An epidemiology report detailing the investigation of EVA virus disclosed on a premises in Shropshire, July 2019

September 2019
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APHA is an Executive Agency of the Department for Environment, Food and Rural Affairs and also works on behalf of the Scottish Government, Welsh Government and Food Standards Agency to safeguard animal and plant health for the benefit of people, the environment and the economy.
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Introduction

Background

Equine viral arteritis (EVA) is occasionally detected in the UK in stallions which are tested for pre-breeding or for export purposes. The last confirmed cases were on premises in Devon and Dorset in 2019; prior to these, the last confirmed case was in 2012. Under the EVA Order (1995) the disease is notifiable in all stallions and in mares that have been mated or inseminated in the 14 days prior to testing positive. It is not notifiable in non-breeding mares or geldings.

Infection with equine arteritis virus (EAV) was confirmed by PCR testing of a semen sample from a pony stallion resident on premises in Shropshire.

The infection was initially disclosed as a result of routine private pre-breeding ELISA-based serology testing, as recommended by the Horserace Betting Levy Board (HBLB) Code of Practice for EVA. These tests returned non-negative results on two separate occasions in a private laboratory, along with a positive semen sample tested by PCR, which were subsequently notified to APHA by the private veterinary surgeon as required under the EVA Order 1995. Official samples submitted by APHA confirmed the seropositive results and that the semen tested positive with two different PCR tests and genetic sequencing. The Deputy CVO confirmed disease on 31 July 2019.

Clinical signs suspicious of acute EVA infection in the single positive stallion were not observed or reported by the private veterinary surgeon, nor were any signs suspicious of acute EVA infection reported in the other horses on the affected premises during the preceding 12 months.

At the time of the official APHA veterinary inquiry, the positive stallion was observed to be in an apparently good state of general health, with a rectal temperature within normal limits. He did demonstrate a degree of increased bilateral lacrimation (facial tear-staining) and some nasal discharge. However, it was considered that these signs could be equally attributable to a number of other factors and it was reported that he is particularly sensitive to fly nuisance.

The only other relevant clinical history relating to the positive stallion was his treatment by a veterinary practice in Suffolk at the beginning of 2019 for dentistry and investigation of lameness, with treatment for a back problem.

Epidemiological investigations found no evidence to suggest that the Shropshire case is in any way connected with the previous confirmed cases of EVA in Devon and Dorset during
March and April of 2019 (i.e. no evidence of any direct or indirect links, including any common attendance by any horses from the respective premises at competitive events).

The results of epidemiological investigations are further supported by laboratory sequence analysis which has been able to rule out any evidence of a direct transmission pathway linking the current positive stallion and any of the stallions involved in the Dorset and Devon outbreaks.

**Description of the premises**

The premises is a private yard and currently contains five horses, all owned by the owners of the premises.

Horses are housed in stables overnight during the winter months, but whenever possible are kept outdoors at pasture at other times.

Details of the horses currently resident on the premises are provided in Table 1.

**Table 1: Details of horses currently on the premises**

<table>
<thead>
<tr>
<th>Horse number</th>
<th>Sex</th>
<th>Age</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>Mare</td>
<td>10 years</td>
<td>Arrived on the premises in 2017. Originally from Germany.</td>
</tr>
<tr>
<td>3</td>
<td>Colt</td>
<td>6 months</td>
<td>Offspring of horses 1 and 2.</td>
</tr>
</tbody>
</table>
Nine other horses (six geldings and three mares) have departed from the premises since January 2018.

The positive stallion (horse 1) is kept separate from other resident horses. He has his own stable (albeit that it is at the end of a row of stables used by the other resident horses) and when at grass he has his own paddock separated from the other horses by an empty paddock and an electric fence.

However, he did manage to serve one of the resident mares (horse 2) in March of 2018 and she subsequently gave birth to a healthy foal, which remains on the premises.

Other than this isolated incident no other breeding, or semen collection for use in artificial insemination, has taken place on the premises for a number of years.

