EQUINE VIRAL ARTERITIS: HOW SIGNIFICANT A THREAT TO THE HORSE?

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ABSTRACT

Equine viral arteritis (EVA) achieved international notoriety following an extensive occurrence of the disease on numerous Thoroughbred breeding farms in Kentucky in 1984. Outbreaks of EVA have increased in number over the past 20 years, the majority being reported from North America and Europe. Aside from the ability of the aetiological agent, equine arteritis virus (EAV), to cause abortion and illness and death in young foals, the virus has been responsible for a number of extensive outbreaks of non-fatal disease at racetracks. Understanding the patterns of EAV shedding in acute and chronic phases of the infection is pivotal to establishing the modes of viral transmission. The most important means of spread at a racetrack is by aerosol transmission. The carrier state has only been demonstrated in the stallion, which sheds the virus solely in semen. Strategies are suggested for the prevention and control of EVA on breeding farms and at racetracks.

INTRODUCTION

For many years, equine arteritis virus (EAV) has been recognised as one of a triad of viral pathogens historically associated with the 'equine influenza-abortion' syndrome, the others being equine herpesvirus-1 and equine influenza virus (Timoney and McCollum 1993). In 1953, the virus was identified as the aetiological agent of equine viral arteritis (EVA), an acute contagious disease of equids (Doll *et al*. 1957). Subsequently, EAV was shown to be an RNA virus belonging to the genus *Arterivirus*, family *Arteriviridae* (Cavanagh 1997). Equine viral arteritis is principally characterised by fever, inappetance, depression, leukopenia, dependent oedema, especially of the limbs, scrotum and prepuce in the male and mammary glands in the female, conjunctivitis and, in a variable percentage of cases, respiratory signs and a skin rash. Infection of the pregnant mare can result in abortion or, if viral exposure occurs very late in gestation, the birth of a live but diseased foal, affected with a fulminant interstitial pneumonia or pneumo-enteritis (Timoney and McCollum 1993).

Equine viral arteritis (EVA) achieved little international prominence, much less notoriety, until the extensive outbreak on Thoroughbred breeding farms in Kentucky in 1984. That occurrence caused major concern over the potential for spread of EAV through the movement of horses (Timoney and McCollum 1993). Severe restrictions on the international trade in horses and equine semen with respect to this infection quickly followed, many of which are still in force.

OCCURRENCE AND DISTRIBUTION

The past 20 years have seen an increase in the number of recorded outbreaks of EVA, nearly all of which have been reported from North America and Europe. Factors contributing to this increase include greater awareness, improved monitoring and surveillance and enhanced laboratory capability to diagnose the disease. There is a sharp contrast between the global distribution of reported outbreaks of EVA, however, and the known geographic range of the causal virus, antibodies to which have been demonstrated in most equid populations in which they have been looked for, with the exception of Japan and Iceland. The prevalence of EAV infection varies widely among countries and frequently between breeds in the same country (Timoney and McCollum 1993). Available evidence indicates that most cases of EAV infection are asymptomatic.

Currently, greatest emphasis is placed on the importance of EVA occurring on breeding farms because of the potential risk of abortions, deaths in

