WOAH Collaborative Centre Reports Activities 2023

Activities in 2023

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Centre Information

Title of WOAH Collaborating Centre	Viral Genomics and Bioinformatics
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Name (including Title and Position) of Head of the Collaborating Centre (WOAH Contact Point):	Prof Massimo Palmarini
Name of the writer:	Prof David L Robertson

TOR1 AND 2: SERVICES PROVIDED

1. Activities as a centre of research, expertise, standardisation and dissemination of techniques within the remit of the mandate given by WOAH

Category	Title of activity	Scope
Disease control (true)	Membership of national and international consortia	SARS-CoV-2 is continuing to evolve, and after the success of G2P-UK, 'G2P – Global Knowledge Exchange to Enable In Country Risk Assessment of SARS CoV-2 Variants' was established in 2022. Taking advantage of established working partnerships between teams in the UK, Africa and India, G2P-Global comprises 4 G2P-UK members, including the CVR, and 4 established LMIC partners. G2P-Global aims to implement standardised methodologies that enable rapid in-country risk assessment of the biological and antigenic properties of SARS-CoV-2 variants of concern (VOCs), undertake discovery-led molecular, cellular and in vivo analyses of variant phenotypes, assess the potential for spill-overs from animal species, and establish communication networks and laboratory resources that will build technical and logistical preparedness for G2P-Global partners and additional collaborators. The CVR will play a key role in achieving these aims by assessing variant pathogenesis in vivo, employing reverse genetics to generate recombinant viruses to understand the genetic basis of pathogenesis and transmission differences between variants, and providing training in bioinformatics analyses. - Capacity building consortia CVR researchers are co-investigators in a Bill and Melinda Gates Foundation-funded multi-pathogen national sero-surveillance consortium in Malawi, in partnership with the Malawi-Liverpool-Wellcome Clinical Research Programme (MLW), Malawi Epidemiology and Intervention Research Unit (MEIRU), LSHTM, and

		UCL. In addition, the CVR co-leads a Wellcome Trust-funded longitudinal community serosurveillance study in an urban and rural cohort in Malawi (2020-2022), in collaboration with MEIRU, the Public Health Institute of Malawi (PHIM), and Malawi Ministry of Health. CVR researchers also have strong links with the MRC/UVRI & LSHTM Uganda Research Unit in Entebbe, Uganda.
Training, capacity building (true)	Viral Bioinformatics and Genomics Training Course	Viral genomics and bioinformatics deliver bespoke training to a range of CVR trainees, including postdoctoral researchers and PhD students, as well as external training to international partners with the aim of stimulating collaborations, supporting capacity building and increasing the CVR's response capacity. The 2023 external Virus Genomics and Bioinformatics course ran in Glasgow in August. The focus is on Linux/Unix command line analysis and using software for analysing NGS sequencing data. Topics include: data formats, read cleaning, de novo sequence assembly and reference alignment, consensus and variant calling, multiple alignment and phylogenetics, relevant statistics, metagenomics and virus discovery, and transcriptomics. The team also play a major role in designing and delivering external Viral bioinformatics and genomics courses in collaboration with Wellcome Connecting Science. Since 2022, see https://coursesandconferences.wellcomeconnectingscience.org/our-events/global-training/.
Zoonoses (true)	Evolution and transmission of SARS-CoV-2	- Development of online tools and apps for tracking SARS-CoV-2 mutations and variants Our genomics and bioinformatics team had a leading role in developing SARS-CoV-2 dashboards. Using our virus resource dedicated platform, GLUE, we were able to develop a mutation centric resource, COV-GLUE (https://cov-glue.cvr.gla.ac.uk), which was set up online by early February 2020. This was one of the first resources to share access to mutation data from the accumulating SARS-CoV-2 genome sequences in GISAID. SARS-CoV-2 genome sequences in non-human species were monitored (numbers, mutations etc.). This information was fed into the WOAH SAR-CoV-2 expert group. - SARS-COV-2 evolution and links to animal origin Our bioinformatics team has participated in landmark evolutionary studies to understand the origins of SAR-CoV-2. These include papers on SARS-CoV-2 phylogenetic relationships to the horseshoe bat virus reservoir once recombination had been accounted for (doi: 10.1038/s41564-020-0771-4; doi: 10.1093/gbe/evac018), expert reviews of the available evidence (https://doi.org/10.1016/j.cell.2021.08.017) and the strong link of the early SARS-CoV-2 cases to the Huanan market (doi: 10.1126/science.abp8715), confirming emergence via the live-animal trade much like the first SARS virus (doi: 10.1126/science.abh011). - SARS-CoV-2 in cats A CVR team demonstrated that human-to-cat transmission of SARS-CoV-2 occurred during the first wave of the COVID-19 pandemic in the UK, with infected cats displaying mild to severe respiratory disease (doi: 10.1002/vetr.247). Further studies are being conducted to assess the spread of SARS-CoV-2 in cats in the UK in subsequent waves. Analysis of global feline SARS-CoV-2 sequences from the GISAID database, has shown that convergent mutations observed in feline sequences are widespread in the human population. It is likely that these evolved in humans and are not associated with feline adaptation. Publications include: doi: 10.1002/vetr.247; doi: 10.3390/v15030731; doi: 10.3201/eid2906.221755; doi: 10.3390/v15081769. - SARS-CoV-2 in mink/ferrets SARS-CoV-2 has a broad mammalian species tropism infecting humans, cats, dogs, and farmed mink. Since the start of the pandemic, several reverse zoonotic outbreaks of SARS-CoV-2 have occurred in mink, one of which reinfected humans and caused a cluster of infections in Denmark. We investigated the molecular basis of mink and ferret adaptation and demonstrated that the spike mutations Y453F, F486L, and N501T all specifically adapt SARS-CoV-2 to use mustelid ACE2. Furthermore, we risk assessed these mutations and concluded that mink-adapted viruses are unlikely to pose an increased threat to humans, as Y453F attenuates the virus replication in human cells and all three mink adaptations have minimal

