DEFRA / AHT / BEVA

EQUINE QUARTERLY DISEASE SURVEILLANCE REPORT

Volume 5, No.2: April – June 2009

Highlights in this issue:

- **Borna Disease**
- **West Nile Virus**

Important note:
The data presented in this report must be interpreted with caution, as there is likely to be some bias in the way that samples are submitted for laboratory testing. For example they are influenced by factors such as owner attitude or financial constraints or are being conducted for routine screening as well as clinical investigation purposes. Consequently these data do not necessarily reflect true disease frequency within the equine population of Great Britain.

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Introduction

Welcome to the second quarterly equine disease surveillance report for 2009 produced by Department of Environment, Food and Rural Affairs (Defra), British Equine Veterinary Association (BEVA) and the Animal Health Trust (AHT). Regular readers will be aware that this report collates equine disease data arising from multiple diagnostic laboratories and veterinary practices throughout the United Kingdom giving a unique insight into equine disease occurrence on a national scale. We would like to acknowledge the contribution of four laboratories which have reported their data for the first time to us.

National disease occurrence

The UK's Department for Environment, Food and Rural Affairs (Defra) gave notice on 29th July 2009 of confirmation of isolation of ***Taylorella equigenitalis*** from one subclinically infected stallion with no known sexual contact with the other 23 horses at premises near Bishops Stortford, Hertfordshire, England. The stallion entered the United Kingdom from mainland Europe one month ago. The stallion was clinically healthy and was swabbed for **contagious equine metritis (CEM)** as part of a pre-export procedure. The test results were positive with diagnosis by agent isolation and PCR by the UK's OIE reference laboratory for CEM at the Veterinary Laboratories Agency, Bury St. Edmunds. From available information, the stallion has not been involved in any breeding activities (covering mares or donating semen) since moving to the United Kingdom. Investigations and control measures including movement restrictions are on-going. (copy)
The detailed report can be found at
http://www.oie.int/wahis/public.php?page=single_report&pop=1&reportid=8330

International disease occurrences

A dead crow which tested positive for **West Nile Virus (WNV)** was found in Ontario, Canada and initially caused some trouble in the UK at the end of July as the bird was first wrongly reported by the internet platform “promed” to have been found in London’s East End. West Nile Virus is endemic in Ontario, but not in the UK. Earlier on this year Defra has published a qualitative risk assessment on WNV, which specifically addressed the likelihood of the introduction of WNV from abroad to the UK (http://www.defra.gov.uk/animalh/diseases/monitoring/riskassess.htm). Within this context Katherine Edgar from Defra has contributed a focus article on….

In the United States, as of 10 August 2009, a total of 21 stallions and five mares have now been confirmed as positive for ***Taylorella equigenitalis*** **(CEMO).** There were two new cases since the last quarterly disease report: At the end of May a Dutch Warmblood stallion which was imported into the United States in 2000 and epidemiologically linked to the Wisconsin outbreak, was confirmed positive through test-breeding for Taylorella equigenitalis (CEMO). At the same time an Arab gelding (stallion in 2007), also linked to the Wisconsin outbreak, was confirmed positive for Taylorella equigenitalis.
The event is continuing as reported by the OIE (Click here, please see also previous reports Vol. 4, No. 4 and Vol. 5, No. 1).

At the beginning of June 2009 16 new outbreaks with 209 cases of **equine influenza** were reported in the North and North West of India, giving a total of 38 outbreaks throughout India since July 2008 (see also previous reports Vol. 4, No. 4 and Vol. 5, No.1).

At the beginning of June 2009 an outbreak of **Equine piroplasmosis (*Theileria equi*)** was reported in Missouri, U.S. The affected index horse was a seven-year-old quarter horse gelding which was purchased six months ago and stood on a yard with 66 other equides. The affected horse was isolated and the whole yard was placed under quarantine. Six additional horses were confirmed as positive for *Theileria equi.*
Five of the seven positive horses were euthanized while two positive horses were illegally removed from the premises. No treatment of affected horses was allowed and a stamping out policy of positive animals was applied. Transmission is believed to have resulted from the use of shared needles for medication and not through ticks (www.oie.int).
Only the United States, Canada, Australia, Japan, England, Iceland and Ireland are not considered to be endemic areas. With regards to the World Equestrian Games in Kentucky in 2010, where piroplasmosis positive horses will be allowed to compete, an information document has been prepared by the Veterinary Services of the U.S. Department of Agriculture
(http://www.aphis.usda.gov/animal_health/animal_diseases/piroplasmosis/downloads/ep_2010_weg_wp.pdf).

On 29th May 2009 a horse in Texas was found to have swelling and vesicular lesions on the muzzle. About ten days later **Vesicular Stomatitis (VS)** was confirmed by competitive ELISA and complement fixation test at the National Veterinary Services (NVSL) and the affected horse was placed under quarantine. None of the three in-contact horses and the ruminants on the property showed any clinical signs. The serotype involved was classified as New Jersey. This was the nation's first case of VS for 2009. As of 11th August a total of five outbreaks (two in Texas, five in New Mexico) involving seven cases and 22 susceptible animals have been diagnosed in the U.S. Positive animals were placed under quarantine and were released 21 days after the lesions of all animals on the premises had healed. General control measures include quarantine and control of arthropodes, but vaccination is prohibited and there is no treatment of affected animals except supportive (www.oie.int). As of 19th August 2009 the event was confirmed as closed.
Horses are often the first animals to be confirmed with VS. This helps in differentiating between VS and foot-and-mouth disease (FMD), the main concern if vesicular lesions are seen in cloven-hoofed animals as horses are not susceptible to FMD. Defra has published a preliminary outbreak assessment for the current disease outbreak:
http://www.defra.gov.uk/animalh/diseases/monitoring/pdf/vs-horses-usa-090618.pdf

On the 8th of August 09 a horse died due to a **Hendra virus (HV)** infection on a stud farm in Queensland, Australia. The natural hosts for HV are flying foxes (fruit bates), which can spread HV to horses and rarely to humans. It was first isolated in 1994 in a stable in Hendra, Brisbane. The onset is acute and there is almost always rapid progression to death associated with fever and respiratory or neurological signs. Human infections, sometimes resulting in death, have occurred from handling infected horses. For more information please see:
http://www.dpi.qld.gov.au/cps/rde/dpi/hs.xsl/4790_11127_ENA_HTML.htm

Eastern Equine Encephalitis was confirmed in several States of the U.S. including Georgia, Louisiana, Florida, Missouri, Virginia, Texas and North Carolina. The virus is endemic to the bird population but is transferred to horses and humans through the bite of mosquitoes. It is fatal in 90% of cases, but there exists a vaccine for efficient prevention in equides (www.promedmail.org).