young foals and establishment of the carrier state in stallions. Aside from its accepted significance for susceptible breeding populations, EAV can also cause outbreaks of non-fatal disease at racetracks, horse shows, riding establishments, sales and equine clinics (Burki and Gerber 1966; McCollum and Swerczek 1978; Scollay and Foreman 1993; Timoney and McCollum 1993). To date, such occurrences have been infrequent, representing a minority of known outbreaks of EVA. Over the course of the past 30 to 40 years, however, there have been a number of occasions where the virus has spread widely among large groups of closely congregated horses, resulting in very extensive occurrences of EVA. In fact, the first isolation of EAV in Europe was obtained during a major outbreak of the disease among a group of 400 remount horses at Bern, Switzerland in 1964 (Burki and Gerber 1966). Epizootics of EVA occurred at 2 Standardbred racetracks in Kentucky in 1977, involving several hundred horses (McCollum and Swerczek 1978). The majority of the horses at the Red Mile Racetrack in Lexington became infected, with most of them displaying some clinical evidence of EVA. There have been 4 other widespread occurrences of the disease at racetracks, all in North America and all of them involving hundreds of clinically affected horses. EVA occurred among Thoroughbreds and Standardbreds at tracks in Edmonton and Calgary in Alberta, Canada in 1986, from which it spread to local breeding farms where it was responsible for outbreaks of virus-related abortion. In 1989, extensive outbreaks of EVA took place at racetracks in Nebraska and Iowa, which involved Arabians, Thoroughbreds and Quarterhorses. The most recent major occurrence of EVA was at Arlington International Racecourse, Illinois, in 1993, during which more than 10% of the Thoroughbreds stabled at the track became affected with the disease (Scollay and Foreman 1993). The outbreak had significant repercussions on international participation in the Arlington Million, the feature race of that particular meeting. Experience has shown that, given the appropriate circumstances, the potential certainly exists for widespread transmission of EAV among a susceptible population of closely congregated horses.

EPIDEMIOLOGY

A range of factors, virus-, host-, and environment-related, are known to be involved in the epidemiology of EVA (Timoney and McCollum 1993). Those considered of greatest significance include: variation in pathogenicity and other phenotypic properties among naturally occurring strains of EAV; modes of virus transmission during acute and chronic phases of the infection; occurrence of the carrier state in the stallion (Timoney et al. 1986); nature and longevity of acquired immunity to infection; and economic trends within the horse industry. Outbreaks of EVA are most frequently associated with the movement of horses or the use of virus infective semen (Timoney and McCollum 1993).

Viral pathogenicity

Variation in pathogenicity is known to exist among field strains of EAV. Viral isolates have been categorised as lentogenic, mesogenic and velogenic based on experimental infectivity studies (Timoney and McCollum 1993) and growth characteristics in equine endothelial cells (Moore et al. 2002). While many carrier stallions shed virus of low pathogenicity, some shed variants of EAV capable of causing clinical disease and outbreaks of EVA.

Modes of transmission

The principal modes of EAV transmission are by the respiratory route in the case of the acutely infected horse and via the venereal route by the acutely or chronically infected stallion (Timoney and McCollum 1993). Following an incubation period, usually of 3 to 8 days duration, large quantities of virus are shed into the respiratory tract of the acutely infected horse for 7 to 16 days. Transmission of infection takes place through direct contact with infective, aerosolised respiratory secretions. This is the principal means of spread of EAV during widespread outbreaks of EVA, for example, at racetracks, or wherever horses are closely congregated (Timoney and McCollum 1993). Furthermore, it is an important mode of dissemination of virus on breeding farms. During the acute phase of the infection, EAV is also shed, but in lesser concentrations, in conjunctival secretions, urine, faeces, vaginal secretions and semen.

Of additional importance on breeding farms is venereal transmission of the virus by the acutely or chronically infected stallion (Timoney and McCollum 1993). EAV can also be spread by this route through artificial insemination of mares with infective, fresh-cooled or frozen semen. Exposure of the pregnant mare can result in *in utero* transmission of the virus to the unborn foal. In such cases, the placenta and placental fluids, together with foetal tissues and fluids are usually productive sources of EAV.

Although not considered of major epidemiological significance, EAV can be spread either on breeding farms, at racetracks, or other equine establishments through indirect contact with virus-contaminated fomites, such as tack or other equipment shared between horses (Timoney and McCollum 1993). Also, personnel coming in contact with infected animals may inadvertently act as mechanical carriers of the virus on their hands or outer apparel.

