaculates with poor sperm quality and viability can often be managed by centrifugation of their ejaculates and re-suspension of the sperm in an extender with supportive nutrients. Furthermore, the quality of an ejaculate can be enhanced through gradient centrifugation that can separate "good" and "bad" sperm before insemination. Further research to enhance our understanding of relevant sperm biology and advanced diagnostics to identify preferable sperm populations in an ejaculate is expected to create a need for robust and efficient technologies to select superior sperm within an ejaculate.

Research that makes it possible to separate x- and y-bearing sperm through flow cytometry (based on the fact that x-bearing sperm contains more DNA), has been commercialized and is currently offered by specialized laboratories. The method is very accurate, but efficiency is still quite low and results in a low number of sex-sorted sperm. This hurdle has been a limiting factor for the method and more research is needed to improve the technique. Recent research exploring the use of nanotechnology to target specific sperm DNA sequences has the potential to develop a new efficient technology to select sperm for sex-sorting, and maybe more importantly—to select

sperm based on other genetics that can be used as a replacement or complement to pre-implantation genetic testing of embryos. Preliminary results from studies using nanotechnology for sex-sorting in donkeys and other species appear to be promising. Logically, technologies that involve sperm selection in the laboratory will always result in limited number of sperm that can be used for breeding. However, recent scientific and clinical advances make it possible to use a single sperm from an infertile stallion and inject it into an egg that has been retrieved by ultrasound guided follicular aspiration from the ovary, culture the conceptus in a petri dish under controlled conditions and transfer the embryo into a recipient mare or back into the same oocyte donor mare at a later time. Intracytoplasmic sperm injection (ICSI) has become increasingly popular and a suitable method to use genetically valuable semen of poor quality or with restricted access because of disease or death of the stallion. Research to improve the efficiency of this technology would greatly benefit breeding operations that allow assisted reproduction.

Few, if any, breed registries accept foals resulting from somatic cell cloning of an existing individual. The technology is nevertheless offered for horses, and has become popular among some horse owners that don't need to register their foals. Although cloned horses have been used successfully as athletes, the greatest benefit of the technology may be the possibility to produce an intact stallion as a genetic copy of an existing gelding with a successful athletic career. It should be kept in mind that even if the genetic make-up of the cloned copy is identical to the original horse, the phenotype (looks and performance) may be different because of the influence of the environment that the embryo and fetus were exposed to in the test tube and uterus of the surrogate mother (epigenetics). The best prospective for the cloned intact stallion may therefore, be for breeding purposes. In other words—an expensive way to put testicles back on a gelding.

CONTACT:

Mats H.T. Troedsson, DVM, PhD, DACT, DECAR
 m.troedsson@uky.edu
 (859) 218-1133
 Maxwell H. Gluck Equine Research Center
 University of Kentucky
 Lexington, KY





Clostridial Myositis

Clostridial myositis is a rare but serious bacterial infection, which causes inflammation and death of muscle and release of bacterial toxins into the bloodstream. This condition is also referred to as myonecrosis, malignant edema, and gas gangrene. It occurs most often in horses that have recently received an intramuscular injection. Clinical signs appear 6-72 hours following the injection, and horses initially exhibit acute swelling, heat, and pain of the affected area. The disease progresses rapidly and the horse's condition may decline within hours. The affected animal exhibits signs of systemic toxemia; death can supervene rapidly in severe cases. Clostridial bacteria produce gas that results in a characteristic emphysematous (bubbly) feel or crepitation of the region. Clostridial myositis is a true medical emergency, with survival linked to prompt intervention through aggressive antibiotic treatment and wound debridement.

The *Clostridium* genus consists of over 150 known species of Gram-positive, anaerobic, spore-forming bacteria. The spore-forming ability of

these bacteria allows survival for long periods of time in the environment. When spores encounter a location without oxygen, such as damaged muscle, they are triggered to proliferate and produce exotoxins, which cause extensive tissue and vascular damage. The clostridial species that commonly cause myositis include *C. perfringens*, *C. septicum* and *C. chauvoei*.

Clostridial myositis has been reported following intramuscular inoculations of vaccines, ivermectin, antihistamines, phenylbutazone, vitamins, prostaglandins, and most commonly, flunixin meglumine. Infrequently, cases occur following inadequate perivascular administration of compounds, foaling trauma, or puncture wounds. In a study by Peek *et al* in 2003, stallions and Quarter horses were overrepresented, and the authors hypothesized this might be due to the heavy muscling of these groups.

The mechanism by which the bacterial spores arrive in the muscle of the horse is unknown. It is possible spores are introduced at the time



STEVE PATTON, UK AGRICULTURAL COMMUNICATIONS

Equine Disease Quarterly Newsletter

Department of Veterinary Science
Maxwell H. Gluck Equine Research Center
University of Kentucky
Lexington, Kentucky 40546-0099

Presorted Standard
US Postage Paid
Permit 51
Lexington KY

Address Service Requested

28 40 *****AUTO**MIXED AADC 450
Wisconsin State Horse Council
PO Box 72
Columbus WI 53925-0072



of injection. Another theory is that bacteria are translocated from their normal environment in the intestine, in times of inflammation or colic and arrive in the muscle via the bloodstream. No association linking whether or not the injection site is cleaned prior to injection and the development of myositis has been established. A higher incidence of myositis with irritating substances such as nonsteroidal drugs and vitamins is reported, potentially due to increased tissue damage and creation of an oxygen-free environment.

Diagnosis involves aspirating a small amount of fluid for anaerobic culture and Gram-staining to look for the presence of gram-positive rods. To treat the infection, large incisions are made into the muscle and fascia to expose the bacteria to oxygen and debride dead tissue. General supportive care is critical because these bacteria produce toxins that have secondary effects on the horse, including the potential to reduce the contractility of the heart. Clostridial toxins may also result in anemia, thrombocytopenia, and leukopenia. Horses are commonly treated with high doses of intravenous penicillin, intravenous fluids, cardiovascular sup-

port, and wound care. Hyperbaric therapy, where available, is suggested as an adjunct to routine treatment.

Survival has been reported to range from 31% to 73% and appears to be better for infections with *C. perfringens* compared with *C. septicum* or *C. chauvoei*. Horses that survive the initial toxemic stages of disease have an improved prognosis. The wounds created by a combination of infection and treatment are usually large, and may take weeks to months to heal entirely. Horses which do not survive show signs of intravascular coagulation and multi-organ failure.

There is no definitive prevention for clostridial myositis. When giving intramuscular injections, use large and well vascularized muscle groups, and when possible avoid giving irritating substances in the muscle if there is an alternative route such as oral or intravenous administration.

CONTACT:
Rebecca Ruby, BVSc (dist), MSc, DACVP
Rebecca.Ruby@uky.edu
(859) 257-8283
UK Veterinary Diagnostic Laboratory
University of Kentucky
Lexington, KY