Institution:
UNIVERSITY OF LIVERPOOL

Unit of Assessment:
UOA6 - Agriculture, Veterinary and Food Science

Title of case study:
Foal Immunodeficiency Syndrome - Control of a Fatal Equine Genetic Disease

1. Summary of the impact

Foal Immunodeficiency Syndrome (FIS) is an emerging fatal inherited equine disease which has caused much concern in the equine industry. Research at the University of Liverpool (UoL) into the genetic basis of this disease has identified the genetic mutation and developed a carrier test which led to equine population screening to understand the spread of this disease (>40% adult carriers in one breed, Fell ponies) and provided a tool for vets and owners to design selective breeding programmes to eradicate the disease. Since the introduction of the test in 2010, the number of cases has drastically fallen (only 1 detected in any breed in 2012) and FIS spread into other breeds is now considered most unlikely.

2. Underpinning research

Foal Immunodeficiency syndrome (FIS), first reported in Fell ponies in 1997, is a 100% fatal disease that affects foals of 4-12 weeks age. Requested by the Veterinary Laboratory Agency to investigate this disease, which was increasingly reported, Professor Stuart Carter of UoL and clinician Knottenbelt (1999-2005), identified the key biological defects in affected foals: profound anaemia and loss of B lymphocytes leading to reduced immunoglobulin production, so that foals succumb to opportunistic infections. In addition to its significance for equine welfare, the data clearly showed that FIS is a novel immunodeficiency providing insight into mammalian immune system function.

Thomas (UoL PGR) examined stud books which showed that FIS has an autosomal recessive genetic basis, explaining why affected foals were being produced after matings of clinically normal mares and stallions which were both FIS carriers. Thus, the mutated gene was spreading undetected in the equine populations and there was every likelihood of it being passed to other equine breeds by cross breeding. Carter reported the first FIS case in Dales ponies (2009) and has detected carriers in two more breeds subsequently. Prior to 2007, analysing genetic associations with equine diseases was quite difficult because comprehensive tools were not available. Once the equine genome became available, it was possible to take a molecular genetic approach to identify what was likely to be a small, but very deleterious change in equine DNA.

Utilising the equine genome, Fox-Clipsham (UoL PGR) identified microsatellite markers that clearly indicated strong FIS linkage to chromosome 26. The location of the aberrant genetic mutation was confirmed and refined with a whole genome SNP array (Illumina) identifying 54,600 equine single nucleotide polymorphisms (SNPs), enabling a genome wide association study. This clearly identified a candidate region of 2.6Mb on chromosome 26 which was captured and resequenced (GS FLX Titanium @ UoL). Eight SNPs were identified in affected foals, narrowing the critical region (to 0.3 Mb) and enabling Sanger sequencing which identified the one SNP completely associated with FIS. A PCR test was developed in 2010 to identify this SNP in samples and made available (commercially via the Animal Health Trust (AHT), Newmarket) to owners to enable identification of FIS carriers prior to breeding.

This work was entirely carried out by those named above at the UoL. Over 2,750 samples of horse DNA were screened to identify carriers and affected foals. The research benefited from collaboration with the Animal Health Trust, breeders, vets and others.

3. References to the research

1. **Carter SD**, **Fox-Clipsham LY**, **Christley R** & Swinburne JE (2013) Foal Immunodeficiency Syndrome: Carrier testing has markedly reduced disease incidence. Vet Rec 172: 398 DOI: 10.1136/vr.101451 Citations: 0 Impact Factor: 1.803

2. **Fox-Clipsham LY**, **Carter SD**, Goodhead I, **Hall N**, **Knottenbelt DC**, May PD, Ollier WE, Swinburne J (2011) Identification of a Mutation Associated with Fatal Foal Immunodeficiency Syndrome in the Fell and Dales Pony. PLOS Genetics e1002133. doi:10.1371/journal.pgen.1002133 Citations: 7 Impact Factor: 8.517

3. **Fox-Clipsham LY**, Brown EE, **Carter SD** and Swinburne JE (2011) Population screening of endangered horse breeds for the Foal Immunodeficiency Syndrome mutation. Vet Record 169: 655-658 Citations: 2 Impact Factor: 1.803

4. **Fox-Clipsham L**Y, Swinburne JE, Blunden AS, **Malalana F**, **Knottenbelt DC** & **Carter SD**. (2009). Immunodeficiency/anaemia syndrome in a Dales pony. Vet Rec.165: 289-90 Citations: 6 Impact Factor: 1.803

5. **Thomas GW**, **Bell SC**, **Phythian C**, **Taylor P**, **Knottenbelt DC** & **Carter SD** (2003). Aid to the antemortem diagnosis of Fell pony syndrome by the analysis of B lymphocytes. Vet Rec 152: 616-621 Citations: 13 Impact Factor: 1.803

6. **Thomas GW**, **Bell SC**, & **Carter SD** (2005). Immunoglobulin and peripheral B-lymphocyte concentrations in Fell pony foal syndrome. Equine Vet J 37: 48-52 Citations: 13 Impact Factor: 2.286