Defra news

Focus articles

In this report we are pleased to include two focus articles. As mentioned above, Katherine Edgar from Defra has prepared an overview on West Nile Virus lineages and Dr Maria Puorger and Professor Felix Ehrensperger from the Institute of Veterinary Pathology, Vetsuisse Faculty, University of Zurich present a report about Borna Disease. We reiterate that the views expressed in these focus articles are the authors' own and should not be interpreted as official statements of Defra, BEVA or the AHT.

Access to all of the equine disease surveillance reports can be made on a dedicated page on the Animal Health Trust website at http://www.aht.org.uk/equine_disease.html or via the BEVA and Defra websites:
http://www.beva.org.uk/
http://www.defra.gov.uk/animalh/diseases/vetsurveillance/species/horses/index.htm

We would remind readers and their colleagues that a form is available on the AHT website for registration to receive reports free of charge, via e-mail, on a quarterly basis. The link for this registration form is available via
http://www.aht.org.uk/equine_disease_registration.html.

Virology Disease Report for the Second Quarter of 2009

The results of virological testing for April to June 2009 are summarised in Table 1 and include data relating to Equine Viral Arteritis (EVA), Equine Infectious Anemia (EIA) and West Nile Virus (WNV) from the Veterinary Laboratories Agency (VLA), Weybridge. The sample population for the VLA is different from that for the other contributing laboratories, as the VLA's tests are principally in relation to international trade (EVA and EIA), although with recent Defra concessions VLA now provides testing for WNV as part of clinical work up of neurological cases on specific request and provided the local DVM has been informed.

Table 1: Diagnostic virology sample throughput and positive results for the second quarter 2009

	Number of Samples Tested	Number Positive	Number of Contributing Laboratories
Serological Tests			
EVA ELISA	3149	99#	5
EVA VN	1497	175#	3
VLA EVA VN	293	11#	1
EHV-1/-4 CF test	669	13*	1
EHV-3 VN test	1	1	1
ERV-A/-B CF test	242	9*	1
Influenza HI test	302	10*	1
EIA (Coggins)	911	0	4
EIA ELISA	906	0	4
VLA EIA (Coggins)	71	0	1
VLA WNV (PRNT)	0	0	1
Louping ill	0	0	0
Virus Detection			
EHV-1/-4 PCR	65	17	1
EHV-2/-5 PCR	2	0	1
Influenza NP ELISA**	201	9	1
Influenza Directigen	18	0	1
Influenza VI in eggs	0	0	1
EHV VI	201	19	1
EVA VI/PCR	1	0	1
VLA EVA VI/PCR	10	0	1
Rotavirus	111	50	6

ELISA = enzyme-linked immunosorbent assay, VN = virus neutralisation, VLA = Veterinary Laboratories Agency, CF = complement fixation, HI = haemagglutination inhibition, Coggins = agar gel immuno diffusion test, PCR = polymerase chain reaction, NP = nucleoprotein, VI = virus isolation, EVA = equine viral arteritis, EHV = equine herpes virus, ERV = equine rhinitis virus, EIA = equine infectious anaemia

= Seropositives include vaccinated stallions, * = Diagnosed positive on basis of seroconversion between paired sera

** = The relatively high number of NP ELISA tests performed is largely due to requirements for international equine movement. All horses travelling to Australia must now have 2 NP ELISA tests performed prior to travel. The figures above include tests performed for international trade purposes.

Of the 11 EVA VN positives detected by the VLA, two were export samples, one was an AI sample, two samples were from overseas, two others were diagnostic samples and four were private requests. The ten semen samples received for virus isolation were all negative for EVA virus isolation after three passages in RK13 cell culture and negative for EVA by the one-tube RT-PCR. They all were import-, export- or samples for private testing purposes.

The 71 agar gel immuno diffusion tests for EIA (AGID; Coggins) were conducted for international trade purposes and they were all negative. No testing for equine viral encephalitides was performed.

Virological Diagnoses for the Second Quarter of 2009

EHV-1 Abortions

Sixteen cases of EHV-1 abortion have been diagnosed by histology, immunohistochemistry, PCR and/or virus isolation. This includes a foal which was born normally but developed respiratory complications and died at four days of age (diagnosis with immunohistochemistry on the foetal lung), an outbreak on a polo yard with a total of three abortions (only one with laboratory diagnosis of EHV-1; pregnant mares in contact with yearlings) and a farm with three stillborn foals (only one with laboratory diagnosis of EHV-1).

EHV-1 paralytic disease

A mare presented off colour for a few weeks (was positive on strangles ELISA) and then developed mucopurulent nasal discharge and neurological signs (including incoordination). Histology of the brain showed changes consistent with viral encephalitis and PCR was EHV-1-positive on the brain tissue. No virus could be isolated from a NPS, brain tissue or CSF. All the 6 in-contacts were clinically fine but two of them showed a seroconversion to EHV-1 in the complement fixation-test. VI was negative on heparinised blood of the in-contacts with the exception of one sample where EHV-2 was isolated.
A five year old Welsh A gelding showed typical signs of paralytic EHV-1 infection including bilateral hind limb paresis, ataxia and urinary retention. Virus isolation on heparinised blood was negative but the pony showed a seroconversion to EHV-1 in the complement fixation-test. No other horses from the same premises were affected.
A nine year old thoroughbred gelding presented neurological signs including ataxia and urinary incontinence. EHV-1 was isolated from a heparinised blood sample. Serology also suggested recent contact with EHV. The horse recovered well with symptomatic treatment. Some of the 30 in-contacts at a riding school developed respiratory, but no neurological signs. The yard was isolated and no further cases were reported.

EHV-4 Respiratory infection

One case of EHV-4 respiratory infection was reported this quarter.

