Editorial

Equine infectious anaemia in Europe: Time to re-examine the efficacy of monitoring and control protocols?

Introduction

Equine infectious anaemia (EIA) is a disease with an almost worldwide distribution and is of considerable importance to the equine industry, primarily because it is one of only 11 notifiable equine specific diseases listed by the World Organisation For Animal Health (OIE). Equine infectious anaemia is caused by EIAV (equine infectious anaemia virus), a blood-borne retrovirus belonging to the lentivirus genus [1]. It is unfortunate that the disease only receives significant attention when an EIA outbreak has a significant financial impact [2]. Equine infectious anaemia outbreaks have been reported in Belgium, Bosnia, Croatia, France, Germany, Greece, Hungary, Ireland, Italy, Latvia, Romania, Serbia, Slovenia and the UK between 2007 and 2014 [3]. In countries where EIA re-emerged after several years of absence such as in Belgium, Germany, Ireland and the UK, epidemiological studies suggested that equine biological products (such as blood plasma) imported from Italy or Romania were the source of the outbreaks. Moreover, several European countries (including Hungary, Poland and Serbia) have recently reported new EIA outbreaks and cases, underlining a wider extent of the disease in Europe than previously reported.

Recent outbreaks of EIA in European Union (EU) countries have prompted a closer look at the disease and alternative routes of transmission. Traditionally, EIA is seen as a blood-borne disease that can be mechanically transmitted by various blood-feeding insects, mainly Tabanidae. However, human actions (either iatrogenic or not) are suggested to have been an important contributory element, especially in recent outbreaks like the one in Ireland in 2006 [4]. In at least some of the EIA cases in the Irish outbreak (16 out of 38), it has been hypothesised that infection might have occurred through aerosolisation of infectious particles as a result of cleaning with pressure washers [5].

A few European nations reported EIA to the OIE between 2013 and 2014, namely Bosnia, Croatia, Hungary, Italy, Latvia, Romania and Serbia [3]. Two of these countries had ongoing (Romania) or have initiated (Italy in 2007) active surveillance programmes for EIA including systematic yearly testing of equids. Equine infectious anaemia is reported as endemic in the 2 EU countries that use systematic serological testing (Italy and Romania). We recently performed an 18 year retrospective study in Romania comparing the numbers of reported EIA outbreaks, number of individual cases and number of seropositive equids subjected to euthanasia as a result of the diagnosis. The data was collected from the OIE’s World Animal Health Information System (WAHIS for information after 2005) and Handistatus II (for information prior to 2005) and from the Institute for Diagnosis and Animal Health (IDAH), Bucharest, Romania. It is important to note that the Italian National Surveillance Programme that ran between 2007 and 2011 [6,7] demonstrated that testing exclusively using agar gel immunodiffusion (AGID), (the OIE prescribed test) can yield false negative results from a sizeable proportion of EIAV infected equids (perhaps exceeding 20%), thus emphasising the necessity to combine AGID with more sensitive tests such as ELISA or immunoblotting for more accurate detection and diagnosis.

Progress in EIA diagnosis and control in Romania

Equine infectious anaemia serological testing in Romania is based on the AGID test. By contrast, between 2008 and 2012, in several regions with no EIA positive cases or with low numbers of cases, testing was performed using ELISA followed by AGID confirmation to increase testing sensitivity and reduce the reporting of false negative results. The regional reference laboratories report their results to the IDAH which is also a testing centre. Compulsory annual testing of the entire equine population over 6 months of age from house-holdings (representing around 99% of equids in Romania) for EIA has been the policy in Romania since 1996 but was erratically enforced until the 2010 ‘Plan for eradication of EIA in Romania’ was initiated by the National Sanitary Veterinary and Food Safety Authority [8].

Over the investigated period (1996–2013), Romania reported 19,017 EIA outbreaks, including 49,651 identified cases of which a total of 14,166 horses were destroyed. All positives cases were registered to house-holdings rather than large commercial stables or studs. The statistics prior to 2010 are incomplete because the numbers of samples tested each year are not available, even though the census population was reported. Following the initiation of the rigorous EIA eradication plan in 2010, accurate and transparent records have been compiled and a steady reduction from 2973 cases in 2010 and 3154 cases in 2011 to 634 cases in 2014 (until August) has been demonstrated (Table 1). Over the last 5 years (2010 to 2014), EIA incidence (of animals tested) was 0.25, 0.48, 0.29, 0.20 and 0.12. Over the same period, respectively 85.1, 93.3, 97.5, 98.4 and 91.9% of the total equid population were reportedly sampled and tested. Importantly, the diagnostic procedures remained unchanged throughout the observation period. Starting in 2007, a marked drop in the total number of equids, reported at 800,000–900,000 horses per year from 1996 to 2009, was observed reaching 540,376 horses in 2013 (IDAH and [9]). This dramatic drop of ~300,000 horses (>30%) can only be marginally attributed to animals officially destroyed due to their positive EIAV status (14,264 horses) [3,10]; most were instead slaughtered for the meat industry [9].

