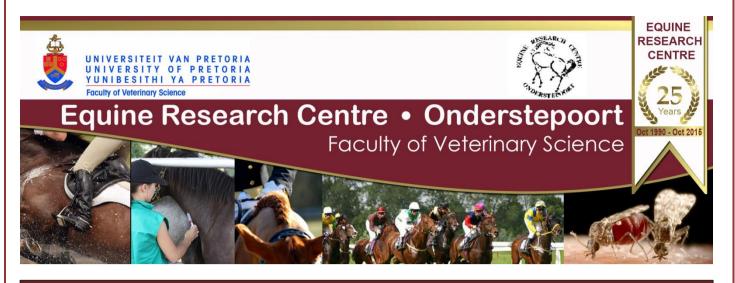
Equine Research Centre News Update – May 2016



EQUINE RESEARCH ... what you need to know

Brought to you by the Equine Research Centre, University of Pretoria

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COMPLETE GENOME SEQUENCES OF THE FOUR AHS VIRUS STRAINS FROM A COMMERCIAL TETRAVALENT LIVE ATTENUATED VACCINE

In our last Newsletter we reported on the groundbreaking study and report of the complete genome sequences of the three virus strains included in Bottle I of the OBP AHS Vaccine. We said that a study was now underway on the four virus strains included in Bottle II of this live attenuated vaccine. Here are the findings of this study :

African horse sickness (AHS) is caused by AHS Virus (AHSV), the genome of which is comprised of ten segments. In South Africa, a polyvalent AHS attenuated live virus (ALV) vaccine is manufactured by Onderstepoort Biological Products (OBP) Ltd. This vaccine is supplied in two separate vials each of which contains different AHSV types: Bottle I is trivalent and contains serotypes 1, 3 and 4 while Bottle II is tetravalent (x 4) and contains serotypes 2, 6, 7 and 8. This study is focused on Bottle II.

The research team studied the full genome sequences of AHSV2, AHSV6, AHSV7 and AHSV8 which were isolated from Bottle II of the AHS-ALV vaccine. The individual serotypes were independently isolated, using plaque selection. Each of these viruses was then passaged and AHSV dsRNA was extracted. Sequence reads were analysed followed by mapping to obtain full-length genome sequences of the four viruses.

The genome sequences of AHSV serotype 2 and serotype 6 viruses, which are precursors of the respective AHSV-ALV virus strains, are available from GenBank. The whole genome sequences of the AHSV-2 and AHSV-6 vaccine strains were more than 99.350% identical to that of the precursors of these strains.

The genome sequences of an AHSV7 strain with a truncated VP2 protein (AHSV7-tVP2) are available from GenBank. The sequence identity of AHSV-7 derived from OBP Vaccine and AHSV7-tVP2 was more than 99.3% for all segments except those encoding the VP5 and non-structural 2 (NS2) proteins, which were lower at 77.4% and 97.9%, respectively. Our analysis showed that the AHSV7 vaccine strain was a reassortant comprising 8 segments from AHSV7-tVP2 and 2 segments from an AHSV6 strain.

The genome sequences of AHSV serotype 8 are available from GenBank. The sequence identity of the AHSV8 vaccine strain for four of the ten segments was >99.9%, while that for the remaining segments was between 76.8% and 97.4%. Analysis showed that AHSV-8 vaccine strain is a reaaortant which includes segments from both AHSV8 and AHSV5.

<u>Comment</u> : This paper further supports the previous paper on Bottle I, and that when outbreak investigations are conducted, it can be categorically determined whether a field virus or a vaccine related virus is involved. These findings therefore provide the basis for investigations into any future outbreaks, and the potential role of vaccine viruses. They also highlight the importance of vaccinating before midge season to minimise the risk of the virus being transmitted.

