MANCHESTER 1824
The University of Manchester

**ANIMAL WELFARE AND ETHICAL REVIEW BODY**

**Minutes of the meeting held on 4 March 2021 via Zoom**

Present: [redacted]

Apologies: [redacted]

Observer: [redacted]

## 1. Minutes

*Agreed:* That the minutes of the meeting held on 21 January 2021 were approved subject to a revision of a typographical error:

Item 4.1, Bullet point 3, line 3, Type of anaesthetic used will (instead of 'with') minimise this interaction

## 2. Applications for New Project Licences

### 2.1. [redacted], The Importance of Comorbidities in the Pathophysiology of Heart Failure

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

*Interviewed:* [redacted]

*Discussed:*

- [redacted] has worked with the applicant a lot on this licence and supports the number of animals being requested for use based on the calculations done to determine group sizes.

- The number of sheep to be used within the BSF was discussed including any welfare aspects of husbandry. The Acting Director of the BSF stated no concerns about catering for the number of sheep over the licence period and explained that some sheep go directly for Schedule 1 culling. The NVS stated that there is a square metre limit for housing animals and this is monitored closely.
- The applicant clarified that for the animals that receive only an injection, these are the animals that are going to be humanely killed via a Schedule 1 method, and that the injection increases the amount of cells that can be obtained after culling. The data available from these animals would not be available from other groups.
- The use of female sheep only was discussed. The applicant explained that the availability of male sheep is limited as these are castrated early and used for food. The disease does have a strong female prevalence so the use of only female sheep is in line with the human condition.
- When asked about the worst life experience of a sheep during the experiments the applicant outlined that some animals may suffer sudden death during surgical procedures. Given that the work requires the animals to get heart failure this cannot be avoided but at the first signs of heart failure the animals will be humanely killed.

*Revisions:*

- Please ensure that it is clear in the application that the heart failure with preserved ejection fraction is initially a model development protocol. This issue was raised in the meeting by the NVS.
- Please change the humane end point for sheep regarding weight loss. Rather than 20% body weight loss a body conditioning score should be used. Please make it clear what score would lead to the animal being humanely killed.
- Please include an average for adverse effects based on the discussion during the meeting and your previous experience of similar procedures on your current or previous licences. The committee discussed that adding all the percentages up gives a high percentage but understand after discussing that a sum of the numbers does not represent the true overall average of adverse effects observed in previous work.
- Page 31 - In the section "Lead or Device Failure" please check if the first sentence needs words adding.
- Page 112 - Under Protocol justification (b) the second word should be "are".
- Page 125 - Under Treatment toxicity please check if Pharmacodynamics should be Protocol 5 not 6.
- A number of comments were made regarding your Non-Technical Summary which are listed below. Please update your NTS based on the comments and send it to the following lay members for their review [redacted]
    - It might be helpful to mention that you are using gender balanced mice but only female sheep, with an explanation as per the main PPL.

