Ref: 03/CN
Project: African swine fever virus vaccine correlates of protection

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Research Group: ASF Vaccinology

Project Summary:

Pig farmers are threatened by an epidemic of a lethal haemorrhagic disease of domestic swine and wild boar, which is spreading through Eastern Europe called African swine fever. A viral vectored vaccine against African swine fever has been developed at Pirbright, but further development of this is hindered by a lack of understanding of the mechanisms of immunity and reliable surrogates of protection. The vaccine induces both lymphocytes and antibodies that recognise whole virus, but the relative importance of the response to the individual proteins that make up the vaccine is unclear. In addition we do not know which types of lymphocytes, in particular T-cells, recognise the virus either. Therefore, the laboratory work in the project will consist principally of phenotyping immune cells using flow cytometry and developing ELISAs to define the antibody response. The results from the project will contribute towards refining a novel viral vectored vaccine against African swine fever.

Details:

Pigs immunised with a viral vectored African swine fever virus (ASFV) vaccine are protected against lethal challenge by virulent virus. Our aim is to identify which of the proteins are required for protection and so reduce the complexity of the vaccine. Analysis of samples taken from these animals show the development of both an antibody and cellular immune response to ASFV. Lymphocytes collected from these animals recognise whole virus and secrete a cytokine called interferon gamma, however we do not know which T-cell subsets are responsible for this and whether the same cells secrete other cytokines (multi-functionality). Similarly, we don't know which of the proteins that make up the vaccine are recognised by which T-cells. Serum from the animals contain antibodies that recognise cells infected with ASFV, however we do not know which proteins these antibodies recognise. Therefore, the project will focus on two separate areas:

1) Identify T-cell subsets that respond to ASFV and the individual antigens. Flow cytometry assays and reagents are available in the lab, frozen cells from three animal experiments are available and new experiments are scheduled for 2018 which are likely to overlap with the time that the student will be at Pirbright. The student will have the opportunity to design further flow cytometry panels to enable discrimination of effector and memory T-cells.

2) Generate recombinant proteins to viral antigens that are not available as yet. Bacteria expressing viral proteins are available and the student can purify proteins from bacterial culture and use them to develop ELISAs to help understand the antibody response. Proteins and ELISAs for the other two proteins have already been established in the lab and are available for the project. These proteins can also be used to examine the T-cell response.

The student will be trained in all of the techniques and methods required to complete the project. The student will be trained to work in high containment, will gain experience of cell culture as well as the techniques outlined above. The laboratory is based in a multi-user lab shared with three other groups all working on viral diseases of swine.

References for Suggested Reading:

- Jancovich JK, Chapman D, Hansen DT, Robida MD, Loskutov A, Craciunescu F, Borovkov A, Kibler K, Goatley L, King K, Netherton CL, Taylor G, Jacobs B, Sykes K, Dixon LK (2018). Immunization of Pigs by DNA Prime and Recombinant Vaccinia Virus Boost To Identify and Rank African Swine Fever Virus Immunogenic and Protective Proteins. *Journal of Virology*. 92(8) doi: 10.1128/JVI.02219-17.
- Takamatsu HH, Denyer MS, Lacasta A, Stirling CM, Argilaguet JM, Netherton CL, Oura CA, Martins C, Rodríguez F. (2013) Cellular immunity in ASFV responses. *Virus Research* 173:110
- Gerner W, Talker SC, Koinig HC, Sedlak C, Mair KH, Saalmüller A (2015) Phenotypic and functional differentiation of porcine αβT cells: current knowledge and available tools. *Molecular Immunology*. 66(1):3-13