EHV-2

In a 20 year old TB mare EHV-2 was isolated from a NPS but not from heparinised blood sample.
In a two year old filly from a racing yard EHV-2 was isolated from heparinised blood but not from a nasopharyngeal swab. Paired serology for Influenza, EHV-1/-4, ERV A/B and Adenovirus did not show viral activity.
In a nine year old pony gelding and another animal EHV-2 was isolated from heparinised blood.

Equine Influenza

In Northumberland a 14 year old pony stallion which showed respiratory signs, tested positive for equine influenza by nucleoprotein ELISA on a nasopharyngeal swab. Subsequently influenza virus was isolated and sequenced. This pony has not been vaccinated against influenza for five years. The same pony tested also positive for *S.equi* by culture on a nasopharyngeal swab. It was the only flu case on the yard. It was not possible to do further virus analysis on this case.

In a trekking yard in Scotland equine influenza was diagnosed in four horses. The outbreak seemed to have started after a new horse was bought in from central Holland and started coughing two days after arrival. There were a total of eight affected horses and five in-contacts. The affected horses showed serous profuse nasal discharge, coughing at rest or occasional, glandular swelling, anorexia and pyrexia up to 105° F. The isolate belonged to clade 2 of the Florida sublineage of the American lineage of H3N8 equine influenza virus.

EHV-3

A mare with a positive EHV-3 titre on the virus neutralization test was reported to have developed pustules around the vulva after mating.

FOCUS ARTICLE: Borna Disease

Maria Puorger, DVM (Zurich); Dr. med. vet. (Zurich) and Felix Ehrensperger, Prof. Dr. med. vet. (Zurich), Dipl. ECVP, Institute of Veterinary Pathology, Vetsuisse Faculty, University of Zurich

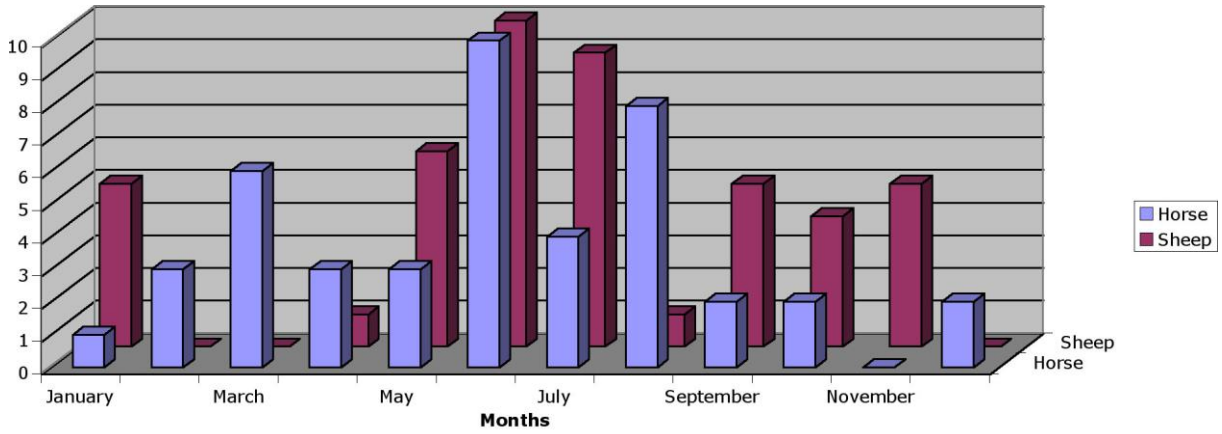
Introduction

Borna disease virus (BDV) is an enveloped virus with a negative-stranded non-segmented RNA genome of approximately 8.9 kb. It replicates and transcribes its genome in the nucleus and uses the cellular RNA splicing machinery to regulate gene expression. Mainly because of these features, BDV has been classified as the prototype virus of a newly established family, Bornaviridae, within the order Mononegavirales. BDV is the causative agent of Borna disease (BD), a mostly fatal meningoencephalitis originally detected among horses of Germany. Natural hosts of BDV are horses, sheep and other farm animals. Many other warm-blooded vertebrates ranging from rodents to non-human primates are susceptible to experimental infection with BDV. In these animals, BDV infection may either remain clinically inapparent, or it may lead to severe neurological abnormalities and eventually to death. Numerous studies with experimentally infected rats and mice have conclusively demonstrated that BD is caused by immunopathological mechanisms in which the antiviral T cell response results in neurological disorder.

Borna disease in horses and sheep is restricted to central Europe

BDV infections can result in neurological disease that mainly affects horses and sheep in certain areas of Germany. The endemic area also includes parts of the upper Rhine valley between Switzerland, Austria and the Principality of Liechtenstein. Between 1894 and 1896 a large epidemic of BDV-induced disease occurred among cavalry horses in the town of Borna in the state of Saxony (Germany). The disease and its inducing viral agent are named after the location of this first documented large outbreak. In recent years, the number of animals diagnosed with classical BD was relatively low, usually affecting less than a total of 100 horses and 100 sheep each year. To our knowledge, until very recently, no confirmed cases of BD had been reported in horses or sheep outside the endemic areas described above. The exception is a BD case in a horse from eastern Austria. Unlike infected animals from the classical endemic areas, this horse was shown to be infected with a novel genotype of BDV. Serological studies in horses outside the known endemic regions (Italy, Finland, Iran...) indicate that BDV could be more widespread.

Regarding the seasonal incidence of natural BD cases, the figure below shows the distribution of 107 BD cases in Switzerland, which had been diagnosed at the Institute of Veterinary Pathology in Zurich (47 horses, 46 sheep; not included in the graphic are 4 donkeys, 2 mules, 3 goats, 2 cows, 2 rabbits and 1 cat) during the last 30 years (1976 until today).
The figure shows a peak during the summer months, but there are as well cases during the other months. So the question aroused if this appearance would correlate with the occurrence of possible vector populations. However the incubation time is supposed to be up to several months, which might explain BD cases occurring in wintertime.