- The "Why do you need to use animals to achieve the aim of your project?" could be edited down quite a bit
- In what will be done to the animals, it might be helpful to add that up to 4 devices could be inserted as one reviewer understands the work (pacemaker, intracardiac defibrillator, vagal nerve stimulator and telemetry).
- Also in 'what will be done' section, perhaps add something on the effect of Vagal nerve stimulation but that it will resolve after a few days, i.e. Coughing, dysphagia and dysphonia as coughing has 100% incidence.
- It might be helpful to say in the refinement section that animals are housed in social groups at all times apart from the immediate post-operative period recovery period.
- Please consider if the (very helpful) section on what the animals feel when they have heart failure and significant Ventricular Arrhythmias be added to the NTS.
- Writing as a 'lay' member the NTS overall is clear and does well to mostly avoid technical scientific terms. However, it is a little 'wordy' - a quick edit to reduce surplus words would make it more concise and easier to read. For instance, on page 2 of 200 one could remove "In this programme of work" so as to begin the sentence with "We will investigate " to create a more concise to the point sentence. However, one might also consider whether the section "Why is it important" requires detail of the "what" of the work together with hypothesis etc.; one could just explain why it is important (lack of effective treatments, worse survival rates over 5Y relative to breast/prostate cancer; and what we will learn/gain from the work (better knowledge of the mechanisms of disease and more effective treatments?).
- Is it possible to use an alternative to pathophysiology in sections that appear in the NTS or ensure that it is briefly explained?
- The description on high impact journals and open access is welcome but perhaps belongs maximise outputs; otherwise this info appears twice. This would produce a leaner on point answer to "what outputs do you think you will see at the end of this project?".
- Again, we may not need the hypothesis for the NTS only the intended output described concisely as "more effective therapies for heart disease".
- Page 7 of 200 - First paragraph under replacement "Diseases such as... new ways to treat them" is more of a justification for the importance of research programme which occurs earlier in NTS and is not required here. You could begin with second paragraph, starting with something like "We need to use whole living organisms because, whilst heart failure.... "
- Page 11 of 200 - for the question "Why can't you use animals that are less sentient?" might you wish to state the sheep is the best model because...? I realise this is covered in earlier questions but it feels like this answer needs to explain why a less sentient animal to a sheep could not be used... though I write as a lay person and am presuming for example a mouse would be less

sentient than a sheep though I imagine mouse experts may disagree about this.

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

**2.2. [redacted], Cellular Homeostasis & Brain Development**

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

*Interviewed:* [redacted]

*Discussed:*

- The number of animals requested for use on the licence is supported by the statistician with the caveat at there is not much information to base the power calculations on.
- The applicant explained to the committee the measures put in place for animals following the Morris Water Maze test. These include keeping the water at a warmish temperature, keeping the room warm, during the animals and placing them in a heated cabinet, and using tubing to slide the animals down into the water.
- The NVS asked for clarification on the time that the pups are away from the mother and if the use of anaesthesia causes rejection of the pups when they are replaced with their mothers. The applicant stated that 1 in 100 may be rejected.
- The applicant explained that a topical anaesthetic is used before the injection.
- The applicant was asked if the weight loss after tamoxifen is due to changes in metabolism or if the animals appear to not want to eat because they feel unwell. The applicant thinks the reduction in weight is metabolic. The NVS stated that the food may not taste nice for the animals.

*Revisions:*

- Page 6 - Point 1 - include option of giving tamoxifen via diet or gavage also
- Please include in the licence application that the injection is in the somatosensory cortex.
- Page 7 - "Organoid" is used without a description this first time. It could be usefully described here.
- Page 42 - Under Group size (1) there are figures given without any explanation. It should be stated what the numbers refer to.
- Page 59 - Similarly in Group Size (1) there are numbers without any indication of what they are.
- Protocol 1- step 2 – Please update 0.15mm to 1mm in the sentence “For removal of tip of tail (AA/AB), no more than 0.15 cm will be removed.”
- Protocol 2- Step 1 – the use of the term “chemical” needs to be replaced with “substance”. For example in “Administration of chemical to induce or modify gene expression”. All instances of “chemical” in the licence need changing.
- Protocol 2 – Step 2 (optional) – please discuss with the NVS the amount of times that the labelling agent will be administered. The

NVS does not feel that every 3 hour for 2 days is appropriate or possible.

