Royal Veterinary College
University of London

Minutes: AWERB meeting

Status: Chair approved

Meeting held: 09 December at 10am by MS Teams

Present
Attendees: 8 plus 1 in attendance, plus 4 by invitation and 7 apologies.

1 THANK YOU TO EXTERNAL LAY PANEL MEMBER

It was noted that this was the external lay panel member's last AWERB meeting as she would be moving onto another committee for 2021. The member was thanked for her contribution over the year to AWERB – her input as a lay person member had been very valuable. Another external lay panel member was scheduled to re-join AWERB.

2 PPL PRESENTATION: NEW PROJECT LICENCE APPLICATION

The project licence holder was welcomed to the meeting. It was explained that they were applying for a new project licence to replace their existing licence which had secondary availability at the RVC. As this would be their third project licence, the project licence holder was asked to provide an update on how the research had progressed so far and what the new project licence would be building upon. They were also asked what had been learnt from the work with the animals and how they should be treated in order to improve their welfare and minimise their use.

The project licence holder explained that the overall purpose of their research programme was to better understand how the brain functioned to facilitate active listening. From their current project licence they had made real progress in three areas:

(1) **How visual stimuli can change the way in which neurons in auditory cortex process sound mixtures**. This was work that had been motivated by behavioural observations made in humans. The researchers were actively exploring training paradigms that helped listeners to use visual information more effectively. This training was feeding back into motivating the animal work to understand the neural correlates of the behavioural changes. In this audio visual line of work there was a really fruitful exchange between the clinical human work and the animal work.

(2) Listening in noise: It was explained that they had specifically identified a key role for auditory cortical in this process. They had been able to document auditory cortical spatial receptive fields in freely moving, naturally foraging animals for the first time, yielding novel insights into the coordinate frame in which auditory space was represented. They had also explored hearing in noise, providing the first demonstration that auditory cortex plays a causal role in listening in complex situations.

They had developed a novel behavioural paradigm in which animals listen to a continuous stream of speech in order to identify a target word. This would form a bedrock of future work exploring how perceptual invariance and selective attention operate in the auditory cortex when processing complex sounds. Through collaboration with auditory cognitive neuroscientists they

had developed the first animal paradigm for statistical regularity detection, demonstrating that like humans, these animals were able to detect statistical regularity in complex tone sequences. This was work that could become useful in terms of understanding how to build better signalling process devices for cochlea implants and hearing aids.

(3) How brain constructs a sense of auditory space: The team had identified that auditory cortices play a key role in integrating with different sound localisation cues. Unlike vision and touch, sensory receptors don't encode position in space, they encode frequency, so the brain constructs this by comparing the sound as it arrives at the 2 ears. One of the things that the researchers had done under the previous licence was to record freely moving animals as they navigated through a sound environment. They had been able to demonstrate that while a lot of cells in the auditory cortex encode sound in a head set of co-ordinates; a sub-population of cells mapped the sound source position. Irrespective of which way the animal was facing, these cells were able to indicate where the sound was (for the example near the door or by the window). The team were working to recreate this information for people with hearing loss. Although MHRA and NICE have now decreed that people could have two cochlea implants, because these implants cannot communicate with each other, they do not provide cues of where the sound has come from. By understanding which cues are important to the brain, and how they construct this sense of space, they were exploring how different hearing aid settings allow this sense of space to be constructed. This was another example of the to and fro link between human and animal work.

With regards to welfare of the animals the following had been learnt: they had published a large number of normative datasets of animal weight changes with season which when used as a proxy for health, enabled them to understand the changes in body weight that they observed, allowing them to be more nuanced in their ability to detect when they should be concerned about an animal in terms of its weight change trajectory.

The project licence holder had a joint PhD project with the RVC which involved studying boredom in animals and what enrichment that could be given to improve their lives. The PhD student had undertaken a large survey (with thousands of responses from the animal owners and neuroscience labs that used these animals from all around the world) that examined the experiences that these animals have. She was now analysing the results. If it was possible to find solutions to boredom, that other labs could implement too, this would be very beneficial.