Diagnosis of Borna disease

Reliable intra vitam diagnosis of BD is difficult. Horses and sheep with BD exhibit a variety of clinical symptoms, predominantly behavioural abnormalities, apathy and movement disorders, which are not specific for BD but may also be seen in animals infected with other microorganisms that invade the CNS. Cerebrospinal fluid (CSF) of animals with BD may display pathological alterations, such as increased protein content and mononuclear pleocytosis. However, these changes are not specific for BD but rather represent non-specific indicators of viral meningoencephalitis. BDV-specific antibodies in serum and/or CSF are better indicators. Among the currently used methods of detecting these antibodies, indirect immunofluorescence assay (IFA) appears most reliable. The percentage of horses and sheep with confirmed BD that scored positive in this serological assay varied considerably between different studies. For these reasons, intra vitam examination alone can usually not provide firm proof of BD. Post-mortem confirmation by histological analysis of brain tissue is required.

Histologically, variable degrees of encephalitis are observed in brains of animals with BD. Lymphocytic infiltrations are usually most prominent in the hippocampus, the brain stem and in parts of the cerebral cortex. Inflammation is usually absent or less prominent in the cerebellum. $CD4^+$ T cells are predominantly present at perivascular sites, whereas $CD8^+$ T cells are found both in the perivascular cuffs and in the brain parenchyma. To clearly distinguish BD from encephalitis induced by other viruses, it is mandatory to prove that BDV infection of the CNS has occurred. Traditionally, Joest-Degen inclusion bodies in nuclei of infected neurons have served as BDV-specific markers, but they cannot consistently be seen by routine histology in brains of diseased animals.

RT–PCR or RT-nested PCR analysis of native or formalin-fixed brain tissue is a sensitive alternative technique that may be used to confirm the clinical diagnosis of BD.

Borna disease in other animals

BD is not strictly limited to horses and sheep, although the frequency at which other animals get the disease appears to be very low. BDV was found in donkeys, goats and cattle with neurological disease and strong lymphocytic infiltrations of the CNS. Some of the diseased bovines were from farms in regions of Germany in which BD is not endemic in horses and sheep. BDV antigen and infectious virus was shown to be present in the CNS of two rabbits with neurological disease which originated from the endemic region in Switzerland. An earlier report described the isolation of BDV from the brain of a rabbit with neurological disease. BDV antigen and RNA were further found in brains of several zoo animals in Erfurt that showed neurological disease. However, the identification of BDV in hosts such as cats, dogs, lynx, rabbits, and even ostriches indicates that the virus has a broad host repertoire of various birds and mammals.

Virus reservoir?

The search for vectors for BDV has mainly concentrated on subclinically infected horses and small ruminants, wild rodents and other small mammals. We identified BDV-infections without evidence of disease in wild shrews (Crocidura leucodon) captured from an endemic area.
Shrews belong to the order insectivora. Their appearance can be described as something between a mouse and a mole. The bicolored white-toothed shrew, Crocidura leucodon, occurs in the eastern part of Switzerland (Rhine Valley, Tessin), in the Rhône Valley, in the region of Basel, and the Principality of Liechtenstein and it belongs to the genus Crocidura together with two other species which occur in Europe (C.russula and C.suaveolens).

This species have to be considered as a reservoir species, because virus antigen could be demonstrated in various organs using different techniques like immunohistochemistry and real-time RT PCR without showing any pathological lesions.

Studies in rats had shown that the clinical course and histopathology of Borna disease varies with the age of the animal at the time of infection. In adult Lewis rats, BDV infection results in severe encephalitis accompanied by clinical symptoms that include hyperactivity, aggressiveness, and ataxia. At a later stage of the disease, surviving animals are apathetic and show signs of dementia and behavioral abnormalities, and their brains show a dramatic loss of neuronal tissue.
Experimental infection of adult mice takes a nonsymptomatic course, an observation previously believed to indicate that this animal species is not suitable for pathogenesis studies. However, BDV frequently induces severe neurological disease in infected newborn mice.

In wild birds, fragments of the BDV p24 and p40 genes from faecal samples collected at a bird pond could be amplyfied. Recently, the existence of an avian bornaviruses was demonstrated and provided as a compelling candidate in the search for an etiologic agent of proventricular dilation disease (PDD).
Ticks had been suspected to be possible vectors for BDV. This however has not been proven so far.

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FOCUS ARTICLE: The importance of West Nile Virus lineage

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Since 1996 West Nile Virus (WNV) has re-emerged as an arbovirus of both public health and veterinary concern. WNV is a non-contagious viral disease transmitted predominantly by *Culex* mosquitoes in a bird and ornithophilic mosquito enzootic amplification cycle. Incidental infections occur in horses, humans and other mammals as a result of bites from haematophagus bridging vectors.

Though initially thought to be two lineages, phylogenetic analysis of West Nile Virus has defined five distinct lineages that differ from each other by 20-25% at the complete genome level.