- Protocol 2 – Step 4 – Please clarify the sentence "A maximum of 5 behaviour test types will be conducted per animals without repetition. The amount of time of an animal spent on performing these behaviour tests (cumulatively for all 5 tests) will not exceed 24 days". Do you mean a maximum of 5 tests per day for a maximum of 24 days in total?
- Protocol 2 – Step 4 – humane end points. Please include distress along with pain and discomfort. Please pick either 2 or 3 trials. The NVS suggests "withdraw from study if show adverse effects for 3 tests in a row".
- A number of comments were made regarding your Non-Technical Summary which are listed below. Please update your NTS based on the comments and send it to the following lay members for their review [redacted]
    - Page 3 of 64 - Why is it important…the first paragraph of "benefits" below (4.64) is a tighter and much clearer summary of what appears in this section. Please consider updating.
    - Page 3 of 64 - para 2, line 9 – please clarify if you mean changes over time or differences by comparison with non ASD.
    - Page 3 of 64 - para 3, line 1 – consider a brief explanation of neuronal & non-neuronal.
    - Page 4 of 64 - What outputs… para 1, line 3 "environmental insults" please explain technical use of "insults".
    - Page 4 of 64 - para 3, line 4 please explain briefly "transcriptome" datasets
    - Page 6 of 64 - This whole section gets very technical in language: it feels as if it has been lifted from elsewhere in the application rather than written for/edited for the NTS. It introduces abbreviated references (e.g. BrdU, or AA) with no explanations, and consider is the terms such as intraperitoneal, (& intracerebral, etc), & stereotaxic, appropriate for the NTS.
    - Page 7 of 64 -Why where they not suitable. Language gets technical again, introducing terms such as "cytoarchitecture".
    - Page 8 of 64 - How have you estimated… para 1, lines 1 & 2 Slips into power calculation language - "effect size" etc. Please remove these.
    - Page 10 of 64 - How will you .. para 1, lines 1 & 3, please briefly explain the terms "transgenic" and "full knockout" or use alternatives.
    - Page 9 of 64 - Refinement - Mention could be made of the culture of care of the animals for example; of avoidance of stress, the handling of the animals, and the temperature maintenance during the Morris water maze tests.
    - Please include if you are using male and/or female mice.
    - In terms of the Morris Water Maze, I do feel it needs to be made clear the length of time the animals are left in the water if they

do not find the platform. I understand they are guided to the platform after 60 seconds if they don't find it - which I assume is not too stressful, but it would be good to know more about how this test is carried out and briefly documented accordingly in the NTS. (For information, there has been a campaign by animal rights activists recently on the forced swim test so it's worth being clear to avoid misinformation and confusion). I assume there is no alternative to the mice being picked up by the tail?

- In terms of the rotarod test, perhaps explain this may cause transient stress. I assume there is no alternative to the mice being picked up by the tail?
- Page 2 of 64 - As a lay reader, I do not know what "pyramidal cells" are or what "microglia" are - could these and other technical terms be either explained briefly or substituted for non-technical language? [I realise that you define microglia on p.36 - I wonder if you could move this to the first time you use the term?]
- Page 3 of 64 - The answer to the importance question could be far more concise; for instance instead of asking the rather long question that starts paragraph 3 could you just state the importance of having an answer?
- Page 3 of 64 - The answer to the importance question could be far more concise; for instance instead of asking the rather long question that starts paragraph 3 could you just state the importance of having an answer?
- A general point on title - "Cellular homeostasis and brain development " does not provide a lot of indication as to the research and its relevance to autism spectrum disorder etc. I wonder if it may help to refresh the title so it conveys more specific meaning?

*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. Report on licences processed from 04/01/2021to 18/02/2021

The following amendments were approved by the executive committee.

### 3.1. Amendments to Project Licences

[redacted], How Does Sinus Node Disease Maintain Atrial Fibrillation.
[redacted], Studies of Cancer Inflammation & Immunity In Vivo (Primary at CRUK)
[redacted], Combination Immunotherapy for the Treatment of Cancer (Primary at CRUK).
[redacted], Fish Physiology in an Era of Climate Change.
[redacted], Brain Networks for Memory & Executive Function in Health & Disease
[redacted], Mechanisms Regulating Local & Distal Immune Responses In Barrier Site Health & Inflammation
[redacted], Identifying New Therapies to Prevent Internal Scarring.