		antigenic impact. Finally, we showed that certain SARS-CoV-2 variants emerging from circulation in humans may naturally have a greater propensity to infect mustelid hosts and therefore these species should continue to be surveyed for reverse zoonotic infections (doi: 10.1016/j.celrep.2022.110344).
Zoonoses (true)	Studies on rabies	Rabies virus (RABV) is a neglected zoonotic disease that causes tens of thousands of human deaths each year, with a near 100% mortality rate after the onset of symptoms. The virus is a member of the Lyssavirus genus, within the Rhabdoviridae family, which is characterised by a single stranded, negative-sense RNA genome. Vampire bat transmitted rabies virus is an agriculturally important and pervasive zoonotic threat in much of North, Central and South America. Studies have included longitudinal monitoring of and/or field experiments in wild bats and livestock in Peru (since 2007), Belize (since 2014) and Costa Rica (since 2022), viral genomics and phylogenetics, and spatiotemporal modelling of emergence risk. With Dr Kirstyn Brunker we have developed an online resource (RABV-GLUE - http://rabv-glue.cvr.gla.ac.uk/#/home) which provides an analysis tool providing genotyping, analysis and visualisation of submitted FASTA sequences. The data is organised in a database of RABV sequences and metadata from NCBI, updated daily and arranged into major and minor clades. Pre-built multiple-sequence alignments of NCBI sequences, can be downloaded in user-defined sections. Notable recent publications from our work on rabies include: doi: 10.1098/rspb.2023.1739; doi: 10.1073/pnas.2216667120; doi: 10.1126/sciadv.add7437; doi: 10.1098/rspb.2022.0860.
Zoonoses (true)	Studies on Bluetongue Virus	Bluetongue virus (BTV) is a vector-borne virus within the Orbivirus genus of the Reoviridae family. It causes the bluetongue disease in various common domestic ruminant and wild animal species; an outbreak starting in 2006 threatened livestock industries in northern Europe. Our online resource includes: a database of BTV sequences linked to isolates, with curated metadata and pre-built multiple sequence alignments for all 10 segments, which may be downloaded in user-defined section, and an analysis tool providing genotyping, and visualisation of submitted segment 2 sequences. (http://btv-glue.cvr.gla.ac.uk/#/home). We are currently advising Marc Guimera and Caroline Wright from the Pirbright Institute on experimental design and associated bioinformatic analyses of superinfections and reassortment in BTV. We have also developed a machine learning framework to identify the correlates of diseases severity in acute arbovirus infection, using bluetongue virus as model system. (https://biorxiv.org/cgi/content/short/2024.02.23.581333v1).
Zoonoses (true)	Collaboration on IBV	Infectious Bronchitis is the most economically important infectious disease affecting poultry globally. The CVR has a collaboration with the Pirbright Institute titled 'Investigating Host and Viral Factors for Improved Design of Future Live Attenuated Vaccines for IBV'. (https://gow.bbsrc.ukri.org/grants/AwardDetails.aspx?FundingReference=BB/V016067/1) from the BBSRC. The Pirbright have shown previously that attenuation of the avian coronavirus, infectious bronchitis virus (IBV) by serial egg passaging is likely a multifactorial process rather than being driven by a single pathway. Little is known regarding the mechanisms underpinning this process. The research from this grant will use deep learning methods to identify patterns within existing whole genome sequence datasets generated during egg passaging of 2 viruses (QX and M41-CK) to attenuation. We will then generate novel sequence information for whole S gene/whole IBV genome to explore virus genome sequence changes occurring post-vaccination. Changes will be compared with pathogenic IBV isolates to link to phenotype. We will use spatial transcriptomics to identify changes in host gene expression within tissues from vaccinated birds. This will identify cellular responses driving virus changes at a cellular level. We will take the outputs of these three components, combining them into a single dataset and process them using deep learning to make final predictions of

