

Providing animal technicians with the latest news from the NC3Rs

# Tech3Rs

## Welcome to the latest edition of Tech3Rs

This newsletter is for animal technicians working in research establishments to help identify opportunities to embed the 3Rs in practice and ensure high standards of animal welfare.

If you have any ideas for future issues or are working on a 3Rs approach you would like us to feature, please get in touch – we would love to hear from you! You can email us at [tech3rs@nc3rs.org.uk](mailto:tech3rs@nc3rs.org.uk).

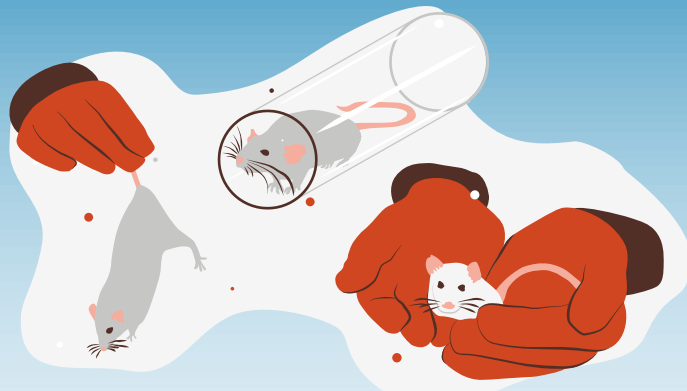
In this issue we share details of our newly launched e-learning course on refined mouse handling and highlight two Tech3Rs champions who share their work on promoting animal welfare through improvements in experimental procedures. We also summarise two research papers describing refinements for large animal species.

## Don't miss the next issue!



Tech3Rs is published online: [www.nc3rs.org.uk/tech3rs](http://www.nc3rs.org.uk/tech3rs)

If you would like to receive an email when the next issue of Tech3Rs is published, please email us at [tech3rs@nc3rs.org.uk](mailto:tech3rs@nc3rs.org.uk) with the subject line "Subscribe".



## Refined mouse handling e-learning course

### A new interactive course on non-aversive handling is now available to complete

We are excited to launch the refined mouse handling e-learning course, created by the NC3Rs and the US-based 3Rs Collaborative (3RsC) in an international collaboration with partners from academia and industry. Through a series of interactive lessons the course is a comprehensive and accessible overview of the evidence base, benefits and practicalities of refined handling, for the mice, the handler and the research.

Whether you are already using refined handling methods, preparing to make the switch or interested in learning more about tunnel and cup handling, the e-learning resource will help you feel confident in picking up mice using refined methods. It offers practical advice on implementing refined handling in your facility addressing biosecurity issues, efficiency and how tunnel and cup handling can be used in combination with restraint and procedures.

The e-learning course serves as an introduction for those new to refined handling or a refresher if you already have experience in these methods.

With demonstration videos and quizzes to test your knowledge, the course supports efficient and effective hands-on training. Completing the e-learning course ahead of hands-on refined handling training will give you a solid foundation in the background and theory ready to put into practice, to help you get the most out of your practical training session.

The course is designed for anyone who works with laboratory mice at all levels of experience. It should take no longer than 45 minutes in total and you can pause between lessons to fit the training into your working day.

**Access the e-learning course and other refined mouse handling resources:** [nc3rs.org.uk/3rs-resources/mouse-handling](http://nc3rs.org.uk/3rs-resources/mouse-handling)

**For questions about the e-learning course or refined handling in general, contact:** Dr Khia Dobbinson [enquiries@nc3rs.org.uk](mailto:enquiries@nc3rs.org.uk) or 3RsC Dr Megan LaFollette [info@na3rsc.org](mailto:info@na3rsc.org)

# Tech3Rs champions

In this issue we feature two refinements of experimental procedures which have improved animal welfare, user performance and success rate of the techniques.

**Chris Moore is a senior laboratory research scientist at the Francis Crick Institute. He spoke to us about improving the safety of intratracheal dosing in mice.**

## What 3Rs idea have you implemented?

Our research group studies mouse models of lung cancer, which requires us to accurately and consistently administer liquid adenoviruses or retroviruses directly into the lungs of mice, via the trachea (also known as the 'windpipe'). To avoid the growth of tumours in unintended places outside the lungs, such as the throat or nose, it is essential that these substances only make contact with lung tissue. To achieve this, we originally used the method of intratracheal intubation outlined in the DuPage *et al.* 2009 paper on [conditional mouse models of lung cancer](#). This involves using an intravenous catheter needle to place a flexible plastic cannula into the trachea of anaesthetised mice. The needle provides rigidity to the cannula allowing it to be guided down the narrow trachea of the mouse. The needle is then removed leaving the cannula in place, ready for substance administration directly to the lungs. I have refined this approach by adding a small 3D printed 'collar' to the intubation needle, greatly reducing the risk of injury to the mice and improving user confidence and competence.