Nevertheless, between 1996 and 2013, Romania eliminated 19.82% of all EIAV-positive animals. From 2002 to 2004, while the number of cases was at its peak with 29,709 cases reported, corresponding to 60% of the total cases recorded during 1996–2014, Romania did not routinely eliminate seropositive animals, with the exception of 2004 when 55% of the EIAV-positive horses were destroyed. The likely economic impact was probably of primary importance in the decision to keep these EIAV-positive animals alive. There was no regional repartition of EIA cases, infection being detected throughout Romania, with minor variation from year to year. Notably, since 2011, all equids tested and found to be positive for EIA are reported to have been subjected to euthanasia.

The Irish episode

The first outbreak of EIA in Ireland was notified in June 2006. Over the following 6 months, a total of 38 EIA seropositive cases were confirmed in 2 clusters [11,12]. Diagnosis was established based on one or more available diagnostic tests: AGID test, one or more commercially available ELISA tests, immunoblot and/or quantitative PCR and RT-PCR. The results of the epidemiological investigation revealed that plasma, believed to have been imported from Italy, was the source of the outbreaks [11,12]. Later studies led to the isolation and characterisation of the field isolate of the BAVg [Ireland] complete genomic sequence [13]. This is very valuable since it represents the only European isolate and is one of only 4 currently published EIAV genome sequences, beside the Chinese, Japanese and USA isolates and could contribute to the future development of more sensitive PCR-based diagnostic tests as well as to a possible EIA vaccine.

Romania via Belgium to UK

In 2010, Belgium made the headlines of the international press after an interesting outbreak of EIA. The epidemiological investigation started in the UK which, in January 2010, reported 2 cases of EIA introduced from...
revealed clustering with some Romanian isolates characterised previously. gag samples of the Belgian positive horses, focusing on their probably in the inapparent stage of disease, perhaps with very low levels of circulating antibodies at the time of their initial testing in Romania. Genetic characterisation of the EIAV strains isolated from different tissue samples of the Belgian positive horses, focusing on their gag gene, revealed clustering with some Romanian isolates characterised previously in 2009 [14].

**Table 1: Detailed epidemiological situation of equine infectious anemia (EIA) in Romania after restarting the EIA eradication plan in 2010 [Institute for Diagnosis and Animal Health (IDAH) data]**

<table>
<thead>
<tr>
<th>Year</th>
<th>2010</th>
<th>2011</th>
<th>2012</th>
<th>2013</th>
<th>2014</th>
</tr>
</thead>
<tbody>
<tr>
<td>Horse population</td>
<td>763,988</td>
<td>610,857</td>
<td>596,380</td>
<td>533,086</td>
<td>507,788</td>
</tr>
<tr>
<td>Number tested</td>
<td>562,350</td>
<td>606,894</td>
<td>594,198</td>
<td>531,866</td>
<td>466,591</td>
</tr>
<tr>
<td>Outbreaks</td>
<td>1513</td>
<td>3073</td>
<td>1542</td>
<td>817</td>
<td>573</td>
</tr>
<tr>
<td>EIA positive cases</td>
<td>2973</td>
<td>3154</td>
<td>1992</td>
<td>1075</td>
<td>634</td>
</tr>
<tr>
<td>Total eliminated*</td>
<td>1647</td>
<td>3153</td>
<td>1987</td>
<td>1075</td>
<td>634</td>
</tr>
</tbody>
</table>

*Includes elimination of EIA positive horses from previous years; **Number of equines tested by August 2014.

Belgium. The equids were originally exported from Romania, along with 16 other animals and only used Belgium as a temporary holding station before being moved to the UK. Later that year, the Belgian veterinary authorities tested 95 horses that could still be traced and identified as introduced from Romania and found 6 additional positive cases [14]. However, none of the horses that had been in contact with the positive cases was found to be positive using the AGID test. Moreover, none of the EIAV positive animals was reported to have shown clinical signs of disease, so they were probably in the inapparent stage of disease, perhaps with very low levels of circulating antibodies at the time of their initial testing in Romania. Genetic characterisation of the EIAV strains isolated from different tissue samples of the Belgian positive horses, focusing on their gag gene, revealed clustering with some Romanian isolates characterised previously in 2009 [14].

