On 11 August 2008, a seven year old Quarter horse in Florida was presented to a veterinarian with a three week history of fever, sweating, not drinking and abnormal urination. The horse had a history of a similar illness six months earlier, but veterinary attention had not been sought.

On 13 August 2008, the veterinarian notified the Florida state animal health authorities of a suspicion of equine piroplasmosis – a disease considered exotic to the United States. The diagnosis was confirmed the next day, and the horse was euthanased.

Over the next month, 20 horses on six separate properties tested positive to piroplasmosis, 25 properties were quarantined and approximately 200 horses had been tested by 19 September 2008. The investigations were continuing at the time of writing.

The disease
Equine piroplasmosis is a blood-borne disease caused by the protozoan parasites Babesia caballi and Theileria equi (formerly called Babesia equi). The parasites can be transmitted by the bite of infected ticks, or mechanically from infected to uninfected horses through the re-use of blood-contaminated needles, syringes and surgical instruments. Foals of carrier mares can also be infected in utero.

Clinical signs of piroplasmosis are non-specific, and are associated with red blood cell destruction and release of inflammatory mediators. The severity of the clinical signs depends on the rate and number of red cells destroyed and the degree of the inflammatory response. In general, B. caballi infections are less severe than T. equi infections, in which a greater proportion of red blood cells are destroyed.

Mildly affected horses show poor appetite, poor performance and weight loss.

Acutely affected horses may show some or all of the following signs:

- fever
- jaundice
- anaemia
- increased heart and respiratory rates
- colic
- limb oedema
- petechial haemorrhages on the mucous membranes
- red-tinged urine (haemoglobinuria)
- neurological signs.

Recovered horses may become long-term carriers, and present a potential source of infection for other horses. For this reason, many countries place restrictions on the importation of horses that test positive for equine piroplasmosis.

Vectors
Only some species of ticks are competent vectors of B. caballi and T. equi (that is, they are able to support the life cycle of the protozoa) (Figure 1). Australia has at least two tick species, Boophilus microplus and Rhipicephalus sanguineus, that are capable of transmitting equine piroplasmosis between horses, at least experimentally. B. microplus is generally restricted to northern and north-eastern coastal and subcoastal areas of Australia by climatic conditions. R. sanguineus has a widespread distribution, but is not commonly found on horses, preferring the dog as its host.

Diagnosis
In the acute case, where there are many circulating protozoa, the organisms can be seen on blood smears prepared with Romanovsky-type stains, such as the Giemsa stain. This is how the Florida case, described above, was initially suspected. B. caballi form pairs of merozoites connected at one end at an angle (Figure 2), while T. equi organisms congregated in pairs or sets of four, in a ‘Maltese cross’ form (Figure 3).

Diagnosis by direct examination of blood smears is not reliable when there are few circulating parasites, such as in mild cases or carrier horses, or many B. caballi infections. In these cases, diagnosis relies on serological tests or detection of parasite DNA.

Serological tests
Several serological tests are available for equine piroplasmosis. Unfortunately, they are all subject to false positive and false negative results. In suspected cases, it is common for a laboratory to do a combination of tests.

The most commonly used serological tests are:

- the indirect fluorescent antibody (IFA) test, which can be used to distinguish between T. equi and B. caballi
- the enzyme linked immunosorbent assays (ELISA), which can detect antibodies to both species in infected horses but may not distinguish between T. equi and B. caballi
- the complement fixation (CF) test – this test is not as useful as IFA or ELISA tests for detecting long-term carrier horses in which the CF titre may have waned, but the horse can remain serologically positive with the IFA and ELISA.

Figure 1: A horse infested with Dermacentor nitens, a tick vector for Babesia caballi in the Americas (courtesy of Associate Professor Katherine Comendouros DVM, MS, PhD, Dept. of Veterinary Sciences, Veterinary Institute, Universidade Federal Rural do Rio de Janeiro, Brazil)
Equine piroplasmosis is a notifiable disease in Australia. Any suspected case or positive test result for equine piroplasmosis must be reported to the veterinary authority of your state or territory government so that confirmatory testing can be carried out.

**Detection of DNA**

The protozoan DNA can be detected by polymerase chain reaction (PCR) test in host blood or tissues, or in the tick vector.