AWERB asked the following questions:

- Numbers of animals in the application: presumably the numbers of animals that would be used was based on their experience of what they had used for their previous licences? It was confirmed that was the case. This was the third licence that had been written and this had the least amount of animals (with 33% less animals than in the previous licence). Improvement in technology had enabled them to obtain more data from the animals than what they could previously; they were also much better at maintaining the implants and extending their capacity. It was noted that there was a philosophical debate about whether it was better to use fewer animals though more individual animals could be exposed to pain and suffering; or more animals that didn't experience so much pain and suffering. AWERB were advised by one of the NACWOs that for the animals used they did not seem to show any signs of pain and suffering after the implants and didn't even seem to notice that the implants were there and behaved like normal playful animals, happily interacting.

- A query was raised about the use of the word "chronic" in the project licence. Did that mean the implant remained? It was confirmed that was the case.

- As part of a tour to the Ethics and Welfare Committee, the lay panel member reported that they recalled seeing the ferrets and that they had seemed very happy. It was queried whether they were just kept in that one room though. Did they have access to outside space? It was confirmed that unless they were in the lab where the behavioural testing was done; they were kept in the same room. They did have a lot of play time when not in their cages such as a floor pen filled with plastic balls and other novel items. The extra enrichment seemed to benefit the animals and they did not exhibit stereotypical behaviour that might be seen in captive animals in the zoo. It was confirmed that if the animals were stressed it would impact on the scientific recordings. The researchers were interested in how the brain processes sensory stimuli so if they had an animal that did not experience a rich variety of sensations then it would give pretty impoverished data in return.

- A query was asked with the animals now being kept longer, were they seeing any problems with the implants? The project licence holder explained that there were a couple of things that determined how well the implants last: the first one was could they still record signals from them. They had got better at ensuring this because they were more effective in building them; the second thing was lot of the electrode implants were movable and could be moved down but not up so a judgement call had to be made on how much data they got at each step in the cortex. At some point they had to decide that there wasn't any more data they could effectively get. They therefore had to trade off the risk that the implant might cease to function versus getting as much meaningful data as possible. As they had got much better in maintaining the quality of the signals they were able to record from the implants it was now a scientific determinant in trying to get the data from the whole of the cortical depth that set the upper boundary on recording rather than the implant itself.

- A query was raised whether there had been any circumstances of infections causing problems with the implants? There had been just two cases where the implants had come off but these had been due to physical failure rather than an infection problem.

- A query was asked about the humane endpoints for water restriction. It was noted that if an animal whose weight loss approached the maximum permitted, or who exhibited signs of dehydration, would be removed from water regulation and given access to free water. What would happen though if the animal did not then improve or respond in terms of increase of body weight as there was no 2nd stage end point included? Although it was recognised that this might not happen very often, it was suggested that it would be useful if the licence set out what the next step was, even if it was just consult the NVS.

- Another query was raised about the ear plugs. If there was an infection would the ear plugs be removed? It was confirmed that they were and this would be made clearer in the licence.

- It was noted that these animals were susceptible to corona virus. What extra care was being taken to protect them? The NACWO explained that they made sure they used standard alcohol hand sanitisers; had hairnets, masks and were fully gowned up when they went into the room and there was no physical contact. They also did not go in to the room if they had any cold or flu symptoms. The NVS added that they were also checking with the suppliers to see if they were able to screen any new animals before they were bought into the unit. It was noted that APHA were working with corona viruses in relation to mink and could be a group to contact about

screening. APHA have been developing a diagnostic test for the mutated viruses that have recycled back into humans.

The project licence holder was thanked for attending the AWERB meeting to discuss her project licence. Their licence would be discussed further by AWERB and that a written summary of the discussions held would be sent to them. They should be proud of the work that had been done in improving the enrichment provided to the animals but that this was an area that should be continually monitored to see what future improvements could be implemented.

Overall AWERB were of the consensus that this was very valuable work. They had no issues in terms of the implants that were carried out on the animals, for once the surgery had been done they had been reassured that the animals quality of life was not affected. What did need to be monitored was ensuring that the animals did not become institutionalised. This should not happen as they were kept in a group and were able to interact with each other and had a lot of environmental enrichment provided. The life span of this species of animal was generally 6 to 7 years; the average time the animal had an implant was 18 months. The animals could not be rehomed after they have had surgery as under A(SP)A the implants could not be removed once they had been installed so they had to be euthanased.

A query was asked why the animals were transported to another institution for the terminal procedures as that would be a stressful process. It was explained that it was because this institute had the relevant sophisticated equipment and facilities in their labs that the RVC did not have. Data were recorded from the brains of the animals once they were fully anaesthetised and these particular studies were run over a period of time.