		genomic markers or signatures contributing to attenuation and validate these ex-vivo. This work will identify better ways to attenuate viruses for use as live attenuated vaccines, whilst minimising the potential for reversion to virulence.
Zoonoses (true)	ICTV's Virus Taxonomy report	David Robertson has been a member of the ICTV Executive Committee, since 2020. Richard Orton is a member of the ICTV Herpesvirales study group and Joseph Hughes is a member of the ICTV Parvoviridae study group.
Zoonoses (true)	Flu: TrailMap-One Health	The CVR's Dr Ed Hutchinson and others are part of a consortium taking a multipronged approach to risk assess in depth the current clade 2.3.4.4b H5N1 avian influenza viruses for human spillover infection and pandemic potential. The contemporary viruses will be compared with those of the early 2000s, and with other influenza viruses that did cause human pandemics in 1968 and 2009. State of the art approaches will be used to study virus/host molecular interactions, and define how these vary with different isolates of the clade 2.3.4.4b virus and between different host species. The interactions the virus makes with the human airway from children and adults will be considered, to understand who is most likely to be infected by and transmit the virus and who is most at risk of disease. Modelling approaches will be incorporated to inform surveillance, asking where and how the virus is most likely to infect mammals that could serve as intermediate hosts. Systems by which mitigations such as antiviral drugs or vaccines could be assessed will be developed, if the virus were indeed to jump species.

TOR3: HARMONISATION OF STANDARDS

2. Proposal or development of any procedure that will facilitate harmonisation of international regulations applicable to the main fucus area for which you were designated

Proposal title	Scope/Content	Applicable area

3. In exercising your activities, have you identified any regulatory research needs* relevant for WOAH?

Yes

Research need : 1

Please type the Research need: Regulatory research is needed on the standardisation and harmonisation of whole viral genomes sequencing and associated standardisation of bioinformatic analyses. Guidelines on the sharing of raw data and transparency of the tools used for consensus sequence generation are needed.

Relevance for WOAH Standard Setting,

Relevance for the Codes or Manual Manual,

Field Diagnostics, Vaccines,

Animal Category Terrestrial,

Disease:

Kind of disease (Zoonosis, Transboundary diseases) Zoonosis,

If any, please specify relevance for Codes or Manual, chapter and title

(e.g. Terrestrial Manual Chapter 2.3.5 - Minimum requirements for aseptic production in vaccine manufacture)

Answer: Standards for high throughput sequencing, bioinformatics and computational genomics for the WOAH Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (pending - under review by editor)

Notes:
Answer:

4. Did your Collaborating Centre maintain a network with other WOAH Collaborating Centres (CC), Reference Laboratories (RL), or organisations designated for the same specialty, to coordinate scientific and technical studies?