**Other diagnostic methods**

Two less commonly used diagnostic methods are described for cases in which serological and molecular test results are equivocal.

A non-infected known tick vector can be fed on a suspect animal and the identification of *B. caballi* or *T. equi* within the tick vector is used to confirm the diagnosis.

A susceptible splenectomised horse can be inoculated with whole blood from a suspect horse, and then the demonstration of clinical signs and parasitaemia in the recipient horse can be used to confirm the diagnosis.

The index infected horse in Florida in 2008 tested positive to equine piroplasmosis by PCR, the IFA and CF tests, but negative by the competitive ELISA test. Parasites were also seen on direct blood smears.

**Tests available in Australia**

The IFA test for equine piroplasmosis is available at a private laboratory in Western Australia. The Australian Animal Health Laboratory in Geelong is currently developing test capability for equine piroplasmosis.

Equine piroplasmosis is a notifiable disease in Australia. Any suspected case or positive test result for equine piroplasmosis must be reported to the veterinary authority of your state or territory government so that confirmatory testing can be carried out.

**Treatment**

In countries with endemic equine piroplasmosis, horses may be treated to alleviate clinical signs of the disease. Diminazene diaceturate, phenamidine isethionate and amicarbalide disethionate are used for treatment of *B. caballi* infections.

Buparvaquone and other antitheilerial drugs can be used to treat *T. equi* infections. Imidocarb, one of the treatments recommended for *T. equi*, has a narrow safety margin in horses and other equids, and treatment is associated with toxic side effects, especially in donkeys.

Treatment of equine piroplasmosis is usually ineffective at eliminating the carrier state. While some *B. caballi* infections can be sterilised, *T. equi* infections tend to persist despite treatment. Therefore, surviving horses in a country otherwise free of equine piroplasmosis would most likely be subject to lifetime quarantine controls or euthanasia.

**World distribution**

Piroplasmosis is endemic in many areas of the world, including Africa, Europe, Asia, Central and South America. Countries believed to be free of the disease include Australia, New Zealand, Canada, the United Kingdom, Ireland, and Japan. Equine piroplasmosis was thought to have been eradicated from the United States until the 2008 outbreak in Florida.

**History of equine piroplasmosis in Australia**

Equine piroplasmosis has occurred in Australia in the past. *T. equi* was introduced into Australia on three occasions by carrier horses imported from the United States and Spain in the 1950s, 1960s and 1970s.

An outbreak of equine piroplasmosis was described in polocrosse horses in the Moss Vale area of NSW in 1976, and was attributed to the re-use of needles and blood-letting equipment on imported carrier horses and locally bred horses. It is believed that the disease did not establish in the local tick population, and Australia is now considered free of equine piroplasmosis. Testing requirements for imported horses have since been amended to prevent seropositive horses entering the country as permanent imports.

Occasional Australian horses have tested seropositive for equine piroplasmosis in pre-export testing in recent years. However, re-testing of these horses has failed to confirm a diagnosis of the disease, and the initial test results are believed to have been false-positives.

**Measures to exclude equine piroplasmosis from Australia**

Horses testing positive to equine piroplasmosis are not permitted to enter Australia as permanent imports.

Seropositive horses may enter Australia temporarily for exhibition, racing or other competition, under strict conditions. These horses must be inspected for and treated to remove ticks before and after arrival, and periodically during their stay in Australia. Seropositive horses are subject to quarantine control while in Australia.

Piroplasmosis positive horses are permitted to compete in a controlled environment (as they did at the 2000 Sydney Olympic Games), but they are not permitted to compete in events where the potential exists for prolonged exposure to vegetation and opportunity for tick attachment (e.g. endurance events).

Full details of import requirements for horses can be obtained from Biosecurity Australia: www.daff.gov.au/ba/reviews/current-animal/horses and AQIS: www.daff.gov.au/aqis/import/live-animals. These requirements are subject to change from time to time.

**What to do if you suspect equine piroplasmosis**

Equine piroplasmosis is a notifiable disease. A suspected case must be reported to the veterinary authority of your state or territory government department of agriculture or primary industries, so that confirmatory testing can be carried out.

**Suggested reading**