Ref: 05/JF

Project Title: Develop and validate a Luminex assay to serotype all AHSV strains within a single test and integrate this assay into diagnostic repertoire

Supervisors: John Flannery, Martin Ashby, Paulina Rajko-Nenow **Research group:** Non-Vesicular Reference Laboratories

Project Summary:

The Luminex xMAP technology has the capacity to allow for a high throughout multiplexing tool. Such a technology has advantages in situations where disease caused by different pathogens can't be clinically differentiated, or in situations where numerous serotypes of a particular pathogen circulate. This project will concern the development of an xMAP assay for the detection and differentiation of all AHSV serotypes. This will aid in the understanding of AHS epidemiology and will facilitate rapid diagnosis and timely deployment of vaccine.

Details:

The primary purpose of the Non-Vesicular Reference Laboratory (NVRL) is to provide a diagnostic service and characterise outbreaks of livestock diseases caused by Bluetongue virus (BTV), African horse sickness virus (AHSV), African swine fever virus, Peste des Petits Ruminants Virus, Rinderpest virus and Capripox viruses. The NVRL also performs applied research such as the development and validation of new diagnostic assays as well as characterising and investigating the molecular epidemiology of arboviruses. Our group also maintains the Orbivirus Reference Collection which is a unique collection of dsRNA virus isolates.

Recent changes in the epidemiology of BTV has fueled concerns regarding the potential incursion of other orbiviruses into Europe such as AHSV, which is also transmitted by the *Culicoides* midge. AHSV infects all equine species and is often fatal in horses and mules. A potential outbreak of AHSV in Europe could be a major threat to the equine sport and companion animal industries.

Viruses within the Orbivirus genus, family *Reoviridae,* are characterised by high genetic diversity and the existence of multiple serotypes. Currently AHSV serotype is determined using serotype-specific RT-qPCR assays for which there are 9 assays available. Whilst serotyping is guided by epidemiological information, a single AHSV sample would have to be typed against 9 separate assays to truly exclude the presence of unexpected serotypes or to assess possible dual and triple infections.

Luminex technology enables the quantification of multiple nucleic acids within a single sample by tagging specific primers to uniquely-labelled microspheres, making each bead differentiable from another when read on respective Luminex instruments. Hence Luminex technology would offer a new approach to detect all serotypes within a single sample simultaneously. Although the technology is more expensive during initial establishment and validation, the cost of analysing a sample for all AHSV serotypes would be significantly lower than using the respective separate RT-qPCR assays. The overall cost of this technology will not make it a front-line test for every sample submitted. Instead we are envisaging the technology as a screening assay for samples submitted from areas with multiple circulating strains.

During this one-year placement, the student will be expected to implement and validate a Luminex assay for the detection of multiple AHSV serotypes within a single sample. The student will be under direct supervision during the initial training the high-containment facility then she/he will be expected to work independently. In addition, the student will support the NVRL in routine duties such as housekeeping, consumable management, sample reception and other duties as required. This placement will provide the successful student with a unique opportunity to work in an ISO/IEC 17025-accredited Reference Laboratory and to gain practical knowledge of diagnostic techniques.

References for Suggested Reading:

1. Carpenter, Simon, Philip S. Mellor, Assane G. Fall, Claire Garros, and Gert J. Venter. "African horse sickness virus: history, transmission, and current status." *Annual review of entomology* 62 (2017): 343-358.
2. Reslova, Nikol, Veronika Michna, Martin Kasny, Pavel Mikel, and Petr Kralik. "xMAP technology: applications in detection of pathogens." *Frontiers in microbiology* 8 (2017): 55.
3. Aklilu, N., Batten, C., Gelaye, E., Jenberie, S., Ayelet, G., Wilson, A., ... & Bachanek-Bankowska, K. (2014). African Horse Sickness Outbreaks Caused by Multiple Virus Types in E thiopia. *Transboundary and emerging diseases*, *61*(2), 185-192.

To Apply:
Please email your CV (no more than two sides of A4) and a covering letter detailing why you would like to undertake the placement and the knowledge and skills that you will bring to the Institute to yvonne.walsh@pirbright.ac.uk.
Closing date to apply: 31.01.20