Horses from the premises regularly participate in competitive events within Great Britain.

**Overview of biosecurity**

The premises are considered to be well run and are kept in a very clean and tidy state. However, levels of biosecurity were consistent with those encountered in similar enterprises. Advice on measures to improve biosecurity were provided following disclosure of the positive stallion.

**Vaccination**

The owners do not have a current EVA vaccination protocol in place, and are not aware that any of the resident horses have ever previously been vaccinated against EVA. Checks of passports of four of the resident horses showed no evidence of EVA vaccination. However, the passport of the confirmed positive horse was not available for inspection at the time of the epidemiological investigation as it had accompanied him to a private veterinary surgery where he underwent castration on 07 August 2019.
Breeding practices

The owners previously kept eventing mares on the premises that were also used for breeding. Mares would travel to an AI centre for insemination and would then return to the premises. Normal practice would be for them to return to the AI centre for foaling. The last foal to be born in this system was born eight years ago, although she was born early and was delivered on the premises.

None of the mares previously used for breeding in this system remain on the premises.

The EVA positive stallion is considered to have excellent blood lines and the owners had therefore begun to consider using him for breeding, which had initiated the pre-breeding EVA testing (in accordance with the HBLB Codes of Practice) and led to infection being detected by private testing, and subsequently confirmed by official testing of serum and semen.

Following confirmation of EVA, the owners voluntarily had the positive stallion castrated on 07 August 2019, thus effectively eliminating the risk of further onward transmission of disease to other horses.

Movements

As previously described, horses from these premises regularly participate in competitions within Great Britain, with one historic report of attendance at an event in the Netherlands in 2006. A number of events had been attended, both by currently resident (including the positive stallion) and recently departed horses, since January 2018.

All such events were single day attendance with no overnight stabling involved, with the single exception of one event attended by a gelding, which was subsequently sold onwards. The positive stallion did not attend any events involving overnight stays.

However, the gelding referred to above and the positive stallion were consigned to a yard in Suffolk for schooling for a number of months spanning 2018 to 2019, with the intention for them to be subsequently marketed for onward sale.

Only the gelding was subsequently sold and the positive stallion returned to the affected premises in the spring of 2019. Whilst resident on the premises in Suffolk, the positive stallion received veterinary treatment on two occasions in early 2019 from a local veterinary practice (dentistry and investigation of lameness/treatment for a back problem respectively). However, he did not attend any events during this period.
**Imports**

There is no recent history of horses being imported onto the premises from outside Great Britain. Other than the foal born in January 2019 (the result of the positive stallion mating with a resident mare) no horses have been introduced for a period of approximately 18 months.

The positive stallion was imported in 2012 from premises in the Netherlands and was not tested, either as a pre-export voluntary measure, nor sampled post-import. The Netherlands experience occasional cases of EVA, and three previous cases detected in the UK, one in 2004 and two in 2010 were also in horses originating from the Netherlands.

**Visiting horses**

Horses that have moved off will occasionally come back for specific reasons, but these are reported to be kept separate from resident horses.

For example, one gelding was initially relocated to premises in the south west of England but then had a prospective buyer from the north of England. All relevant parties met at the affected premises, which served as a half-way point, so that the prospective buyer could try him out.

However, horses do not routinely return for further training.

**Description of surrounding area and other known susceptible livestock on locality**

The owners report that there are no other horses in the immediate vicinity and therefore direct contact with horses on neighbouring premises is not possible. Observations during the epidemiological investigation are supportive of this.

**EVA testing on the premises**

In addition to the pre-breeding testing that initially disclosed the positive stallion (horse 1) the other four horses on the premises also subsequently underwent private testing for EVA on two separate occasions two weeks apart. A gelding (horse 4) returned seropositive results on both occasions.

The remaining three horses, including the mare (horse 2) that was mated by the positive stallion in March 2018, remained seronegative on both occasions.