Publication : Genome Announcements

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DRAFT GENOME SEQUENCE OF *TAYLORELLA EQUIGENITALIS* ISOLATED FROM THE SEMEN OF A LIPIZZANER STALLION IN SOUTH AFRICA

Taylorella equigenitalis is the agent of contagious equine metritis (CEM), a highly contagious sexually transmitted infection of horses, characterised in infected mares by vaginal discharge and various degrees of vaginitis, cervicitis and endometritis, and it may result in temporary infertility or embryonic death.

In stallions, the long-term presence of *T. equigenitalis* does not cause clinical signs, and asymptomatic carrier mares have also been reported. CEM is a World Organisation of Animal Health (OIE)-notifiable disease and is considered part of veterinary certification for international trade purposes. The multilocus sequence typing scheme for taylorellae recently provided a comprehensive overview of the genetic diversity of taylorellae. To date, the genome sequences of only four *T. equigenitalis* strains have been reported, and the genome sequences of most *T. equigenitalis* strains have been reported, and the genome sequences of most *T. equigenitalis* sequence types (STs) remain to be characterised.

This report is of the genome sequence of *T. equigenitalis* ERC_G2224, which was isolated in 2015 from a stored frozen semen sample collected in 1996 from an asymptomatic carrier Lipizzaner stallion from a property in Gauteng. Sequence typing of this strain revealed its membership in the previously non-sequenced ST4, which is not linked to one of the existing clonal complexes. These data will help clarify the source of the CEM strain involved in the South African outbreak.

Publication : Genome A Journal

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CULICOIDES SPECIES ABUNDANCE AND POTENTIAL OVER-WINTERING OF AFRICAN HORSE SICKNESS VIRUS IN THE ONDERSTEPOORT AREA, GAUTENG, SOUTH AFRICA

In South Africa, outbreaks of African horse sickness (AHS) occur in summer; no cases are reported in winter, from July to September. The AHS virus (AHSV) is transmitted almost exclusively by *Culicoides* midges, with *Culicoides imicola* being the most prevalent vector. The over-wintering mechanism of AHSV is unknown. In this study over 500 000 *Culicoides* midges, consisting of at least 26 species, were collected in light traps at weekly intervals between July 2010 and September 2011, near horses in the Onderstepoort area. The study found that, despite low temperatures and frost, at least 17 species, including *C. imicola* were collected throughout winter, albeit in much smaller quantities (< 100) than in March (>50 000) – there were no midge-free periods. Even in below freezing conditions (-4.5°C), with frost occurring, *Culicoides* midges were present, with the presence of males in addition to females indicating that breeding continued throughout winter. However, this study showed that the rate of infection correlated to high numbers of midges. So, although no virus was detected during the winter period, continuous adult activity indicated that transmission can potentially occur. Therefore cases of AHS in susceptible animals could start as soon as *Culicoides* populations reach a critical level.

Publication : Journal of the South African Veterinary Association

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DETECTION OF EQUINE HERPESVIRUS-4 AND PHYSIOLOGICAL STRESS PATTERNS IN YOUNG THOROUGHBREDS CONSIGNED TO A SOUTH AFRICAN AUCTION SALE

Background

The prevalence of equine herpesvirus types-1 and -4 (EHV-1 and -4) in South African Thoroughbreds at auction sales is currently undefined. Commingling of young Thoroughbreds from various populations together with physiological stress related to their transport and confinement at a sales complex, may be associated with shedding and transmission of EHV-1 and -4.

This study sampled 90 young Thoroughbreds consigned from eight farms, originating from three provinces representative of the South African Thoroughbred breeding demographic to a sales complex. Nasal swabs for qPCR tests to detect EHV-1 and -4 nucleic acid were collected from all sample horses on arrival and departure. Additional nasal swabs for qPCR were obtained from those horses displaying high temperature and/or nasal discharge. Daily faecal samples were used to determine faecal glucocorticoid metabolite (FGM) concentrations as a measurement of physiological stress.

Results

EHV-4 nucleic acid was detected in 14.4% of the horses, while EHV-1 was not detected in any of the horses in the study population. Of this population 93.3% showed antibodies indicating prior exposure to EHV-4, and only 1.1% showed prior exposure to EHV-1. High temperatures and nasal discharge did not necessarily mean that the horses had either EHV-1 or -4.