AWERB confirmed that once the requested changes had been made to the project licence they were happy for the project licence to be submitted to the Home Office.

3 NEW PROJECT LICENCE APPLICATION: TARGETED TREATMENT OF BLOOD-BORNE DISEASE

The project licence holder and two colleagues were welcomed to the meeting.

It was explained that a new project licence had been written to assist in developing a new novel medical device which was used in an extracorporeal system (outside the body) to remove harmful pathogens from the patient's blood stream. This device had the potential to provide targeted and fast treatments for blood-borne diseases. The technology would enable the selective removal of harmful components, such as cells, bacteria, toxins and inflammatory cytokines directly from a patient's bloodstream. The ability to precisely extract unwanted disease causing substances in this way had the potential to revolutionise the treatment of deadly blood-borne diseases, such as sepsis and COVID-19, leukaemia and malaria. Regulatory animal studies were required prior to human clinical trials, hence why a project licence was required.

The aim of this project licence was to provide data for regulatory submission prior to entering human clinical trials. In addition to safety studies for regulatory submission, efficacy studies would also be performed which would lead to advancement of knowledge in treating these diseases and publication of this.

There was an urgent need for new therapeutic tools to help clinicians reduce the severity and mortality of COVID-19 and the number of patients that end up on ventilators. Clinical studies have shown that much of the harm done in severe cases of COVID-19 results from the body's overactive or hyper-immune response. Using the device to reduce this hyper-inflammatory state without systemic delivery of biologics, would provide an early opportunity to control the immune response without causing long-term immunosuppression. The device could treat both the hyperinflammation seen in the lungs that drives hospitalisation and the secondary sepsis in COVID-19 patients that results in significant mortality.

The following queries were asked:

- Were resources available to perform human trials if the clinical data supported that these should be carried out? It was confirmed that a large COVID-19 innovation grant from Innovate UK for this work was in place.
- Were there similar products already on the market? It was confirmed that this was an innovative approach and they were the first taking this approach into the clinic.
- Why was it proposed to use pigs when the study had already been trialled in sheep? It was explained that the study was split into two protocols: the first related to safety testing and was designed to investigate the safety of the device. This protocol would use healthy animals to show that the system could be used safely in large animals. This protocol would probably be done in sheep for practical reasons: anaesthesia in sheep and the recovery side in the barn. The reason pigs were also included was that two of the outcome measures that needed to be run to fulfil the ISO requirements for the safety testing were quite specific tests. It was possible that one of the ELISA'S that had to be used could only be run on pigs. Pigs were on the licence for the efficacy work because they would be using a pig model that had been set up at the RVC for the past 5 years with another collaborator. There were data showing that it worked and how it worked. The outcome measures from that model would fit completely with using their system to measure the efficacy of their system.
- A query was asked about the control groups in the protocols. What would be used as a control group for them? It was explained that at this stage there was uncertainty if a control would actually be required as it was dependent on discussions with the MHRA and what they required. However, this possibility needed to be built into the licence in case it was requested.

- The project licence specified that the maximum numbers of animals that would be used for protocol 1 was 14 animals. How had this number been calculated? It was explained that between 1 and 4 animals would be used for the pilot studies and a maximum of 10 for the main study. Once the pilot had been done, it would be possible to determine the number of animals for the main study. It was anticipated that it would be 6 however it was dependent on the MHRA and the numbers they required for the pre-clinical trial.

- A query was asked about the recovery studies, did the device stay in the animal or was it removed? The animals would stay on the machine for between 2 to 4 hours (to replicate what would happen in humans) so long as the pilot studies showed that the anaesthesia could be maintained for that long. The animal would then be recovered and blood samples taken on an interim basis. Two weeks later the animal would be euthanised. It was suggested that removal of the device should be added as a step to make that clear. It was explained that there was no installation of an actual device, it just involved a catheter going into and out of the blood stream. This needed to be made clear in the project licence.

- Protocol 1 mentioned that heparin might be administered intravenously at the time of the surgery. Were there any potential adverse effects? It was explained that heparin was widely used in people and animals and the amount was carefully calculated. They routinely had small animals on dialysis machines in the hospital and the main clinician who ran the dialysis would be in the background helping with the project once it got to that stage as well as the anaesthetist that ran the heart bypasses. In terms of heparin, the loading dosage was well known as it was used quite widely in efficacy and inflammatory models already. In terms of the machine and changing heparin levels, it was pretty well defined in protocols for human

and veterinary medicine but would be measured bench-side - it was literally a blood drop clotting time. Heparin levels could be titrated so that the level could be adjusted accordingly in order to minimise the risk of bleeding out.