The results of private testing are provided as Annex 1.
In addition to confirming EVA in the positive stallion, official testing by APHA also endorsed the results of private testing of the mare (horse 2) and gelding (horse 4).

The results of official testing are provided as Annex 2.

The results of private testing of the positive stallion on three separate occasions and of the seropositive gelding on two separate occasions indicated the presence of a stable (potentially declining) antibody titre in the stallion, and a stable antibody titre in the gelding (albeit with the caveat that samples were tested on different occasions and so a small degree of inter-assay variation could exist).

**Hypotheses for the source of infection**

**Most likely sources of infection**

EVA can be transmitted through a respiratory route between horses in direct and close contact, through sexual transmission and through fomites (contaminated equipment).

EVA is known to be present in several other EU Member States and there are no requirements under trade rules for animals to be vaccinated, or pre-movement tested, prior to entry into the UK, although vendors and purchasers may agree to private testing.

There is a history of horses on the affected premises (both currently resident horses and horses that have recently left the premises) originating from other countries (Germany, Ireland and the Netherlands) including the positive stallion that was imported from the Netherlands in 2012.

Horses from the affected premises, including the positive stallion and the seropositive gelding, also have a history of regular attendance at a number of competitive events within Great Britain and the positive stallion also spent a number of months between 2018-2019 on premises in Suffolk, all of which could potentially have represented sources of infection via direct or indirect contact with another undetected but infected (and infectious) horse.

**Estimated date of introduction of disease onto the premises**

Current working hypotheses for the source of infection for these premises include:

1. **The positive stallion was already infected when imported from the Netherlands in 2012.**
   
   There is existing APHA evidence of stallions from the Netherlands previously being imported and subsequently testing positive for EVA (October 2004, June
2010 and September 2010). It is recognised that between 10-70% of stallions acutely infected with EVA can develop a persistent infection and subsequently continuously shed the virus in their semen (Balasuriya et al 2016 and references contained therein). However, although whilst not conclusive, a degree of reassurance that the stallion was not actively shedding virus in his semen in March 2018, when he mated the mare, may be provided by the fact that the mare remains seronegative after repeat testing. It is considered that 85-100% of seronegative mares bred to persistently infected stallions will become infected and demonstrate seroconversion within 28 days (Balasuriya 2014).

2. **The positive stallion was infected whilst resident on the current premises by a horse that is still present or one that has since left.**

Given the absence of evidence of further transmission of disease to other horses on the premises this is considered to be a less likely hypothesis. The serologically positive gelding may have been previously exposed to infection before he arrived, may have been infected by the positive stallion whilst on the premises, or may have been exposed to infection at an event attended.

3. **The positive stallion was infected whilst competing at an event elsewhere within Great Britain.**

4. **The positive stallion was infected whilst resident on/visiting the two premises in Suffolk.**

The presence of stable / declining antibody titres in both the positive stallion and the seropositive gelding suggest that infection with EVA was not a particularly recent event and that active disease circulation was not present on the premises at the time of detection. This is further supported by the fact that the other three horses on the premises remain serologically negative after repeated testing.

It is known that following natural infection with EVA a solid and prolonged immunological response develops and that detectable antibodies can persist for several years (Balasuriya et al 2013, 2016 and references contained therein). One study suggests that following natural infection of mares, antibodies may persist for up to eight years (Amat et al 2016). There is no evidence of persistent infections developing in mares or geldings (Balasuriya et al 2016 and references contained therein).
Given the considerable uncertainties surrounding the potential source of infection it is not possible to provide a realistic estimate of the most likely period of source and spread of infection for this case.

**Tracings**

EVA is only notifiable in stallions and mares that have recently been mated or inseminated. Given the absence of evidence that disease was actively propagating on the affected premises, and the fact that the positive stallion had only ever once mated a mare (March 2018) that subsequently gave birth to a healthy foal and remains seronegative after repeated testing, additional extensive tracing investigations for this case were not considered to be proportionate. None of the events previously attended by the positive stallion or seropositive gelding since January 2018 involved overnight stays where more prolonged direct contact with other horses would be more likely to have occurred, and therefore these were not considered to represent a high risk of onward disease transmission.