Carrier state

Chronic infection with EAV or the carrier state has been demonstrated only in the stallion (Timoney *et al.* 1986) or pubertal colt and not in the mare, gelding or sexually immature colt (Timoney and McCollum 1993). The carrier state is testosterone dependent, with the virus localised to certain of the accessory sex glands in the reproductive tract (Little *et al.* 1992). Persistently infected stallions shed EAV constantly in semen, but not in any other bodily secretion or excretion (Timoney and McCollum 1993). Under appropriate conditions of management, carrier colts or stallions do not represent a risk of transmitting the infection to susceptible, in-contact horses in a racetrack setting. Persistence of EAV in the stallion does not appear to have any adverse effect on its clinical condition nor on its fertility (Timoney and McCollum 1993). The carrier stallion is considered the primary reservoir of EAV and of major epidemiological significance in enabling the virus to persist in equine populations around the world.

Immunity

Infection with EAV stimulates a strong protective immunity against EVA that is long lasting (Timoney and McCollum 1993). Neutralising antibodies to EAV have been shown to persist for up to 3 years and longer after exposure to the virus. Vaccination can also stimulate an immune response that is protective against development of clinical disease and establishment of the carrier state in the stallion. Some protection is conferred as early as 4 days after vaccination and protection is nearly complete after 10 days. Vaccinated horses may, upon natural exposure to the virus, experience a limited reinfection cycle. At the present time, only 2 commercial EVA vaccines are available, one an attenuated, modified live virus vaccine and the other, an inactivated adjuvanted product (Timoney and McCollum 1993). Serological responses following primary vaccination with the modified live virus vaccine are markedly enhanced by re-vaccination, with the development of high neutralising antibody titres that remain relatively undiminished for a year or longer (Timoney 2000a). The inactivated vaccine also stimulates an immune response against EVA. This tends to be less pronounced, however, than that observed in horses vaccinated with the modified live virus product. There are no published data on the duration of immunity conferred by the commercial inactivated vaccine.

Foals born of mares that have been immunised through natural exposure or vaccination against EVA are protected against the disease if they receive colostral-derived antibodies (McCollum 1976). This passively acquired protection, which lasts for 2 to 5 months, will block stimulation of the immune response by vaccination while the foals remain seropositive.

Industry developments

Of the various developments that have taken place in the major horse breeding, racing and competition countries in the world over the past 30 to 40 years, most of them driven by economics, 2 have had a significant impact on the expanded global distribution of EAV. The first is the considerable growth in the volume of international movement of horses for performance or breeding purposes and the second is the significant increase in the volume of frozen semen being shipped internationally (Timoney 2000b).

International representation at prestigious racing events is not without risk of EAV being introduced by a horse from overseas, either incubating the infection or subclinically infected with the virus. In sharp contrast to past experiences with other respiratory-borne viruses, especially equine influenza, there have been no recorded instances to date where outbreaks of EVA at racetracks have been traced to such a source. Though reassuring, this should not give rise to complacency. Continued growth in the number of lucrative racing events held annually around the world will increase the risk of a disease outbreak at one of these venues at some point in the future, unless the necessary safeguards are in place to prevent or minimise this happening.

Imported carrier stallions have been implicated in introducing EAV into indigenous equine populations previously free of the infection and in causing outbreaks of economically-damaging disease. The second major factor involved in the dissemination of EAV has been the use of transported frozen semen by most of the major horse breed registries. This has resulted in a significant increase in the quantity of frozen equine semen being shipped internationally. There have

been instances where countries have inadvertently imported EAV-infective semen which subsequently has been responsible for outbreaks of EVA.

DIAGNOSIS

Although EVA may be suspected in a horse displaying the clinical signs characteristic of the disease, a diagnosis must be confirmed by carrying out the appropriate laboratory test(s) (Timoney and McCollum 1993). Clinically, a number of other infectious and non-infectious equine diseases can mimic the symptomology of EVA. Of particular importance from a differential diagnostic point of view are equine herpesvirus-1 and -4 infections, equine influenza, purpura haemorrhagica, equine infectious anaemia, dourine, African horse sickness fever, Getah virus infection and toxicosis due to hoary alyssum (*Berteroa incana*).