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

[redacted], Generation of Chil4 KO - BALB/c Mouse Line Using CRISPR
[redacted], Generation of Prlhr-Cre Mouse Line Using CRISPR.
[redacted] Generation of Npff-Cre Mouse Line Using CRISPR.
[redacted], Generation of CLOCK-mRuby3 Mouse Line Using CRISPR

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

[redacted], Generation of B6-Tnfaip3$^{tm1Uman}$ / B6-Tnfaip3$^{tm2Uman}$ Mouse Lines Using CRISPR

**4. Update on applications outstanding from previous meetings and upcoming Project Licence applications**

4.1. As per the paperwork apart from [redacted] application which has just been granted.

4.2. The 15 April 2021 meeting which had been set aside for matters other than licence applications will now be used for the review of licences. An extra meeting is taking place on 12 April 2021 for discussion of operational processes, etc.

4.3. There are two issues involving [redacted] application which is pencilled in for the meeting on 27 May 2021.

Firstly, the application is still at a rudimentary stage. The Chair outlined again that there is a timeline for applicants with a number of deadlines for when meetings should be held. If this information has been provided to the applicant, or any applicant, and they do not stick to the deadlines then the committee are under no obligation to see them at the slot initially given to the applicant.

Secondly, the work does not have funding. The committee discussed if applications that do not have funding should be reviewed by the committee. AWERB members discussed that funded projects would generally have been peer reviewed whereas this would not be the case for unfunded work. There was a discussion regarding obligations to review a licence if the applicant was from industry. The NVS also raised the point that the Home Office would also take funding into account when reviewing the licence application.

For the application by [redacted] it was agreed that the Chair would contact them and let them know that AWERB would not be considering their application but would do so should funding be obtained.

**5. NACWO report**

5.1. A review of import/exports is taking place on a case by case basis. There is now one designated courier used.

5.2. An additional non-compliance not included in the paperwork circulated for the meeting was raised. The incident was a failure to give water to animals due to a new water pouch being incorrectly installed and a subsequent failure for the animals to be checked over the following days. The incident has been reported to the Home Office and Establishment Licence Holder. Standard Operating Procedures have been reviewed and

updated and staff have been retrained. Technicians must now ensure that they have sight of all animals in the cage whereas previously if an animal was in the nest they would not be disturbed. If the cage is moved the water pouch must also be checked. A meeting is taking place between the Home Office Inspectors and the animal technicians involved next week.

**6. Research Compliance Committee report**

6.1. No comments on the submitted report

**7. NVS report**

7.1. No additional comments than those outlined in the circulated report.

**8. Standard Conditions 18s**

8.1. The Home Office Inspector present stated that Standard Condition 18s are the responsibility of the Project Licence Holder and must be submitted to the Home Office within 72 hours.

**9. NC3Rs Regional Programme Manager update**

9.1. The self-assessment tool still requires volunteers to complete it. The NC3Rs RPM hopes to have the results for the extra AWERB meeting on 21 April 2021.

9.2. There is a new research culture hub on the NC3Rs website. This will be promoted soon with Personal Licence Holders.

9.3. The webinar series is continuing. Over 200 people attended the webinar which took place this week.

9.4. A 3Rs symposium will take place online in May. The NC3Rs RPM invites ideas for speakers or other agenda items.

9.5. The first meeting of the 3Rs subgroup will take place on 17 March 2021. The first meeting will discuss how they will operate.

**10. Any other business**

**10.1. Project Licence applicants attending a meeting as an observer**

The Chair suggested that it may be a good initiative to invite Project Licence applicants to attend a meeting as an observer prior to the meeting they will be presenting at.

The committee discussed that the issue of confidentiality would need to be outlined to the observer and applicant and that the applicant being observed should be asked if they agree to someone being present at the meeting that is not an AWERB member.

The Secretary will update the template email for inviting applicants to include information on an observer being present and the issues on confidentiality and the option to opt out of being observed by a future applicant. The updated invite will be reviewed and approved by the Chair.

The Secretary will invite future applicants to be observers at a meeting prior to when they will be presenting.

**The next meeting will be on 15 April 2021 at 10am-12pm,**
**via Zoom**

**Dates of meetings for the 2020/2021 academic year are:**

8 October 2020
Wednesday 18 November 2020
21 Jan 2021
4 March 2021
15 April 2021
27 May 2021
8 July 2021
19 August 2021
30 September 2021