Yes

Name of WOAH CC/RL/other organisation(s)	Location	Region of networking Centre	Purpose
Padova WOAH Centre, Italy Pirbright Institute, UK	Italy	Europe	Research led by scientists at the CVR, and linked to the Padova WOAH Centre, identified the human gene BTN3A3, which is commonly expressed in our airways, as a key human defence against avian flu. Through a series of extensive tests, we were able to show that the BTN3A3 gene is vital to protecting humans against avian flu, as most strains of the virus cannot get past its defences. We were able to show that avian flu viruses like H7N9 (which has infected more than 1,500 individuals with 40% case fatality rate) have a genetic mutation that allows them to 'escape' the blocking effects of the BTN3A3 gene. Tracking the history of influenza pandemics in humans, we were also able to link BTN3A3 resistance with key influenza virus types. All the human influenza pandemics, including the devastating 1918-19 global flu pandemic and the swine flu pandemic in 2009 were caused by influenza viruses that were resistant to BTN3A3. As result, this study suggests that having resistance to this gene may be a key factor in whether any flu strain has human pandemic potential. Pinto, R.M., Bakshi, S., Lytras, S. et al. BTN3A3 evasion promotes the zoonotic potential of influenza A viruses. Nature 619, 338–347 (2023). https://doi.org/10.1038/s41586-023-06261-8 With researchers at the UK Pirbright Institute we are members of the UK G2P2 (SARS-CoV-2 focussed) and TrailMap-OneHealth (influenza focussed) consortia.

TOR4 AND 5: NETWORKING AND COLLABORATION

5. Did your Collaborating Centre maintain a network with other WOAH Collaborating Centres, Reference laboratories, or organisations in other disciplines, to coordinate scientific and technical studies?

Yes

Name of WOAH CC/RL/other organisation(s)	Location	Region of networking Centre	Purpose
WOAH Expert Group on SARS-CoV-2	Global	Europe	Richard Orton is a member of this group.
WOAH Emerging Diseases Group	Global	Europe	Richard Orton is a member of this group.

TOR6: EXPERT CONSULTANTS

6. Did your Collaborating Centre place expert consultants at the disposal of WOAH?

Yes

NAME OF EXPERT	KIND OF CONSULTANCY	SUBJECT
Massimo Palmarini	Expert	Avian influenza viruses - Virus and host genetics determinants of cross-species transmission
Pablo Murcia	Expert	Molecular virology of influenza viruses
David Robertson	Expert	Computational and evolutionary analysis of viruses
Richard Orton	Expert	Computational analysis of virus genome sequence data
Joseph Hughes	Expert	Virus evolution and bioinformatic training
Ana da Silva Filipe	Expert	Sequencing strategies for animal and human viruses

TOR7: SCIENTIFIC AND TECHNICAL TRAINING

7. Did your Collaborating Centre provide advice/services to requests from Members in your main focus area?

Yes

David Robertson provided advice and Richard Orton provided training in bioinformatics data analysis to IAEA's Zoonotic Disease Integrated Action (ZODIAC) project.

8. Did your Collaborating Centre provide scientific and technical training, within the remit of the mandate given by WOAH, to personnel from WOAH Members?

Yes

a) Technical visit : 7

b) Seminars : 0

c) Hands-on training courses: 29

d) Internships (>1 month) : 0

TYPE OF TECHNICAL TRAINING PROVIDED (A, B, C OR D)	CONTENT	COUNTRY OF ORIGIN OF THE EXPERT(S) PROVIDED WITH TRAINING	NO. PARTICIPANTS FROM THE CORRESPONDING COUNTRY
A	Zulma Rojas, Universidad Andres Bello (Chile) visited as part of the "Environmental and anthropogenic drivers of bat-borne pathogens in Latin America" project.	Chile	1
A	Julius Nziza, Gorilla Doctors (Rwanda) visited as part of the "Pathogen genomic analysis at the primate-human-domestic animal interface in Rwanda: prevalence, diversity and transmission dynamics" project.	Rwanda	1
A	4 researchers from Laboratorio Nacional de Servicios Veterinarios (LANASEVE), Servicio Nacional de Salud Animal (SENASA), Heredia, Costa Rica visited as part of the "Molecular characterization of the rabies virus in animals in the countries that make up the OIRSA".	Costa Rica	4
C	Conservation of viral RNA on FTA cards for viral genomic analysis	9 countries in Latin America and the Caribbean	9
C	2023 Viral Bioinformatics and Genomic Training Course	U.K., Poland, Germany, Algeria, Peru, U.S., Belgium, Switzerland	20
C	Participation in the organisation and delivery of a bioinformatics training workshop in collaboration with the Food and Agriculture Association (FAO). The workshop was held in Bangladesh and focussed on avian influenza virus and foot-and-mouth disease virus. Approximately 25 participants from across Asia were taught how to generate genome consensus sequences from illumina high throughout sequencing data using the command line, and how to create and run workflows on Galaxy web based servers to perform the same analyses.	Asia	25