**Perspectives and concluding remarks**

How worrying or threatening EIAV really is for equine stakeholders remains an open question. It has previously been documented that most horse flies will not fly more than 50 m to find an alternative host for feeding but prefer to return to their original host [4]. An interesting aspect regarding the pathogenesis and clinical evolution of EIA is that, after acute and chronic phases, the vast majority of infected equids will eventually establish immunological control of the infection by switching from strain-specific to cross-reactive neutralising antibodies and thereafter enter an inapparent stage of disease [15]. This occurs approximately one year after the initial infection and the virus will then persist in monocytes for the rest of the animal’s life, with infected animals representing potential viral reservoirs. Various situations leading for example to immunosuppression may result in recurrence of disease in these infected animals [15]. Thus, an immunocompetent EIAV-infected horse in the inapparent stage of disease (as is the case for the vast majority of infected equids), is a very low risk for disease transmission but is still a virus reservoir for flies; that horse represents a threat for viral transmission because an immunosuppressive or other event leading to an increase in virus replication could trigger disease recrudescence.

Interestingly, it has recently been suggested by Issel [4] that the psychological fear of EIA greatly exceeds the risk it poses as a biological agent of disease. Indeed, in a clinical setting, EIAV spread is slow, as witnessed by several horse-owners and equine veterinarians in Romania (before 2004) who reported that on farms where there was one EIAV seropositive (Coggins test) horse that was not eliminated, no other horses on the premises seroconverted, even after several years. This is not a rule, however, and situations where one or more horses became EIAV positive over the following years were also reported. However, the Irish episode, in which human intervention was believed to have played an important role in EIAV transmission, should trigger alarm bells and remind the equine community that a risk assumption for the presence of an infectious disease applied to all contacts between equids would help decrease the actual number of animals that become infected [4].

It is certainly time to ask questions about the optimum approach to EIA. For example, is all the money invested in systematic testing (in the countries that apply it) really worthwhile and cost effective? Or should some of that money be dedicated to developing more sensitive diagnostic techniques (PCR based) at the EU level? In Romania, between 2010 and 2014, the incidence of EIA was a mean of 0.27% of the total population at risk. The number of EIAV positive cases in a country seems to be correlated with systematic testing (see the examples of Italy or Romania). If we look at the outbreak map for the last decade, EIA is fairly widespread in continental Europe (except for the Iberian Peninsula). A legitimate question that one might pose is what would happen if other EU countries started systematic testing for EIA? Most probably EIAV positive cases would be diagnosed (see recent outbreaks in Hungary, Poland or Serbia). The next question that arises, as suggested by Issel [4], is a simple one but with a complicated answer: Live with it or eradicate it? No cost estimations have been made at the level of the EU in terms of the money invested in EIA diagnosis (sampling-related costs, reagents, laboratory hours, technicians, veterinarians, etc). This analysis has, however, been performed in the USA and the result was that from 2007 to 2012, after testing more than 8 million equine samples, only 485 positives were found with average costs of USD 650,000 to find each positive animal [4]. Testing in the USA is generally focused on the mobile horse population because testing may be required for congregations. The untested population remains an unquantified potential reservoir [4].

Despite the massive elimination of EIAV positive animals in the last few years in Romania and Italy, EIAV actively circulates in and between EU countries, as proven by the recent outbreaks. This proves that EIAV is present within other European countries and future molecular epidemiological studies will be required to help understand the nature of circulating viruses in new outbreaks. Since Romania restarted the EIA eradication programme in 2010, accurate and transparent records have been compiled and a steady reduction from around 3000 cases in 2010 and 2011 to 634 in 2014 has been demonstrated. This efficient detection of EIAV seropositive animals and the elimination of infected animals should help stimulate reconsideration of the special conditions applicable to equine movements from Romania (Commission Decision 2010/346/EU). Despite testing being mandatory each year, it has not been possible to reach all privately held equids. Control of EIA in Romania has reached a point where eradication is feasible but may require higher compensation or other incentives to encourage full compliance and more complete voluntary testing (most horses in Romania are used for agricultural purposes), given that all positives will be destroyed and perhaps the use of additional test modalities proven to be more sensitive for the detection of antibodies against EIAV.

In the light of the epidemiological investigation in Belgium (which revealed no seroconversion for horses that had been in contact with the EIA positive animals) and the epidemiological situation following the Irish episode (where human intervention likely contributed to airborne transmission of EIAV), it may be time to review the status of EIA from a disease with potential for rapid spread within equine populations similar to equine influenza or African horse sickness to one with slow natural spread.

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**References**


