National Centre for the Replacement Refinement & Reduction of Animals in Research

Researchers

Recommendations for refining rodent head fixation and fluid control

This summary presents the main recommendations from a review of the use of head fixation and fluid control in rodents. The recommendations are the product of an NC3Rs working group and you can find further material on this topic at the dedicated resource page.

This summary is intended for researchers already working in this area or soon to be using these techniques. Summaries for animal care staff and members of ethical review bodies are also available on the resource page.

The recommendations are split into three categories; head fixation surgery, dietary control, and behavioural set-up. Links to sections of the paper and to other useful material are given where relevant for further information.

Head fixation surgery – preparation and conversation are key

1. Pre-surgical steps are key to a successful outcome. Ensure animals are healthy, that there is plenty of time for them to be habituated to the experimenter and facility, and preparations for post-operative recovery have been taken The latter includes preparing recovery chambers and consulting with animal care staff about monitoring, including frequency and intervention points.

2. If animals are already under caloric control, return them to *ad libitum* access and allow them to regain weight appropriate to their age and pre-restriction weight before surgery.

3. Good aseptic technique should always be observed. Use of a trained assistant can help minimise the chances of the surgeon breaking asepsis and should be standard practice. Further advice on this is available, from the Laboratory Animal Science Association and through Research Animal Training.

4. A combined anaesthetic and analgesic regimen should be followed. Seek local veterinary advice on a regimen that includes appropriate pre- and post-operative analgesia and review this with them regularly to ensure best practice is always followed.

5. Deliver fluids before surgery, as well as after for longer surgeries, to prevent dehydration. Avoid delivering fluids during surgery as this may prolong surgery further and not be necessary in addition to pre- and post-surgical fluids.

6. The site of surgery and general health of the animal should be monitored closely in the days following the procedure and observations recorded and reviewed so that appropriate action can be promptly taken. For example, see the example health monitoring templates for key indicators of recovery, including body weight, integrity of the wound site and general activity level of the animal.

7. Group housing of implanted animals is strongly recommended and should be the standard practice to avoid the negative welfare impacts of single housing. Group housing has not been observed to lead to greater post-surgical complications or implant loss.

Dietary control – maintaining motivation and animal health

8. First, consider whether dietary control is needed at all; is any restriction needed to motivate the desired behaviour? Could a highly palatable reward alone achieve the desired level of performance? What else could be done to reduce the need or level of restriction required? Begin from a position

that restriction should only be considered if the alternatives would not provide the desired level of motivation.

9. Where its use is unavoidable, the degree of restriction should be regularly reviewed, adjusted and minimised throughout the study to maintain motivation without unduly impacting on the animal's welfare, for example, easing restriction once initial training is complete. This will require a flexible approach that is individual to each animal.

10. Allow for growth in younger animals, consider "holiday" breaks from restriction, top-ups of food and water outside of testing and periods of recovery of body weight before and after surgery.

11. Consider what else can be done to minimise the degree of restriction, for example habituation to any restraint needed and other techniques to be used to reduce levels of aversion. Equally, if behavioural performance is poor, first review possible technical failures or signs of ill-health in the animal before restricting access to food or water further.

12. The overall welfare of each animal must be monitored and recorded daily and compared against clearly defined intervention points. Discuss these measures and appropriate responses with your animal care staff as well as local ethical review body. For example, see the example health monitoring templates for key welfare measures, including body weight, grooming and general activity level of the animal.

13. Separating an animal for a short period to feed can address situations in which an individual rodent continues to lose weight while others in the cage remain stable, but the time apart should be minimised. Consult animal care staff when carrying this out.

14. Responses to dietary control may differ in mutant rodent lines compared to their wildtype counterparts, so be particularly cautious when first using it in a novel line and ensure that animal care staff are well briefed if additional concerns are anticipated.

Behavioural set-up – improving welfare for better performance

15. First, consider whether head fixation is necessary or if your scientific goals can achieved with less restraint. Check for advances in, , tethered or wireless recording techniques for example.

16. Habituation to restraint should be practiced before formal testing as this will reduce stress responses to head fixation, improving task engagement and decreasing the likelihood of the loss of headcaps.

17. Further steps to reduce stress throughout the task should also be taken, for example allowing for naturalistic behaviours as part of the response and locomotion during testing or using self-initiated head fixation.

18. Self-initiation of trials should be used where large numbers of omissions and/or high response latencies may confound the results.

19. Monitoring factors such as pupil size and facial expressions, even when unrelated to the task, provides useful indicators of welfare and engagement. Consider also other measures of welfare that could be incorporated into the study design, for example noting overt signs of distress, the presence of faeces following restrain and using task-specific measures of engagement as an indicator of habitation to the testing set-up. Review these welfare measures and take appropriate action promptly.