Confirmation of a diagnosis of EAV infection is based on virus isolation, detection of viral nucleic acid or antigen and/or demonstration of a specific antibody response by testing paired (acute and convalescent) sera collected at a 21 to 28 day interval in the virus neutralisation test or an appropriately validated ELISA (Timoney 2000a). Specimens of choice for virus isolation /nucleic acid detection by the reverse-transcription polymerase chain reaction (RT-PCR) assay from the acutely infected horse include nasopharyngeal swabs or washings, conjunctival swabs and unclotted blood using ethylene-diamine-tetraacetic acid or sodium citrate as preferred anticoagulants. Swabs should be placed in an appropriate viral transport medium and kept refrigerated or frozen during transport to the laboratory.

Detection of the carrier state in a stallion involves initially taking a blood sample to determine the individual's serological status for antibodies to EAV. Only seropositive stallions, ie those having a neutralising antibody titre of 1:4 or greater, without a certified history of vaccination against EVA, need to be considered potential carriers of the virus. The carrier state has never been recorded in a seronegative stallion (Timoney and McCollum 2000). Semen containing the sperm-rich fraction of the ejaculate should be collected from any suspect stallion and screened for the presence of EAV either by attempted virus isolation in cell culture or by the RT-PCR assay. Use of this testing procedure for detection of the carrier state, which is described in detail by Timoney (2000a), is based on the fact that persistently infected stallions shed EAV constantly in semen (Timoney and McCollum 1993, 2000). Reliability of the screening procedure is dependent upon the quality of the semen sample submitted for examination and on the competence and experience of the laboratory conducting the test. It is very important to select a laboratory with a proven track-record of proficiency in screening stallions for this infection. Although no longer commonly in vogue, presence of the carrier state can also be determined by test breeding a stallion to at least 2 seronegative mares and monitoring the latter for seroconversion to EAV for up to 28 days after breeding (Timoney and McCollum 1993).

PREVENTION AND CONTROL

Breeding farms

Current control programmes are directed at curtailing the spread of EAV in breeding horse populations to prevent outbreaks of virus-related abortion and/or losses in young foals, and to minimise the risk of establishment of the carrier state in stallions (Timoney and McCollum 1993; Timoney 2002). These programmes are predicated on the epidemiological importance of the carrier stallion in maintaining the virus in breeding herds. EVA can be controlled effectively on breeding farms through observance of sound management practices similar to those recommended for the prevention of equine herpesvirus and other respiratory virus infections, and through a programme of selective vaccination (Timoney and McCollum 1993). Thanks to the availability of safe and effective vaccines, it has been possible to protect against this disease. It is recommended that all stallions used for breeding purposes be re-vaccinated annually against EVA to obviate the risk of establishment of the carrier state. For the same reason, it is also advisable to vaccinate colts between 6 and 12 months of age. In light of the risk of introducing EAV onto a breeding farm by means of infective shipped semen, it is important to establish the infectivity status of semen used for artificial insemination, especially if it has been imported.

Racetracks

Despite the potential economic consequences of a widespread outbreak of EVA, as yet there are no formal programmes specifically directed at preventing the introduction of EAV at racetracks. In large measure, this is probably reflective of the perceived low risk of the disease occurring at racetracks and other equine performance events.

In the absence of any clinical evidence suggestive of EVA, there is no practically feasible

means of currently detecting horses arriving at a track which are acutely infected with EAV. The problematic animal is the one incubating the infection or subclinically infected with the virus. Depending on the numbers involved, implementation of racetrack testing of individual horses for this infection could be logistically burdensome and excessively costly. Based on the rarity of track outbreaks, such a programme would be very difficult to justify. Of proven value in helping to reduce the risk of EVA and other respiratory-borne equine infectious diseases being introduced, is to require that all horses arriving at a racetrack be accompanied by a certificate of veterinary inspection issued within the previous 3 days. This should include a declaration of non-exposure to animals known to be affected with a contagious disease within a specified time frame (Knowles 1994).

In the case of international race fixtures involving horses imported from abroad, it is certainly possible to reduce considerably, if not eliminate the risk of introducing EAV by requiring a period of quarantine with or without laboratory testing for evidence of virus shedding.