TOR8: SCIENTIFIC MEETINGS

9. Did your Collaborating Centre organise or participate in the organisation of scientific meetings related to your main focus area on behalf of WOAH?

No

TOR9: DATA AND INFORMATION DISSEMINATION

10. Publication and dissemination of any information within the remit of the mandate given by WOAH that may be useful to Members of WOAH

a) Articles published in peer-reviewed journals:

24

Manali, M. et al. (2023) SARS-CoV-2 evolution and patient immunological history shape the breadth and potency of antibody-mediated immunity. Journal of Infectious

Diseases, 227(1), pp. 40-49. (doi: 10.1093/infdis/jiac332)

Taylor, E. et al. (2023) The Amazonian Tropical Bites Research Initiative, a hope for resolving zoonotic neglected tropical diseases in the One Health era. International Health, 15(2), pp. 216-223. (doi: 10.1093/inthealth/ihac048)

Cox, M. et al. (2023) SARS-CoV-2 variant evasion of monoclonal antibodies based on in vitro studies. Nature Reviews Microbiology, 21(2), pp. 112-124. (doi: 10.1038/s41579-022-00809-7)

Atim, S. A. et al. (2023) Prevalence of Crimean-Congo haemorrhagic fever in livestock following a confirmed human case in Lyantonde district, Uganda. Parasites and Vectors, 16, 7. (doi: 10.1186/s13071-022-05588-x)

Viana, M. et al. (2023) Effects of culling vampire bats on the spatial spread and spillover of rabies virus. Science Advances, 9(10), eadd7437. (doi: 10.1126/sciadv.add7437)

Griffiths, M. E. , Meza, D. K., Haydon, D. T. and Streicker, D. G. (2023) Inferring the disruption of rabies circulation in vampire bat populations using a betaherpesvirus-vectored transmissible vaccine. Proceedings of the National Academy of Sciences of the United States of America, 120(11), e2216667120. (doi: 10.1073/pnas.2216667120)

Carabelli, A. M., Peacock, T. P., Thorne, L. G., Harvey, W. T., Hughes, J. , Peacock, S. J., Barclay, W. S., de Silva, T. I., Towers, G. J. and Robertson, D. L. (2023) SARS-CoV-2 variant biology: immune escape, transmission and fitness. Nature Reviews Microbiology, 21(3), pp. 162-177. (doi: 10.1038/s41579-022-00841-7)

Simmonds, P. et al. (2023) Four principles to establish a universal virus taxonomy. PLoS Biology, 21(2), e3001922. (doi: 10.1371/journal.pbio.3001922)

Kuhlmeier, E. et al. (2023) Detection and molecular characterization of the SARS-CoV-2 Delta variant and the specific immune response in companion animals in Switzerland. Viruses, 15(1), 245. (doi: 10.3390/v15010245)

Hardy, A. et al. (2023) The timing and magnitude of the type I interferon response are correlated with disease tolerance in arbovirus infection. mBio, 14(3), e0010123. (doi: 10.1128/mbio.00101-23)

Whitlock, A. O. B. et al. (2023) Identifying the genetic basis of viral spillover using Lassa virus as a test case. Royal Society Open Science, 10(3), 221503. (doi: 10.1098/rsos.221503)

Tyson, G. B., Jones, S., Logan, N., McDonald, M., Marshall, L., Murcia, P. R. , Willett, B. J. , Weir, W. and Hosie, M. J. (2023) SARS-CoV-2 seroprevalence and cross-variant antibody neutralization in cats, United Kingdom. Emerging Infectious Diseases, 29(6), pp. 1223-1227. (doi: 10.3201/eid2906.221755)