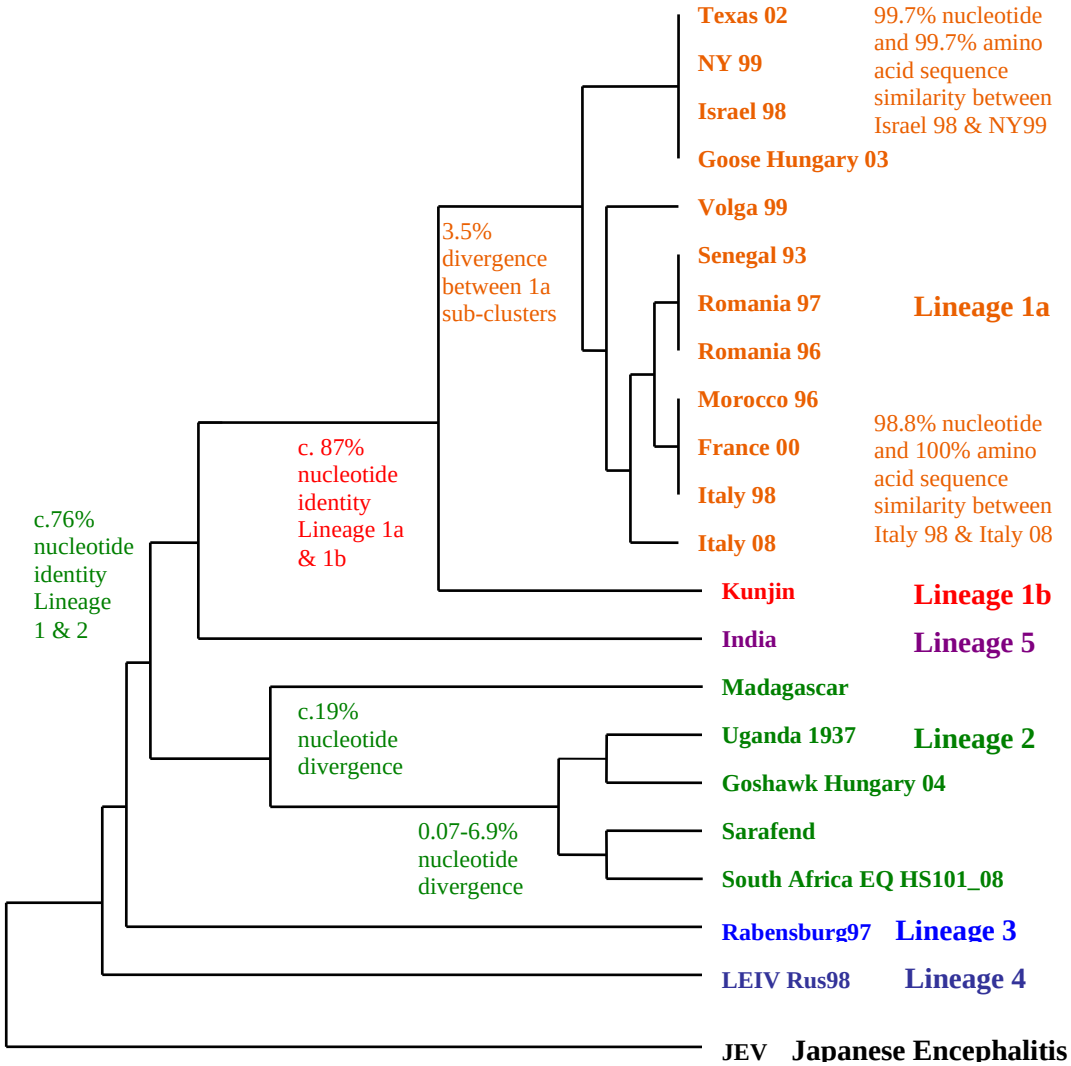

Figure 1: Phylogenetic analysis of West Nile Virus lineages (E-protein genome)

Lineage 1 is the most geographically widespread (1a Europe, Africa, Americas & Asia; 1b Kunjin Australia). Lineage 2 has been found in Africa and recently related to avian mortality in Central Europe (Hungary 2004 & 2005, Austria 2008). Lineages 3 and 4 have been identified in Russia and lineage 5 (formerly classified as 1c) in India.

Horses are susceptible to infection by both lineage 1 and 2, however it is not the individual WNV lineage, but the neuroinvasiveness of the individual virus serotypes that influence the development of clinical signs.

WNV presentation is a function of lesion location and produces a neurological syndrome characterised by a combination of clinical signs. The majority of WNV cases are subclinical or present with non-specific signs such as depression, anorexia and stiffness that resolve within 24-48hrs. Mild transient pyrexia (38-39^{o}C) may occur, but is often undetected. Serological investigations in a number of WNV outbreaks indicate that many horses produce antibodies but few develop clinical signs or mortality. WNV infection can however be life threatening to clinically affected horses.

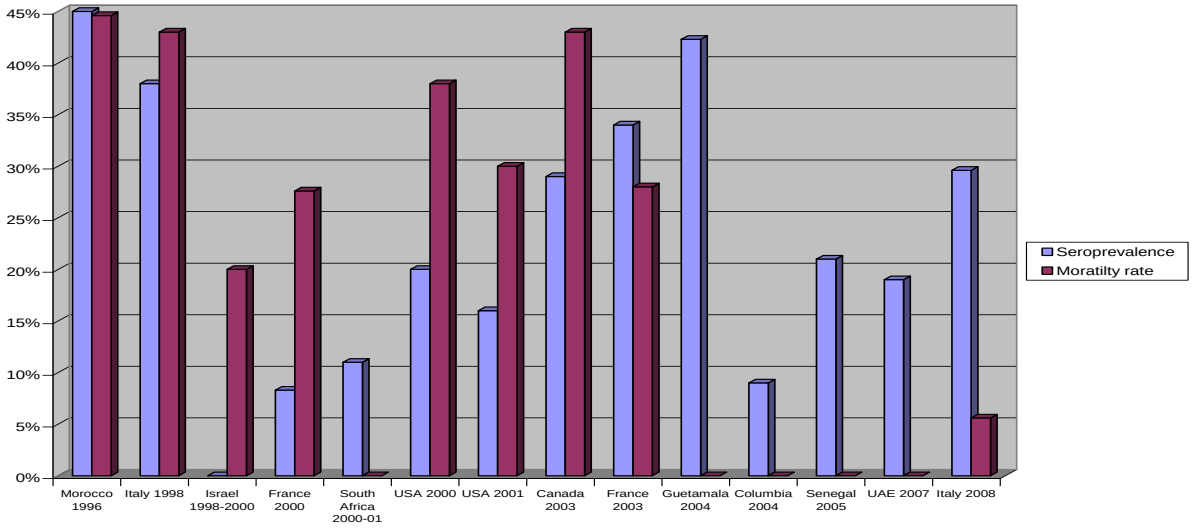

Figure 2: Comparison between observed seroprevalence and mortality rates for WNV lineage 1 and lineage 2 (South Africa 2000-01).

Mortality and recovery rates differ both between and within lineages. In the Italy1998 lineage 1a outbreak the mortality rate was 43%, but all of the horses that did recover did so fully and within a 5-15 day clinical course. This contrasts with the North American lineage 1a WNV outbreaks, where mortality rates ranged up to 43%, but had mean clinical courses >21 days and recovery rates of only 79-93%. This North American lineage 1a WNV serotype then spread into the Caribbean in 2004, with a seroprevalence of 9-42.3%in the absence of clinical disease. A neuroinvasive Lineage 2 strain in South African 2008 killed 5/7 clinically affected horses. The 2 surviving horses did fully recover after protracted rest over several months. Wider seroprevalence of this strain in South Africa is unknown, however in endemic regions equine WNV may reach high seroprevalence rates. In Romania lineage 1a WNV seroprevalence was reported at 56% in the absence of clinical signs, while maximum seroprevalence of African lineages has been reported at 75-86% with an annual infection rate of 11-21%.