The horses' FGM concentrations increased following arrival, and then decreased again for most of the remaining study period including the auction process.

Conclusion

In this study population, there was evidence that the sales process could result in carriers shedding and transmitting EHV-4 despite most horses having been exposed to EHV-4 prior to the sale. There was no evidence of EHV-1 shedding or transmission at the sales despite very few horses having been exposed to EHV-1 prior to the sale. However, the physiological stress response shown by most horses reflected the combination of stressors associated with transport and arrival and these are key areas for future investigation into management practices to enhance health and welfare of young Thoroughbreds during sales consignment.

Publication : BMC Veterinary Research

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THE CARRIER PREVALENCE OF SEVERE COMBINED IMMUNODEFICIENCY, LAVENDER FOAL SYNDROME AND CEREBELLAR ABIOTROPHY IN ARABIAN HORSES IN SOUTH AFRICA

The reason for performing this study was to determine the carrier prevalence of severe combined immunodeficiency (SCID), lavender foal syndrome (LFS) and cerebellar abiotrophy (CA) in Arabian foals in South Africa, in order to quantify the potential impact of these conditions locally. Furthermore, the carrier prevalence of SCID prior to and following the introduction of a genetic test was compared to evaluate the effect of testing in the population.

The objective was to compare the changes in prevalence in these disorders between two groups of registered purebred Arabian foals, born in South Africa in the 2004/5 and 2009/10 foaling seasons.

Severe combined immunodeficiency (SCID) of Arabian horses is a primary immunodeficiency disorder which results in a profound susceptibility to infections and death before the age of 6 months. The carrier prevalence of SCID in Arabian horses varies between 8.4% in the USA, 1-5% in the UK, 7% in Morocco, 1.5% in Brazil and 0% in Poland, Slovenia and Iran.

Lavender foal syndrome (LFS) is a lethal disorder of Arabian foals, characterised by a dilute coat colour and a range of neurological signs, including lying down, severe spasm in which the back arches, paddling movements and rigidity. The carrier frequency of LFS in the USA is estimated to be 10.3% in Egyptian Arabian and 1.8% in non-Egyptian Arabian populations.

Cerebellar abiotrophy (CA) is a progressive neurological disorder, for which the clinical signs usually develop between the ages of 6 weeks and 4 months, and include lack of control of voluntary muscle movements, vision problems and head tremors.

The development of genetic tests for these conditions has provided means of identifying carriers of these disorders. This enables breeders to manage breeding programmes and thereby prevent the birth of affected offspring. Determination of the prevalence of carriers of such disorders in a population allows quantification of the potential impact of the disease on a population. The carrier prevalence of SCID, LFS and CA has not previously been determined in Arabian horses in South Africa.

For the 2004/5 foals, a total of 349 stallions were used, while 378 stallions were used during the 2009/10 foaling season. The carrier prevalence of LFS and CA among foals born during the 2009/10 foaling was 11.7% and 5.1% respectively, with no significant change between 2004/5 and 2009/10. However, the prevalence of SCID was found to have decreased significantly, from 6.4% in 2004/5 to 3.4% in the 2009/10 foals.

Lavender foal syndrome of Arabian horses is most commonly observed in Egyptian Arabians, and the influence of the Egyptian Arabians in South Africa may account for the higher carrier frequency. On the other hand the prevalence of CA, at 5.1% is lower, suggesting that fewer carriers of CA have been used as popular sires in South Africa. The marked decrease in the prevalence of SCID since the introduction of a genetic test for this disorder in 2005 is possibly a positive outcome of the testing. Complete exclusion of carriers from breeding programmes hastens removal of a disease from the population. However, this practice may result in shrinkage of the available gene pool.

This study did not address specific lines or the influence of founder animals or popular stallions, which may warrant further study.

Publication : Equine Veterinary Journal 46

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