Animal Welfare Foundation

GP West Fund - Final Report Form

Primary Contact for Original Application	
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Grant Awarded	
Date	17/01/2018
Title of study	Using modern genomics to better understand the causes of gastrointestinal and respiratory disease in cats

Grant Report

Project summary and impact

250 words max.

Including:

- A brief, non-technical summary of project outcomes and achievements.
- The contribution the project has/ will continue to make to the improvement of animal welfare.

The project aimed to use a technique called next generation sequencing (NGS) to better understand the role of known and yet-to-be-identified infectious agents in feline respiratory and gastrointestinal disease. To achieve this aim we sequenced 60 samples from cats with signs of either respiratory or gastro-intestinal disease. We obtained 30 full-length or near full-length genomes from 7 viral pathogens and partial sequences from many more viruses and bacteria.

The project yielded 12 full-length feline calicivirus (FCV) genomes. These will contribute to ongoing surveillance of the evolution of FCV and help determine whether current vaccines are still appropriate. We also produced full-length or near full-length genomes of feline coronaviruses (FCoV, 3 genomes) and feline parvoviruses (FPV, 4 genomes). These viruses are already known to cause diarrhoea in cats, however, our sequencing has increased the number of available genomes in GenBank (the first complete genomes for UK FPV isolates). These sequences are useful for the continued surveillance of these diseases in UK cats and will be particularly useful if an outbreak of disease occurs. We found partial genomes from viruses for which there is evidence of a possible role in gastro-enteritis in cats but which are not generally currently tested for and not part of core feline vaccines (feline kobuvirus and feline astrovirus). These viruses were found in faecal samples from cats with diarrhoea which tested negative for known gastro-enteric pathogens and suggest the need for further studies to ascertain their prevalence in the UK cat population. Interestingly, we also found a number of novel viruses (with similarity to picobirnaviruses and picornoviruses). These may be emerging viruses in the cat, however, sequencing of better quality samples would be needed to produce longer

genomes for these viruses to help determine whether they are indeed novel viruses and further studies would again be needed to determine their prevalence in UK cats.

In this project we have also sequenced samples from an outbreak of haemorrhagic vomiting in a cat shelter. No definitive disease causing pathogen was identified, however, potential candidates to test for in future outbreaks were identified (feline rotavirus, *Clostridium* spp, *E. coli*) and the methods of sequencing and analysis used here will be applied to future outbreaks.

The project has sequenced known and novel pathogens and has produced 30 full length or near full length genomes which will be submitted to GenBank either in the near future or after publication of the data. We have forwarded relevant sequences to the diagnostic lab which provided samples in the project to enable them to design diagnostic tests to those pathogens they see as important. We have also gained the expertise needed to sequence cat samples and analyse the large amounts of data that NGS produces. We now have the expertise for continued surveillance of cat pathogens using NGS and can use this knowledge in future outbreaks of disease in cats and other small animals to continue to improve the welfare of animals in the UK.

Project evaluation against original hypothesised outcomes

Including:

- Actual versus expected outcomes (including statistical analysis).
- Details of any clinical trials and their key outcomes.
- How practical implementation of your findings will have a direct effect on improvement of animal welfare.

It was estimated we would sequence 20-40 samples in this project. We have recently found that FCV isolates can be multiplexed and sequenced together and the genomes can be untangled bioinformatically following sequencing. This meant we could pool FCV isolates and therefore sequence more than originally planned. In addition, results published by other workers have shown that faecal samples can also be pooled and the number of sequencing reads produced still be of sufficient depth to be able to identify pathogen signatures in the samples. Using sample pooling we were therefore able to sequence 60 samples. This enabled us both to produce more genomes for known viruses and to potentially increase the number of novel viruses detected, giving us more information on the types of pathogens currently causing disease in UK cats.

Project challenges or barriers

Including:

- Any changes made to original project focus, scope, delivery, schedule or evaluation.
- The reason for these changes.

The project aimed to recruit up to four cat rescue shelters. Despite a nationwide appeal by placing an advert in the Veterinary Record, we only received samples from an outbreak in one shelter and from only four affected cats. Whilst we were pleased that this possibly meant outbreaks were not occurring, it limited the number of samples coming from shelter outbreaks.

We also aimed to collect respiratory samples from diagnostic labs to look for potential novel respiratory pathogens. Unfortunately this was not possible as the labs required the whole sample received for testing for their purposes.

Despite these problems we have still extracted the genetic material from 60 samples and by using a pooling strategy have been able to send all of these for NGS sequencing within the budget of the grant.

Please provide details of any additional, work or accolades that have resulted directly from the work supported by this grant

Including:

- Original peer-reviewed research papers.
- Books, book chapters or monographs.
- Intellectual property activity.
- Prizes, awards or commendation, including those awarded to grant funded staff.

Dr Shirley Bonner who carried the work on this grant received a promotion in part recognition for the quality of the work carried out on this and other recent projects.

The pipeline that has in part been developed with AWF funding has now been incorporated as part of national preparedness for companion animal outbreaks in SAVSNET. This contributed to SAVSNETs recent receipt of the BBSRC societal impact team award for 2019.

Following on from the methods developed with AWF funding, SAVSNET has been able to secure additional funding for these kind of studies from the DogsTrust.

How have the outcomes of the project been communicated?

Including:

- The journals/venues at which findings have or will be published or presented.
- Any other routes you have or will use to publicise the outcome of your work

We expect to submit at least 1 publication based on the findings - a paper describing the evolution of FCV and whether existing vaccines are still relevant to current FCV isolates.

We may also publish on the novel viruses and the role of e.g. astrovirus in diarrhoea but this will need further work.

Those genomes of appropriate quality will be published on the international sequence database GenBank, where they will be accessible to all researchers working in the field of feline virology.