WNV infections have been confirmed in horses ranging from 4 months to 38 years. Age prevalence does not seem to be lineage related, though a number of equine case studies have shown an age-related incidence in WNV lineage 1a infection in the 6-10 and 10-16 year age brackets. This may not be a true risk but a factor of equine use and mosquito exposure, or biased dependent on the economic or emotional value of the equines and whether reporting of suspect clinical signs is compulsory in that country.

Concurrent disease does not seem to play a major role in the pathogenesis of lineage1a WNV infection in horses; however co-infection in South Africa with African Horse Sickness, even in vaccinated horses, or Equine encephalosis may facilitate the central neuroinvasive quality of WNV lineage 2 leading to a greater case prevalence of blindness and seizures.

Ataxia and weakness are the major presenting clinical signs of WNV. Ataxia and muscle fasciculation may indicate a better prognosis than paresis, paralysis or central nervous signs. Though this is predominantly a function of diffuse lesions and less severe neuronal damage, recumbent horses have a poor prognosis due to intensive nursing requirement and associated secondary problems.

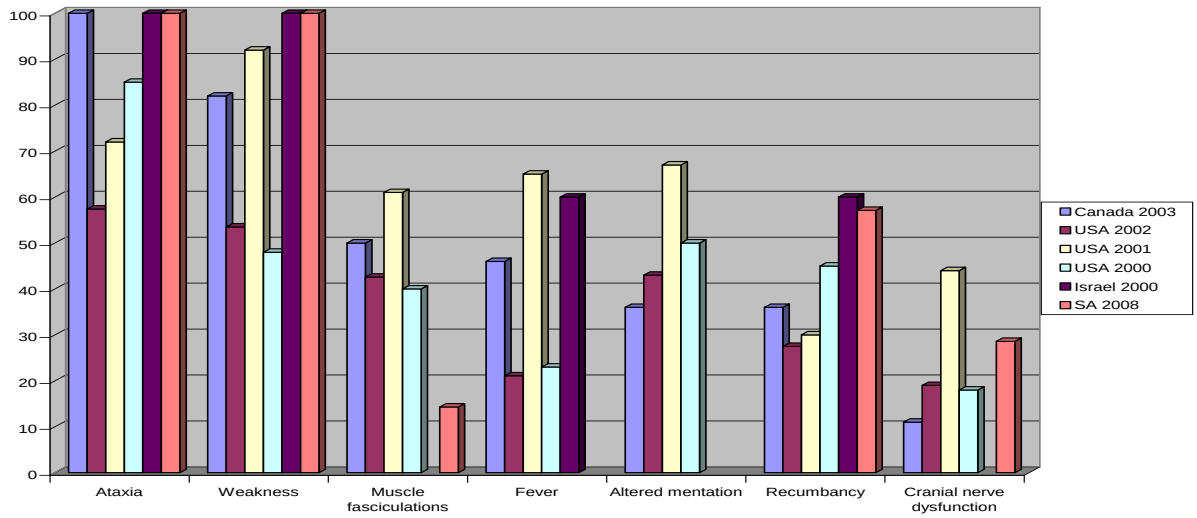

Figure 3: Clinical presenting signs associated with lineage 1 and lineage 2 (South Africa 2008) outbreaks

The risk of WNV introduction into the UK remains very low. Syndromic surveillance and the timely reporting of equine neurological disease can help detect any initial WNV case, regardless of WNV lineage, and provide a vital early warning of WNV circulation in the UK. Timely reporting and diagnosis also allows for more accurate assessment of the geographic distribution of WNV and will guide public health and veterinary preventative control measures with the aim of minimising any subsequent morbidity and mortality.

In the UK WNV is a notifiable disease. Any suspicion of WNV should be reported immediately to your local Animal Health office.

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Bacteriology Disease Report for the Second Quarter 2009

A summary of the diagnostic bacteriology testing undertaken by different contributing laboratories is presented in Table 2. For contagious equine metritis (CEM) 22 of 28 HBLB approved laboratories contributed data.

VLA CEMO Data for the period April to June 2009

We are again pleased to include data relating to CEM testing from the Veterinary Laboratories Agency (VLA), in this quarterly report. The sample population for the VLA is different from that for the other contributing laboratories as the VLA tests are principally in relation to international trade.

No isolates were identified as CEMO positive by HBLB laboratories.

Strangles

Strangles remains endemic in the UK, especially among parts of the non-Thoroughbred horse population. Diagnoses are confirmed in the UK based on traditional culture of *S.equi* and qPCR on respiratory samples and/or seroconversion using a blood-based ELISA.

Table 2: Diagnostic bacteriology sample throughput and positive results for the second quarter 2009

	Number of Samples Tested	Number Positive	Number of Contributing Laboratories
CEMO (HBLB)	7288	0	18
CEMO (VLA)	693	0	1
***Klebsiella pneumoniae*#**	7070[1]	20	18
Pseudomonas aeruginosa	7076[1]	25	17
Strangles*culture	1905	134	17
Strangles PCR	883	97	1
Strangles ELISA	1757	318	1
Salmonellosis	325	6	15
MRSA	258	0	6
Clostridium perfringens	53	6	4
***Clostridium difficile* (toxin by ELISA or immunochromatography)**	54	3	3
Borrelia (by ELISA)	16	3	1
Lawsonia intracellularis**	10	0	3

CEMO = contagious equine metritis organism (*Taylorella equigenitalis*); HBLB = HBLB accredited laboratories; # =capsule type 1,2,5; VLA = VLA reference laboratory**; ***Streptococcus equi* subsp.*equi*; MRSA = meticillin resistant *Staphylococcus aureus. ** Lawsonia intracellularis* identified using PCR applied to faeces; [1] reproductive tract samples only

VLA Salmonella results

The VLA tested a total of 15 samples of which three were positive (including the two positive samples sent to VLA by the isolating laboratory). From the strains typed by the VLA there were one case of *S. agama,* one of *S. mikawasiwa* and one of *S. newport*.