Key grants

1998 – 2010. **Fell Pony 2000**. Fell pony immunodeficiency studies at Liverpool, £35k, **SD Carter**

1999 – 2002. **Home of Rest for Horses**. Immunodeficiency in Fell ponies, £148,894. **SD Carter** & MA Holmes *(funded PhD studies for G Thomas (GT))*

2001 – 2003. **Home of Rest for Horses**. Genetics of Fell pony syndrome, £23k, **SD Carter** & M M Binns

2007 – 2010. **Horse Trust**. Fell pony syndrome: Identifying the genetic defect, £110k, J Swinburne & **SD Carter** *(funded UoL PhD studies for L Fox-Clipsham (LF-C))*

4. Details of the impact

Before 2010, the chance of a foal being born with FIS in the Fell pony breed was very high (up to 15-20%) as the carrier rate in breeding animals was so high (40-44%), but carriers were not identifiable. Whilst the Dales ponies had a lower carrier rate (20%), there was still real uncertainty for breeders and owners and the very significant chance that a foal with no chance of survival could be produced.

The principal impact of the research is the provision and deployment of a carrier test for Foal Immunodeficiency Syndrome to eradicate this fatal equine disease following the UoL's identification of the genetic defect in 2010 and deployment of the carrier test by two commercial laboratories (http://animaldnadiagnostics.co.uk; www.aht.org.uk/genetics_tests.html#equine). This new diagnostic veterinary tool for risk-free breeding was immediately offered to owners of the two most at-risk breeds (Fell and Dales ponies) leading to rapid reductions in FIS rates.

In addition, the screening programme unexpectedly identified the spread of the disease in two further breeds; Coloured ponies and Cob ponies. Their breed societies have been informed about the presence of carriers in their populations through dissemination of documentation prepared by Carter and encouragement to use the test to ensure safe breeding.

The test was introduced in 2010. Peer reviewed analysis of the diagnostic data in subsequent

years, by Carter [1], clearly indicates a major reduction in the numbers of FIS foals born since 2010, with only one FIS foal born in 2012 in the UK. Owner education via concerted knowledge exchange programmes has been a large part of the success of this work. There is clear evidence from the Fell pony society of FIS-positive stallions being gelded to prevent disease spread [7]. Feedback (verifiable) from the two main breed societies shows that the carrier test was readily welcomed and in some breeders' cases it may have saved their financial viability [7,8]. The need for an effective carrier test can be evidenced by the fact that more than half of all the UK breeding stock of at risk ponies was tested in the first few months of the test being available. This number has reduced in subsequent years as the owners are taking our advice about safe breeding practice [7,8,9].

The beneficiaries of this UoL research, all implemented since 2010, are:

- Vets – who can confidently use the new diagnostic tool to advise owners on risk-free breeding combinations.
- Horse breeders, owners and their families – who can now find out the carrier status of mares and stallions and ensure safe breeding combinations are employed to stop FIS foals being born.
- Equine populations – through reduction in incidence and spread of this fatal disease. The UoL research has identified four breeds that are affected or who are carriers of the mutation and FIS has been reported in Ireland, mainland Europe and the USA as well as in the UK.
- Meat retailers - without this test, this disease would still be spreading silently through equine populations and would be a threat to those who export UK ponies for further breeding and supply to the horse meat trade; only healthy horses can be used for meat.

A leading equine geneticist at the University of Kentucky wrote in an editorial in the Veterinary Record *"This test is clearly a boon for breeders of these pony breeds and the veterinarians who care for them. The story is a model for approaching other hereditary diseases in animals."* [10]. The work has also already been adopted in standard textbooks such as Tizard's "Veterinary Immunology" [11].

These impacts have been achieved through placing a high priority on dissemination with information on the benefits and availability of the carrier test being communicated through a number of media avenues including peer reviewed academic publications and popular publications such as Horse and Hound. Carter has made many presentations to breeders' societies in meetings across the country and hosted an open day at the UoL Veterinary Campus (2010) to ensure the test availability became general knowledge and that owners understood how to use the results to ensure no ill foals were produced. Carter was interviewed on television and radio and produced articles for the equine owner press. Many equine websites (20+) championed the research throughout the 15 years and now broadcast the availability of the carrier test (e.g. [12]). This has been instrumental to the virtual eradication of the disease in the UK.

5. Sources to corroborate the impact

Two main horse breeds that have been seriously affected have benefited. The research effort and impacts can be verified by the chair of each breed society or the chair of the charities funding our work.

7. Contact: **Fell Pony Society**

8. Contact: **Dales Pony Society**

9. Contact: **Fell Pony 2000**

Each source listed below provides evidence for the corresponding numbered claim made in section 4 (details of the impact).

10. Bailey E. Vet Rec (2011) 169: 253-254. doi: 10.1136/vr.d8118

11. Tizard I. "Veterinary Immunology" (2012) (9th Edn) (p444 – 5). ISBN-13: 978-1455703623

12. Equine Science Update (http://www.equinescienceupdate.com/articles/fistr.html)