Given the length of time that had elapsed between attendance by the positive stallion and seropositive gelding at events during 2018-2019, and confirmation of EVA in the positive stallion, any other horses that could potentially have been exposed to infection via this route would have subsequently seroconverted and cleared any virus, with the potential exception of stallions. However, compliance with the HBLB Code of Practice for EVA would mitigate against any subsequent risk from future breeding activities if any stallions had been exposed to infection via this route.

The premises in Suffolk where the positive stallion had previously resided for a number of months, and the veterinary practice that administered treatment, will be contacted to inform them that a subsequently confirmed EVA positive stallion had been present there and to advise them to undertake any additional measures that they consider to be appropriate in accordance with the HBLB Code of Practice for EVA.

**Direct tracings to Scotland, Wales or Northern Ireland**

None were identified.

**Spread to other EU Member States**

There was no travel to other EU member states and all events/other locations attended by horses from the affected premises were within Great Britain, other than one reported historical attendance by horse 4 at an event in the Netherlands in 2006 (prior to it being acquired by the current owners).
Conclusion

Given the considerable uncertainties around the potential source of infection for the positive stallion, the fact that it had only bred on a single occasion to a mare that remains seronegative and the in the absence of any substantive evidence for transmission of infection within the affected premises additional tracing activities were not considered to represent a proportionate approach in this case. The risk of further spread of EVA via future breeding activities has since been mitigated against by the owner voluntarily having the stallion castrated.

References


Annexes

Annex 1: Test results for private testing carried out on the premises

<table>
<thead>
<tr>
<th>Horse number</th>
<th>Date of sample collection</th>
<th>Serum VNT</th>
<th>Serum Antibody Elisa</th>
<th>Semen PCR</th>
<th>Laboratory comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>21/05/2019</td>
<td>Positive</td>
<td></td>
<td></td>
<td>Seropositive to EVA with a stable titre</td>
</tr>
<tr>
<td></td>
<td>04/06/2019</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>18/07/2019</td>
<td></td>
<td></td>
<td></td>
<td>Positive</td>
</tr>
<tr>
<td></td>
<td>25/07/2019</td>
<td>Positive</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Seropositive to EVA with a stable/declining titre</td>
</tr>
<tr>
<td>2</td>
<td>25/07/2019</td>
<td></td>
<td></td>
<td>Negative</td>
<td></td>
</tr>
<tr>
<td></td>
<td>08/08/2019</td>
<td></td>
<td></td>
<td>Negative</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>25/07/2019</td>
<td></td>
<td></td>
<td>Negative</td>
<td></td>
</tr>
<tr>
<td></td>
<td>08/08/2019</td>
<td></td>
<td></td>
<td>Negative</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>25/07/2019</td>
<td></td>
<td></td>
<td>Positive</td>
<td></td>
</tr>
<tr>
<td></td>
<td>08/08/2019</td>
<td></td>
<td></td>
<td>Positive</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>* Seropositive to EVA with a stable titre</td>
</tr>
<tr>
<td>5</td>
<td>25/07/2019</td>
<td></td>
<td></td>
<td>Negative</td>
<td></td>
</tr>
</tbody>
</table>
* When testing paired serology, the original sample should ideally be re-tested with the second sample to provide a valid paired test result. Therefore, separate testing of repeat samples could lead to some inter-assay variation between the original results and the re-test of the first sample. Only samples tested as pairs on the same occasion can be validly compared.

### Annex 2: Test results for official testing carried out

<table>
<thead>
<tr>
<th>Disease reference</th>
<th>Horse number</th>
<th>Date of sample collection</th>
<th>Serum SNT</th>
<th>Semen PCR 1</th>
<th>Semen PCR 2</th>
<th>Semen Genotyping</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2</td>
<td>08/08/2019</td>
<td>Negative</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>08/08/2019</td>
<td>Positive</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>