Toxic and Parasitic Disease Report for the Second Quarter 2009

A summary of diagnostic toxicosis and parasitology testing undertaken by contributing laboratories is presented in Tables 3 and 4 respectively. Results for toxicosis are based on histopathologically confirmed evidence of disease only (where applicable).

Table 3: Diagnostic toxicosis sample throughput and positive results for the second quarter 2009

	Number of Samples Tested	Number Positive	Number of Contributing Laboratories
Grass Sickness	36	23	3
Hepatic toxicoses	22	3	2
Atypical myopathy	1	0	1
Tetanus	1	1	1

Table 4: Diagnostic parasitology sample throughput and positive results for the second quarter 2009

	Number of Samples Tested	Number Positive	Number of Contributing Laboratories
Endoparasites			
Ascarids	1029	5	10
Cyathostomes	838	167	8
Dictyocaulus	484	3	8
Strongyles	5111	811	16
Tapeworms (ELISA based testing)*	923	636	4
Tapeworms (Faecal exam)	895	14	7
Trichostrongylus	22	0	1
Strongyloides	1011	5	10
Oxyuris equi	54	0	2
Fasciola	102	14	2
Coccidia	43	6	1
Cryptosporidia	2	1	2
Ectoparasites			
Mites	458	0	11
Lice	330	3	10
Ringworm	411	82	16
Dermatophilus	205	11	6
Candida	16	0	1

Grass sickness surveillance data (www.equinegrasssickness.co.uk):
A total of five Equine Grass Sickness cases were submitted to the surveillance scheme between April and June this year.
It should be noted that the grass sickness surveillance scheme receives data from a wider population in comparison to the data presented in Table 3 and different diagnostic criteria were used. For more information about the grass sickness surveillance please refer to previous reports published in Vol.4 No.2 and Vol.2 No.4.

Report on Post Mortem Examinations for the Second Quarter 2009

East Anglia

A total of 58 cases were examined including 28 aborted fetuses.

Of the aborted fetuses examined this quarter, umbilical cord torsion was suspected as the precipitating cause in 4/28 cases. Other non-infectious causes included one case of premature placental separation and one case of meningocoele.

Equine Herpes Virus 1 (EHV-1) was found to be the cause for 10 abortions. Diagnosis of EHV in general was based on positive PCR and typical histopathologic changes, in some cases including immunohistochemistry. Two aborted fetuses were submitted for EHV clearance only and were negative. Other infectious causes of abortion included two cases of placentitis.

Four cases of non-infectious (intrapartum) stillbirths were examined, one of which was associated with abnormal in utero presentation combined with an early detachment of the placenta.

A male thoroughbred foal was born full term and alive but died immediately after birth due to a non-infective cause. Further cases of neonatal death were reported as dystocia associated (2), sepsis (1), hydrocephalus (1), cleft palate (1), tumorous lesion (1) and intestine strangulation (1).

A newborn thoroughbred foal which survived only for 12 hours had poor viability due to lung atelectasis and placentitis (hypoxia, foetal stress, meconium aspiration in utero).

In a two day old Welsh filly foal which died suddenly, *E.coli* septicaemia was diagnosed post mortem.

In a 36 hour old colt foal that died suddenly, multifocal necro-suppurative embolic nephritis and adrenalitis was diagnosed post mortem. The random distribution of lesions, and the frequent targeting of glomeruli were consistent with sepsis. The pulmonary changes most likely indicated this animal died of septic shock, a finding supported by the presence of adrenal cortical haemorrhage, which is often associated with death due to shock. Septicaemia in foals is not uncommon, and may be associated with in utero bacterial disease, or may be due to neonatal infection and subsequent septicaemia. No evidence of bacterial infection, i.e. umbilical abscess etc, was identified in this case to indicate the source of infection, however in neonatal foals with inadequate colostrum, minor infections may progress rapidly to a septic state. Bacterial culture from liver, lung and kidney revealed infection with *Pasteurella pneumotropica*, an opportunistic bacterium.

A thoroughbred foal was born three weeks early but was initially normal at birth. About four hours later it started to show classical signs of Hypoxic Ischemic Encephalopathy and had to be euthanized at 36 hours of age due to severe progressive seizure activity, renal failure and ileus. Gross post mortem examination was unremarkable.

A three day old Trotter cross foal was examined post mortem after having died while under veterinary care due to septic fetlocks. At death this three day old foal was thin,

dehydrated and had atrophic lymphoreticular tissues. The history and postmortem findings indicated that the fetlock joint sepsis was clinically evident at one day of age, and well established by three days when it died. This, together with the severe deep gastric ulceration, confirmed that the foal had been compromised from an early age. In spite of treatments *Pasteurella trehalosi* (*P. haemolytica* var.) was grown from both a septic fetlock and lung tissues. The recent lung changes, including the pulmonary vascular changes (perivascular haemorrhages and early necrotising vasculitis) are likely to have been of most significance in causing death. However, it is not possible to identify exactly what initiated or potentiated these changes; bacterial toxins and an adverse response relating to the plasma infusion are possible contributory factors.

Grass sickness was diagnosed in a six year old Welsh pony mare and an eight year old thoroughbred mare, based on histology showing widespread degeneration of neurons with extensive chromatolytic changes in the anterior mesenteric ganglion and the anterior cervical ganglion.

An 11 year old warmblood stallion died shortly after bone marrow aspiration from the sternum. Post mortem examination revealed a haemopericardium due to perforation of the right ventricular heart wall.

An eight month old Hanoverian cross colt presented with acute diarrhoea after a short course of NSAIDs and required euthanasia despite of a week of hospitalization and intensive treatment. Post mortem examination revealed that the clinical deterioration of the yearling was attributable to a severe ulcerative typhlitis, with localised peritonitis. Bacterial culture proved negative for *Salmonella* and *Campylobacter* species, but did yield potential pathogens of *Streptococcus zooepidemicus* and *Pasteurella trehalosi*. It is possible that concurrent antibiotic therapy may have modified the results of bacterial culture, but it was concluded that bacterial infection had caused the very severe typhlitis. An additional finding was evidence of nephritis, which may have been secondary to the enteric infection. A further incidental finding was superficial oesophagitis associated with yeast colonisation. There was no evidence of gastric ulceration or renal pelvic necrosis, which are sometimes reported as adverse responses to non-steroidal antiinflammatory drug therapy. There was also no evidence of significant endoparasitism.