Shepherd, J. G. , Davis, C. , Streicker, D. G. and Thomson, E. C. (2023) Emerging rhabdoviruses and human infection. Biology, 12(6), 878. (doi: 10.3390/biology12060878)

Siddell, S. G. et al. (2023) Virus taxonomy and the role of the International Committee on Taxonomy of Viruses (ICTV). Journal of General Virology, 104(5), 001840. (doi: 10.1099/jgv.0.001840)

Pinto, R. M. et al. (2023) BTN3A3 evasion promotes the zoonotic potential of influenza A viruses. Nature, 619(7969), pp. 338-347. (doi: 10.1038/s41586-023-06261-8)

Tyson, G. B. et al. (2023) Increase in SARS-CoV-2 seroprevalence in UK domestic felids despite weak immunogenicity of post-Omicron variants. Viruses, 15(8), 1661. (doi: 10.3390/v15081661)

Mollentze, N. and Streicker, D. G. (2023) Predicting zoonotic potential of viruses: where are we? Current Opinion in Virology, 61, 101346. (doi: 10.1016/j.coviro.2023.101346)

Jones, S. et al. (2023) SARS-CoV-2 in domestic UK cats from Alpha to Omicron: Swab surveillance and case reports. Viruses, 15(8), 1769. (doi: 10.3390/v15081769)

Hartmann, K. et al. (2023) Feline injection-site sarcoma and other adverse reactions to vaccination in cats. Viruses, 15(8), 1708. (doi: 10.3390/v15081708)

Carrozza, M.-L., Niewiadomska, A.-M., Mazzei, M., Abi-Said, M. R., Hue, S., Hughes, J. , Gatseva, A. and Gifford, R. J. (2023) Emergence and pandemic spread of small ruminant lentiviruses. Virus Evolution, 9(1), vead005. (doi: 10.1093/ve/vead005)

Rojas-Sereno, Z. E., Streicker, D. G. , Suarez-Yana, T., Lineros, M., Yung, V., Godreuil, S. and Benavides, J. A. (2023) Detection of antimicrobial-resistant enterobacterales in insectivorous bats from Chile. Royal Society Open Science, 10(11), 231177. (doi: 10.1098/rsos.231177)

Pennisi, M. G. et al. (2023) Feline morbillivirus: clinical relevance of a widespread endemic viral infection of cats. Viruses, 15(10), 2087. (doi: 10.3390/v15102087)

Lytras, S. et al. (2023) Resurrection of 2'-5'-oligoadenylate synthetase 1 (OAS1) from the ancestor of modern horseshoe bats blocks SARS-CoV-2 replication. PLoS Biology, 21(11), e3002398. (doi: 10.1371/journal.pbio.3002398)

Hüttl, J. et al. (2024) Serological and molecular investigation of SARS-CoV-2 in horses and cattle in Switzerland from 2020 to 2022. Viruses, 16(2), 224. (doi: 10.3390/v16020224)

b) International conferences:
0

c) National conferences:
1
- Evolution, Pathogens and Public Health meeting - 18th October 2023 at the University of Glasgow
• The Evolution, Pathogens and Public Health meeting supported by the Genetics Society was a networking meeting of research scientists from public health agencies and academia. Many new interactions between public health agencies and academia were created during the COVID-19 pandemic and we felt it was important to maintain the network that formed during the pandemic research response. The meeting was fittingly held in the Advanced Research Centre, a new building on the University of Glasgow main campus built to host cross-disciplinary research.
• Fifty participants attended the meeting with an equal split between researchers from academia and public/clinical health. Talks covered a range of different infectious diseases of public health importance such as mpox, SARS-CoV-2, avian influenza, HIV and rabies and the discussions shed light on the importance of genomics in public health preparedness and response.

d) Other (Provide website address or link to appropriate information):
2
We have developed some online resources -
RABV-GLUE - http://rabv-glue.cvr.gla.ac.uk/#/home
BTV-GLUE - http://btv-glue.cvr.gla.ac.uk/#/home)

11. What have you done in the past year to advance your area of focus, e.g. updated technology?
Our lab has handled waste-water samples in the past, to assess amplicon-based sequencing methods to quantify SARS-CoV-2 variants. This was a pilot study done in collaboration with the Scottish Environment Protection Agency (SEPA) to generate preliminary data and we now have funding for some further studies.

12. Additional comments regarding your report: