MANCHESTER 1824
The University of Manchester

ANIMAL WELFARE AND ETHICAL REVIEW BODY

Minutes of the meeting held on 8 November 2018

Present: [redacted]

Apologies: [redacted]

1. **Minutes**

Agreed: That the minutes of the meeting held on 13th September 2018 were approved.

2. **Matters arising not covered elsewhere in the agenda**

Noted: a) [redacted]

b) The actions apportioned to the Secretary of AWERB from the meeting in September were deferred to the next meeting.

Reported: c) The Director of the BSF confirmed that he had updated the information he sends to licence holders to state that for those projects with a severe categorisation the licence holders will be required to attend an AWERB meeting 12 months before the end of the licence to discuss their progress including how they have minimised the number of animals they are using on the licence.

d) [redacted] confirmed she had spoken with [redacted] about including the Experimental Design Assistant (EDA) provided by the NC3Rs in the PIL course.

Approved: e) The Chair sought approval of the committee for the appointment of [redacted] from the School of Electrical & Electronic Engineering who has experience of animal work. An appointment period of 3 years was proposed from 1st January 2019 and approved.

3. Applications for New Project Licences

3.1. [redacted], 'Regulation miR-29 Targets in Wound Repair', ([redacted])

Considered: A completed AWERB form, PPL application, and minutes from the Local Management Committee Meeting

Interviewed: [redacted]

Discussed:

a) The Statistician confirmed that the statistics were fine.

b) The committee requested clarification regarding the choice of the sex of the mice. The applicant clarified that the project would use both male and female mice. As rates of healing differs for the sexes, their data must be kept separate for careful analysis.

c) The committee noted that details about the punch biopsy size varied. The applicant responded that as a refinement to the experiment the number of punch biopsies required had reduced from 4 to 2 with a very small increase in the size of the diameter from 5mm to less than 6mm. The committee agreed that this should be made clear in the licence and there should be consistency about the biopsy size.

d) The committee noted that the applicant was breeding 5 times the number of animals required. The committee discussed with the applicant the relative inefficiencies of breeding and noted this was unavoidable.

e) The committee noted there were some inconsistencies in the application regarding number of animals used which would need to be resolved.

f) The Panel also noted that she was limiting herself to 8-10 weeks of age to ensure that second anagen (the active growth phase of hair follicles) is not entered; wound healing cannot be analysed after this period as new hair follicles growing in the back of the mouse would interfere with the measurements.

g) As a general comment, the committee noted that it would be useful for the committee to have a copy any flowcharts prior to the meeting. This was to be fed back to [redacted] in the BSF.

Outcome: The study was given provisional approval based on the applicant making the changes/clarifications listed above to the satisfaction of the Chair/AWERB.

3.2. [redacted], 'Establishment & Healthy Maintenance of Blood & Vascular Systems', ([redacted])

Considered: A completed AWERB form, PPL application, and minutes from the Local Management Committee Meeting

Interviewed: [redacted]

Discussed:

a) The Statistician noted that the applicant needed enough animals to get successful grafts. It was necessary to know the percentage success rate to determine the numbers required. Although he had signed off the application, the Statistician would have liked to see more discussion in the 3Rs section about how the number of animals needed was determined. The applicant explained that she always repeats the experiment 3 times, so needs three mice for each experiment. Based on her experience, only 3-6 animals are required to provide strong significance. It was agreed the applicant should put these details in her reduction section.

b) The committee noted that some of the numbers in the application did not match and this would need to be rectified.

c) The committee noted in Protocols 3 and 4 the applicant had put "no for continued use" which was incorrect. This would need to be changed.

d) The committee noted that Protocols 1 and 2 were in the moderate band. These protocols involved 900 mice. The committee asked the applicant how many mice were likely to experience that severity and what they would experience. The applicant responded that for Protocol 1 there was very little procedure apart from genetic modification. The severity level for 70-80% of animals would be very low. The highest severity would be for mice who do not react well to immunisation. She estimated that genetic modification would impact about 30 mice.

e) For Protocol 2, a portion of the mice would have no blood cell generation and so will experience the subsequent effects which will be monitored closely. An estimate of 10-20% of those mice will need to be killed (about 60 mice). The worst suffering would be general poor health due to lack of blood cells.

f) The committee checked with the applicant how long the experiment would last in order to see the long term effects. She responded that the mice would receive the initial procedure after 4-6 weeks and would be kept for up to 20/24 weeks.

g) The Panel noted that in the Replacement section of the NTS, the applicant should refer to "in-vitro" experiments rather than "a dish or a test tube".

Outcome: The study was given provisional approval based on the applicant making the changes/clarifications listed above to the satisfaction of the Chair/AWERB.

3.3. [redacted], 'How Does Sinus Node Disease Maintain Atrial Fibrillation', ([redacted])

Considered: A completed AWERB form, PPL application, and minutes from the Local Management Committee Meeting

Interviewed: [redacted]

Discussed:

a) The committee required clarification as to whether the project would be using goats or sheep and why goats were the preferred option. The applicant clarified that their collaborator in [redacted] was using male goats, so using male goats at Manchester as well would provide the best comparison. The applicant confirmed that only one species would be used. The committee agreed that there would need to be an overwhelming argument for the use of goats.

b) The committee noted there was a lack of clarity as to whether 16 or 20 animals would be required. The applicants confirmed it was 16. This information should be consistent throughout all the documents.

c) The committee noted the term "put-down" should be replaced with "killed".

d) The committee was concerned that the project would be able to fulfil all its 5 hypotheses with the 16 animals requested. The applicant explained that the structural data will be descriptive. The applicants acknowledged that their first objective was primary and the others were exploratory/secondary. The committee agreed that clarity about the objectives and the descriptive nature of the data would need to be reflected in the application.

e) The committee asked the applicant about his publication strategy and whether he would make the raw data available. He responded that both positive and negative results would be important and publishable. He confirmed he would make the raw data available.

f) The committee noted that the NTS should be reworded to make it clear that the animal will be killed before pain or distress becomes severe.

g) The committee discussed the post- operative care of the goats with the applicant and how to limit the inquisitive behaviour without too much distress with the animals.

Outcome: The study was given provisional approval based on the applicant making the changes/clarifications listed above to the satisfaction of the Chair/AWERB.

3.4. [redacted], 'Defining Immunoregulation During Parasitic Helminth Infection', ([redacted])

Considered: A completed AWERB form, PPL application, and minutes from the Local Management Committee Meeting

Interviewed: [redacted]

Discussed:

a) The Statistician confirmed that he was happy with the statistics.

b) The committee noted that the NTS was too detailed and needed to be anonymised. [redacted] and [redacted] agreed to provide support to the applicant to get this right.

c) The committee noted that a large number of mice were being used. It explored with the applicant what the worst effects would be on the animals. She explained that the greatest risk was with the bone marrow chimera, with the worst effects being diarrhoea and weight loss with bone marrow. She estimated 1% of the mice would experience this level of suffering.

d) The committee explored with the applicant what percentage of animals would go through the most likely complex combination of procedures, as set out in annex 1 figure 5. She estimated it wold be about 15/20 mice.

e) The committee explored with her why mainly male mice would be used and whether this would affect translatability to women. The applicant explained that by using male mice she was keeping variation to a minimum. Female mice were more resistant anyway, so if it worked in male mice it would work more in females.

f) The committee requested the word flora be replaced.

g) The applicant clarified that raw data would be deposited in the Bioarchive and shared on request. Negative results would be presented at conferences.

Outcome: The study was given provisional approval based on the applicant making the changes/clarifications listed above to the satisfaction of the Chair/AWERB.

3.5. [redacted], 'Novel Targets for Anti-Epileptic Drug Design', ([redacted])

Considered: A completed AWERB form, PPL application, and minutes from the Local Management Committee Meeting

Interviewed: [redacted]

Discussed:

a) The Statistician confirmed he was happy with the statistics. The number of animals had reduced from 500 to 372.

The committee was concerned that the applicant was taking the experiment into the seizure model without knowing the PK value. The applicant explained that full toxicity testing had been conducted in flies. They concluded that if the compound had been toxic to the flies they would not use it in the mice. The applicant explained that laboratories around the world are using flies to test mammalian

toxicity and there was a paper about it. However the paper was not cited in the application and the committee agreed that the paper should be referenced in the licence. In addition it was unclear that flies would detect compounds that were toxic to mice that might be harmless in flies. Critically the committee felt it was inappropriate to be testing novel compounds in mice for which there was insufficient pharmacological data. The applicants could derive some of this by modelling and such studies should be included. However the committee felt that some fundamental PK data derived from mice was needed and noted that the applicants had earmarked 60 mice for pilot studies, which the committee felt could be used in this capacity.

b) The committee noted that there were many epilepsy models and questioned the transferability of the chosen model. The applicant acknowledged that there was no "true" model but looking at the literature, the PTZ model was the best to use. The committee also questioned whether an induced seizure would have the same effect as a non-induced seizure. The applicants explained that if they found a compound that reduced induced seizures in normal wild type mice they would progress to a trial in genetically modified mice.

c) The committee noted that the Protocol stated that repeat dosing may be required and wanted to know how that would be decided and calculated. The applicants explained they were using one dose per day for three days, based on pilot data.

d) The committee noted the application needed clarity with respect to the number of compounds to be tested.

e) The committee noted that as a non-schedule 1 procedure was being used justification would need to be provided.

f) The committee explored with the applicant why the seizure level had to be taken to Racine level 5. The applicant explained that this was necessary to see if the compound had an effect. The committee agreed more detail about this was needed in the licence.

Outcome: Unanimously the committee felt strongly that in its current form the application would be declined since it required significant changes. The committee agreed that the above changes should be made and the application resubmitted to the full committee.

3.6. [redacted], 'A Study on Treatment & Penetrance of Inherited Cardiac Conditions', ([redacted])

Considered: A completed AWERB form, PPL application, and minutes from the Local Management Committee Meeting

Interviewed: [redacted]

Discussed: a) The committee noted that the applicant had provided a very thorough and careful breakdown of the number of animals he would need. Although the committee considered this good practice, it

advised the applicant to build in some leeway for the inevitable changes of direction that experimental science would necessitate. Although it was acknowledged that he could submit an amendment if he required more animals.

b) The committee noted that the application required details of an alternative contact.

c) Protocols 2, 3, 4 and 5 stated that more than 20% weight loss for more than 72 hours. This needed rewriting.

d) The NTS needs rewriting with the assistance of [redacted] and [redacted] to improve the English.

e) It was agreed that the NVS, applicant and [redacted] would have a further look at the end points.

Outcome: The study has provisional approval based on the applicant making the changes/clarifications listed above to the satisfaction of the Chair/AWERB.

3.7. [redacted]
'Central Regulation of Appetite & Body Weight' ([redacted])
'Control of Arousal & Autonomic Output from the Brain' ([redacted])
'Anorexia & Cachexia in Ageing & Disease' ([redacted])

Considered: Completed AWERB forms, PPL applications, and minutes from Local Management Committee Meeting for the three applications listed above.

Interviewed: [redacted]

Discussed:

a) The committee noted that these three licences had previously been one very complex licence. There was work still to be done in the writing of the licences.

b) The Statistician had not fully reviewed all three applications but noted there was good practice where the applicant had built in cross-over designs.

c) The committee noted that as a non-schedule 1 procedure was being used justification would need to be provided.

d) The committee explored with the applicant if anaesthetic would be used for the inter-cranial interventions. The applicant explained that the cannula would be fitted under anaesthesia and left for a week so the mouse becomes habituated to the cannula before further interventions. The cannula does not affect behaviour.

e) The committee noted that some animals had to have their food restricted and so would have to be caged on their own. The applicant explained that for the first two licences (P9B0614DD and P4E1FAB01) 95% would have food restricted no more than overnight, just to make them hungry. For the last licence (P8E8F2390) they would be

purposefully making the animals lose weight but not become moribund. They would not need to go beyond 15-20% weight loss.

f) Between 30-40% of the animals would end up living on their own. Once male mice have been separated for a day or two, it is difficult to house them back together. The committee agreed that enrichment should be added where it would not affect the experiment.

g) For application P9B0614DD, the number of animals will be 40k in breeding. 15000 animals will be used for experimental purposes. It is the only licence with breeding so covers all three licences submitted by the applicant to this meeting. 1 in 4 animals will be used for experiments.

h) For application P8E8F2390, the applicant has 3 or 4 models of cancer following collaborators advice at [redacted] He is going to have to do his own studies initially to characterise the cancer model but will not be depriving those mice of food as well. The applicant will start with one cancer model as they know they can characterise that one.

i) The committee queried with the applicant that given they would be measuring food intake will the animals have to be housed on their own. The applicant explained that this would be the case during the characterisation of the model. This information should be added to the Refinement section of the NTS.

j) Given that there are 500-700 experiments the committee queried how many experiments would be included per paper. The applicant explained that a couple dozen experiments would be included per paper.

k) The NTSs need appropriate drafting with the help of [redacted] and [redacted].

Outcome: The studies have provisional approval based on the applicant making the changes/clarifications listed above to the satisfaction of the Chair/AWERB.

4. Report on amendments processed from 13/08/2018 to 18/10/2018

The following amendments were approved by the executive committee.

4.1. Amendments to Project Licences

[redacted], Neural Basis of Tactile Behaviour
[redacted], Circadian Control of Behaviour & Metabolism
[redacted], Understanding Cerebral Folate Metabolism
[redacted], Immunoregulation During Parasitic Helminth Infection

[redacted], The Long-Term Effects of Prenatal Hypoxia on Cardiomyocyte Function
[redacted], The Role of Inflammation in Cerebrovascular Disease
[redacted], Pregnancy Complications: Targeted Interventions
[redacted], Preclinical Evaluation of Cancer Therapeutics

4.2. Amendments to Project Licence [redacted]; Generation, Breeding and Maintenance of Genetically Altered Rodents

[redacted], Generating KOs for Chil3 & Chil4 Function
[redacted], To Produce an Hes1-mScarlet-I Mouse Model

4.3. Amendments to Project Licence [redacted], Generation & Breeding of Genetically Altered Rodents

[redacted], To Create a $KRAS^{LSL-G12C}$ Mouse Model Using CRISPR

4.4. Applications for secondary availability for new or current project licences

[redacted], Murine Models of Cancer Progression & Therapy, [redacted])

5. Any other business

5.1. Home Office statistics on breeding and genotyping of animals

The Home Office have released data the annual statistics on breeding and genotyping of animals for scientific procedures for 2017.

1.81 million non-genetically altered (non-GA) animals were bred for scientific procedures but were killed or died without being used in regulated procedures.

[redacted]

If there is time on the agenda for the next AWERB meeting [redacted] (NVS) will report on the exact figures as there may be some over recording of severity and number of animals.

The next meeting will be on 17th January 2019 at 10am-12pm, in [redacted].

Dates of meetings for the 2018/2019 academic year are:

14th March 2019, [redacted]

16th May 2019, [redacted]

4th July 2019, [redacted]