If this product did work and was then used to help Covid patients that would be an excellent result.

The project licence holder was thanked for attending the meeting. The licence would be discussed further by AWERB and that a written summary of the discussions held would be sent to them.

AWERB noted that the licence was constrained by the regulatory requirements. The device needed to be tested on a large animal model in order to satisfy MHRA requirements before it could go to human clinical trials. The College had the set up to run this project to GLP requirements and there would be a small number of animals used. The research also had potential to help future veterinary patients.

AWERB were not convinced by the answer provided about why two species were needed though. Just because there was no ELISA for sheep seemed to be quite a flimsy reason and should be able to be resolved. As both protocols were suitable for pigs then why did sheep need to be used at all? This question would be asked.

It was confirmed that there were the necessary staff available to observe the animals and give the post-operative care needed. They just needed to be careful what studies were running at the same time.

4 REVIEWING PROJECT LICENCES

It was highlighted that when project licences were downloaded from ASPeL as a PDF, it did not always incorporate the NTS section. For future reviews project licence holders would be asked to download the project licences as word documents. It was noted that the NTS now seemed to be at the beginning of the licences rather than at the end, which AWERB thought was better practice, as it should encourage the project licence holder to focus on why they were doing the research and what the aims were.

5 MINUTES OF PREVIOUS MEETINGS

The minutes of the meeting held on 11 November 2020 were confirmed as an accurate record.

6 ACTION LOG

6.1 Item 2 (November 2020 meeting): Feedback on secondary availability project licence application that had been discussed at the October meeting.

The project licence holder had provided the requested clarification and the project licence had now been approved for submission to the Home Office.

6.2 Item 5 (November 2020 meeting): Ring Tailed Lesions

The ring tail lesions were still occurring in animals in a couple of the rooms. This seemed to be appearing soon after delivery, however early stage ring tail was difficult to see until the symptoms were more severe, so it was difficult to determine whether the animals were arriving with it or developing it upon arrival. Those animals where it was thought that they might develop the condition were being closely monitored. It was not yet known what proportion of animals were infected but this would be found out. It was a small number but significant enough to notice. These lesions could be due to the environmental conditions that they were being kept in. Recently an investigation into the control of humidity had been undertaken which had identified that there were

some blockages in the system that meant the humidity had not been as controlled at it could be and that some of the sensors needed to be replaced.

6.3 Item 5.2: BSU Virtual Tour (May 2020 meeting)
Some starting points for the script/storyboard for the video of BSU had been drafted and were being reviewed.

7 3RS UPDATE

7.1 Proposed ARRIVE guidelines course
There had been discussions about a possible ARRIVE guidelines course. It would depend on the support that could be provided to RVC researchers in terms of understanding the ARRIVE guidelines. A review of recent papers was being carried out to get a feel of the extent of compliance with the guidelines and to identify where the problem areas seemed to be. The course would then be based on these areas. An update would be provided at the next meeting.

7.2 RSPCA Scientist-AWERB Engagement: review section on Dos and Don'ts for AWERB members:
https://www.rspca.org.uk/webContent/staticImages/Downloads/3DosAndDontsForAWERBMembers.pdf

Due to running out of time it was agreed that this item should be deferred to the next meeting.

7.3 New 3Rs self assessment tool
It was reported that there were new online self-assessment tools which had been designed for institutional assessment. The aim was to use them to collate, track and benchmark 3Rs activities. The tools should help researchers and also the institutions identify any local 3Rs opportunities. The system asks a series of questions about 3Rs, which were then scored, and gave feedback and suggestions. It should help identify whether there were specific areas that could look at to be proactive about the 3Rs. The tool would be promoted to researchers and feedback to see how well the tool helped in streamlining things.

7.4 Breeding and Colony Management Resource:
This was now published. A discussion group would be set up to discuss the tool and how it could best be used.

7.5 Improving the quality of research applications involving the use of animals workshop - 25 November @ RVC: feedback on how this went.
This had been well attended. There had been a good level of discussion, with breakout sessions to discuss and critique examples of applications. It had gone well so the event would be run again, but would probably be timed to take place before specific grant calls.