A case of Equine Protozoal Myeloencephalitis (EPM) was presumptively diagnosed in a six year old ataxic thoroughbred mare imported from the USA based on a weak positive result in a Western Blot analysis conducted on a serum sample. This result was suggestive of exposure but did not confirm active infection (please see also pm-report West Midlands Vol.5 Nr.1). The mare initially responded well to treatment, but was euthanized on humane grounds after a relapse (recumbency). Post mortem and histopathological examination revealed lymphohistiocytic, multifocal, severe, meningoencephalomyelitis with intralesional schizonts. The histopathological pattern is consistent with a haematogenous brain infection by microparasites of the Apicomplexa family. Even though intralesional organisms were detected microscopically, any further classification regarding *Sarcocystis neurona*, which induces the so-called equine protozoal myeloencephalitis (EPM), or Neospora species would require further immunohistochemical or PCR testing. The extent of the brain lesion and the associated decay of descending and ascending spinal cord fibres explain the clinical picture sufficiently. The inflammatory and destructive process appeared to have been active for at least several weeks.

A zebra yearling colt was examined post mortem and found to have suffered spinal trauma. He collapsed suddenly andmovement was still possible in all four limbs, but he couldn't get up. Radiographic examination showed instability at C2-C3. The main post mortem finding in this animal was a cervical spinal sub-luxation at C2-C3, presumably due to external blunt trauma, and severe soft tissue alterations such as laceration of the para-axial and epaxial musculature of the neck, rupture of the synovial membrane and ligaments of the C2-C3 articulation and a subsequent contusion of the corresponding spinal cord segment. Thorough investigation of macerated and boiled vertebral column ruled out pre-existing orthopaedic problems or gross instability of the cervical spinal cord that might have led to a so-called Wobbler syndrome. Whether or not the trauma had been caused by other zebras of the same group cannot be answered for sure. Coincidental findings in this animal were a high ascarid burden and associated eosinophilic enteritis. Furthermore, there was a mild multifocal broncho-interstitial pneumonia noted in the samples taken from the lungs, the aetiology of which remains uncertain at the present stage.

A nine month old female Grevy zebra was diagnosed at post mortem with a fracture at the C2/C3 articulation with damage to the spinal cord, and a much older bony lesion at C3/C4. It was very likely that the zebra has sustained two separate neck injuries.

The head of a five year old Dutch warmblood mare was examined post mortem. The mare had shown progressive neurological signs (vestibular signs including head tilt, nystagmus, hyperaesthesia and possible visual impairment) and abnormal behaviour. No abnormalities were found.

A thoroughbred filly of unknown age presenting with a suspected skull fracture and blood coming from the nares was examined post mortem. Macroscopic examination revealed severe comminuted fractures of the cranium, occipital, parietal and sphenoid bones. There was sub-total transection of the spinal cord at C1 and severe haemorrhage in the frontal, caudal maxillary and conchal sinuses. There is no evidence on gross examination of any possible underlying condition in this animal, which may have predisposed her to either pain or a neurologic condition. Fracture of the occipital and basisphenoid bone are the most common sites of fracture in horses that rear back and fall over. Fracture of these bones by other means is difficult to obtain, and considered unlikely in the absence of gross evidence of severe bodily trauma.

Single cases of fibrocartilage embolism, pedunculated lipoma with strangulation of small intestine, dermal and subcutaneous lymphosarcoma, spindle cell sarcoma, tibial fracture and femoral fracture were reported.

Additional single cases of lymphosarcoma, pleuritis, ruptured caecum, gastric rupture, large colon torsion, and enteritis have also been observed during post mortem investigations.

Home Counties

Nine cases were examined this quarter.

Two neurological cases were reported including a case each of equine degenerative myeloencephalitis and cholesteatoma.
Two animals which presented with colic signs were examined post mortem; one was subsequently diagnosed with a strangulating lipoma.

Additional cases consisted of a pars intermedia adenoma, a C-cell adenoma of the thyroid gland, a melanoma, end stage kidney disease, weight loss with hypercalcemia, and a case of ulcerative cystitis.

South West

Four cases were examined during this quarter.

In two donkeys liver cirrhosis was diagnosed by gross post mortem examination and histology. In two other donkeys laminitis was diagnosed clinically and at post mortem examination.
A case each of squamous cell carcinoma in the nasopharynx and rib fracture with pericarditis and pyothorax were diagnosed at gross post mortem examination and histology.

Scotland

Four cases were examined post mortem this quarter.

There were two cases of grass sickness, one case of spinal deviation, and one case of hepatic lipidosis.

Northern Ireland

Ten cases, including one fetus, were examined this quarter.

Hepatic lipidosis was diagnosed in a four year old Shetland pony that had aborted two weeks previously and then presented as dull, stiff and dyspnoeic. At gross post-mortem examination widespread icterus and typical fatty change affecting the liver were evident. No cause for the abortion was found in the foetus.

Clostridial myositis was diagnosed in a two year old pony that had previously presented with sudden onset swelling of the throat and neck 48 hours prior to death. At gross post mortem examination there was marked swelling and oedema of the head and haemorrhage, oedema and necrosis of the left caudal neck musculature. Histological examination showed evidence of myofibril necrosis, emphysema, oedema, leucocyte infiltration and numerous rod-shaped bacteria.

Post mortem examination of a ten day old foal identified the cause of death as a large perforated duodenal ulcer, which had resulted in the leakage of intestinal contents into the abdomen and a widespread fibrinous peritonitis.

Parasitic gastroenteritis with an elevated faecal strongyle egg count was identified in an adult donkey that was presented for post mortem examination in poor body condition.

Colisepticaemia and arthritis was diagnosed in a three day old foal.
Strongyloides westeri infection was diagnosed in an eight week old foal that had died suddenly.

Other post mortem submissions included trauma to the head and tearing of the right longissimus dorsi muscle in a four year old horse that had died suddenly. A case of suspected drowning was also investigated and for one case no diagnosis was reached.

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We would welcome feedback including contributions on focus articles and/or case reports to the following address:

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