Vaccination

It could be argued that the best defence against a possible racetrack outbreak of EVA would be to recommend widespread vaccination against the disease. While such a strategy would probably succeed in protectively immunising the population at risk, it would not be considered an acceptable option by most racing industries at the present time. It should be pointed out, however, that vaccination with a commercial modified live virus product has been used successfully in the past to curtail the transmission of EAV during several extensive outbreaks of EVA in N. America, without any adverse consequences (Timoney and McCollum 1993; Timoney 2000a). Vaccination helped bring such occurrences under effective control within 7 to 10 days (Timoney 2000a).

There is little doubt that the risk of spread of EAV and various other equine pathogens and of outbreaks of EVA on breeding farms and at equine performance events will increase with continued growth in the international movement of horses for racing, breeding or other purposes.

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REFERENCES

Burki, V.F. and Gerber, H. (1966) Ein virologisch gesicherter grosausbruch von equiner arteritis. *Berl Munch Tierarztl Wochenschr.* **79**, 391-395.

Cavanagh, D. (1997) Nidovirales: a new order comprising *Coronaviridae and Arteriviridae. Arch Virol.* **142**, 629-633.

Doll, E.R., Bryans, J.T., McCollum, W.H., and Crowe, M.E.W. (1957) Isolation of a filterable agent causing arteritis of horses and abortion by mares. Its differentiation from the equine abortion (influenza) virus. *Cornell Vet.* **47**, 3-40.

Knowles, R. (1994) Guidelines for EVA. In: *Equine Infectious Disease: Developing an Action Plan*, Ed: D.G. Powell, Lexington, University of Kentucky. pp 37-39.

Little, T.V., Holyoak, G. R., McCollum, W. H., and Timoney, P. J. (1992) Output of equine arteritis virus from persistently infected stallions is testosterone-dependent. *Proc. 6th int. Conf. on Equine Infectious Diseases.* Eds: W. Plowright, P.D. Rossdale and J.F. Wade, R&W Publications, Newmarket pp 225-229.

McCollum, W.H. (1976) Studies of passive immunity in foals to equine viral arteritis. *Vet Microbiol.* **1**, 45-54.

McCollum, W.H. and Swerczek, T.W. (1978) Studies of an epizootic of equine viral arteritis in racehorses. *J. equine Med Surg.* **2**, 293-299.

Moore, B.D., Balasuriya, U.B.R., Hedges, J.F., and MacLachlan, N.J. (2002) Growth characteristics of a highly virulent, a moderately virulent and an avirulent strain of equine arteritis virus in primary equine endothelial cells are predictive of their virulence in horses. *Virology* **298**, 39-44.

Scollay, M.C. and Foreman, J. H. (1993) An overview of the 1993 equine viral arteritis outbreak at Arlington International Racecourse. *Proc. Am. Ass. equine Pract.* **39**, pp 255-256.

Timoney, P.J. (2000a) Equine viral arteritis. In: *O.I.E. Manual of Standards for Diagnostic Tests and Vaccines.* Office International des Epizooties, Paris. pp. 582-594.

Timoney, P.J. (2000b) Factors influencing the international spread of equine diseases. *Vet. Clin. N. Am. equine Pract.* Ed: P.J. Timoney, W.B. Saunders, Philadelphia. pp 537-551.

Timoney, P. J. (2002) Equine viral arteritis. In: *Equine Respiratory Diseases*, Ed: P. Lekeuk, International Veterinary Information Service (www.ivis.org). Document No. B0324.0402

Timoney, P.J. and McCollum, W.H. (1993) Equine viral arteritis. *Vet. Clin. N. Am. equine Pract.* **9**, 295-309.

Timoney, P.J. and McCollum, W.H. (2000) Equine viral arteritis: further characterization of the carrier state in stallions. *J. Reprod. Fertil. Suppl.* **56**, 3-11.

Timoney, P.J., McCollum, W.H., Roberts, A.W., and Murphy, T.W. (1986) Demonstration of the carrier state in naturally acquired equine arteritis virus infection in the stallion. *Res. vet. Sci.* **41**, 279-280.