8 NVS REPORT

- **Ring Tailed lesions**: this was an ongoing concern
- **Ferrets**: one of the ferrets had a mass on its tail tip. This had been removed via surgery and the ferret was recovering well.
- **Anatomy ponies**: One of the ponies had apparently started to become "grumpy" during anatomy sessions. The pony had got tangled in a fence over the summer and had injured her legs. She had been examined: the lacerations had healed well but there was a chip of bone that was just "sitting there" as a foreign body that could be causing her some discomfort when handled. AWERB recommended that she be replaced so that she could be retired from the anatomy

demonstrations as she was not behaving well. Steps would then be taken to rehome her, potentially as a companion pony.

- **Dogs**: one of the bitches had come into season, so depending on how she interacted with the new stud dogs could either be bred from or artificial insemination carried out.
- **Sheep**: there had been some issues with some of the sheep transferred from the farm to Hawkshead BSU stock. They were not being used for any particular study but had developed eye irritation after transportation. The irritation had been treated and the sheep were being monitored. Discussions were being held with the farm though about the quality of animals being provided and where the eye infection could have been picked up from.
- **Cattle at the farm**: some of these were being used by a researcher under one of the project licences. The cattle had been checked for re-use after the procedures but three of them had to be subsequently euthanized due to health issues that were unrelated to the procedure. The researcher was wanting to add more cattle to the licence and had asked if he could add the majority of the herd. The more cattle that there were on the licence though, the more difficult it was to track each animal so increasing the risk of cattle accidentally being used for another licence or sent for slaughter with no official check. They had therefore settled for an additional 15 cattle to be added to the licence.

  The farm staff would be provided with a basic A(SP)A legislative reminder so they were aware of the requirements of the licence, in particular the requirement that if there were any health issues to cattle that were under a project licence, the NVS needed to be informed before anything was done so they could review the animals.

- **Transportation of laboratory animals from overseas**: Laboratory Animal Veterinary Association (LAVA) had recently advised of a number of issues relating to problems with transport of animals (predominantly rodents) over the past few months. Problems have primarily been delays due to cancellation of flights at short notice, re-routing and animals not being put onto the flight they were scheduled to take due to prioritisation of other cargo, especially where smaller planes are put onto the route. The situation did not appear to be improving and in fact was felt to be worsening. LAVA were therefore recommending that transport of live animals into and out of the UK was minimised wherever possible, since the risk of adverse events was higher than usual. Where transport was unavoidable, it was important to consider that delays may be likely and to provide food/ fluids for a prolonged journey (for example many journeys to the USA were now taking 6 to 7 days). They were also recommending that the full itinerary was made available to both sending and receiving establishments and that the couriers were asked to update both on any delays.

  Advice from the Home Office was that the carrier and Establishment Licence Holder must make sure to minimise the risk and establishments must make representation to Defra, as the Competent Authority for regulating the transport of animals, if any issues/criticality arose.

NACWO REPORTS

- **Mice colony**: It was reported that one of the researchers still had a mice colony although he had no funding to pay for them. He did not seem to be doing anything with the mice apart from breeding from them and had no plans to run the colony down. The Establishment Licence Holder would write to him saying keeping tick over colonies was not permissible

9 REVIEW AWERB TERMS OF REFERENCE TO SEE IF ANY CHANGES ARE NEEDED

It was agreed that AWERB would review these terms of reference outside of the meeting and to report back if they thought any changes were needed.

FOR INFO ITEMS

10 NEW PPLS GRANTED BY THE HOME OFFICE

AWERB noted that one project licence had been granted since the previous meeting.

11 PROJECT LICENCES AMENDED BY THE HOME OFFICE

AWERB noted that one project licence had been amended since the previous meeting.

12 END OF PROJECT LICENCE REPORTS

AWERB noted the end of project licence report that had been received. They would feedback if they had any comments on the reports.

13 CONDITION 18 REPORTS

AWERB noted the condition 18 report that had been submitted to the Home Office. No further information or action had been requested.

14 STUDY REQUESTS APPROVED SINCE THE LAST AWERB MEETING

AWERB noted the study request that had been approved since the previous AWERB meeting.

15 ANY OTHER BUSINESS:

15.1 **Date of next meeting:** this was arranged for 26 January 2021 at 2pm

Secretary
22 December 2020