## How did you develop this approach?

When I was asked to train new users in the intratracheal intubation technique I found that they repeatedly had difficulty holding the intubation needle correctly, resulting in injury to the trachea of the mice. When this would happen, the mice would have to be euthanised while still under anaesthesia. Losing animals is never taken lightly and staff would often be very upset when these incidents occurred. The reason such injuries would occur is because the intravenous catheter needle is longer than the plastic cannula (image B, iv), meaning that the person performing the procedure had to hold the needle in a way that its

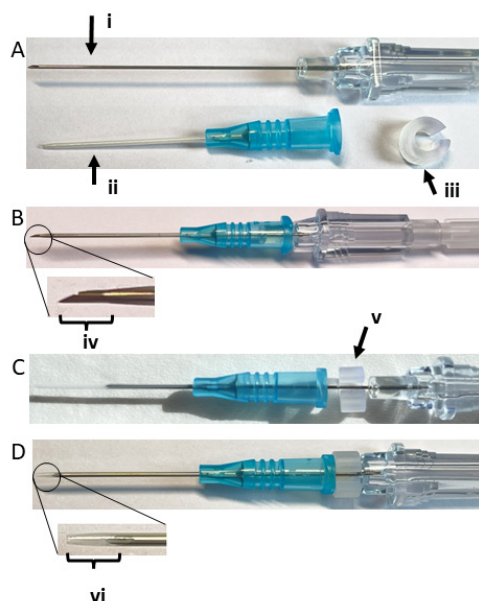
sharp end, the bevel, would not go past the plastic cannula and injure the trachea. Holding the needle and cannula in a way that minimises the risk of injury was uncomfortable and required a lot of practice to get consistently correct.

I thought about how we could get around these issues to reduce the risk to the mice by minimising the more technically challenging elements of the procedure and I came up with a prototype 'collar' device. I took the prototype to our mechanical engineering department and they were able to 3D print my new intubation collars. The collar is a small plastic ring (image A, iii) that easily slots on and off the needle without the user needing to put their fingers near the sharp end. It is also a re-usable attachment, so it is highly cost-effective, as long as you remember to remove it before throwing the needle away! This device prevents the needle bevel from becoming exposed (image D, vi), reducing the risk of harm to the mice, while maintaining the rigidity required to accurately place the cannula. The intubation collar also allows the needle to be held in a more comfortable and natural position so that users can fully focus on the mouse.

## What was the outcome of this process?

I have reduced the time taken to train users to a competent level from three to five days to only two hours. Trainees are much more confident now that the risk of injuring the mice is greatly minimised. Previously, during training, incidents of euthanasia following tracheal injury could be up to 10%. By adding a collar to the needle this rate has dropped to zero.

I have trained around 25 people in using this approach, including people from other UK facilities and overseas. No one has reported difficulty in taking the technique back to their own lab, which is a sign that the skill is transferring well. I am always happy to share our SOP, the 3D printing schematic and any additional expertise with anyone who may find these helpful.



### Intubation needle and collar

(A) Breakdown of the intubation needle; (i) inner bevel metal needle; (ii) flexible soft plastic cannula; (iii) intubation collar.

(B) When cannula is inserted over the needle, the bevel of the needle moves past the cannula tubing (iv). This will lead to injury if inserted into the trachea as pictured.

(C) Attachment of the collar to the needle, by passing the collar through the side of the needle (v).

(D) With the collar attached, the bevel of the needle is no longer exposed and the needle still adds rigidity to the flexible cannula tubing (vi).

**Sheena Wallington is a senior licensed animal technologist and former nonclinical manager at Evox Therapeutics. She spoke to us about improving blood sampling from the saphenous vein in mice.**

### **What 3Rs idea have you developed?**

Some years ago, I saw a poster at the IAT Congress on the welfare benefits of collecting blood from the saphenous vein (less tissue damage and less scarring) which made me want to adopt this technique and switch from tail vein to saphenous vein bleeding. The saphenous vein is more superficial and more easily accessible, so in theory it is a faster and more reliable way of blood sampling. Most bleeds can be obtained with one needle stick as opposed to tail vein, where the supply quickly drops as the blood clots and thus multiple sticks are often needed until sufficient volume is obtained.

When I started using this technique, I realised that it can be difficult to see the saphenous vein, especially as the mouse skin often shows varied levels of pigmentation in that area. Another potential barrier to fast and easy bleeding is that the saphenous vein, unlike the tail vein, travels slightly different paths in each animal, so you really need to be able to see the veins well to be able to collect blood reliably. Due to these aspects of mouse biology, locating the saphenous vein can sometimes take time. However, as saphenous bleeding requires restraint, which the mice can find stressful, it is important to keep the duration of the procedure to the minimum.

I have addressed these challenges by using a small bullet LED light to help visualise the saphenous vein, making it much easier to insert the needle in the right place.

### **How did you develop this project?**

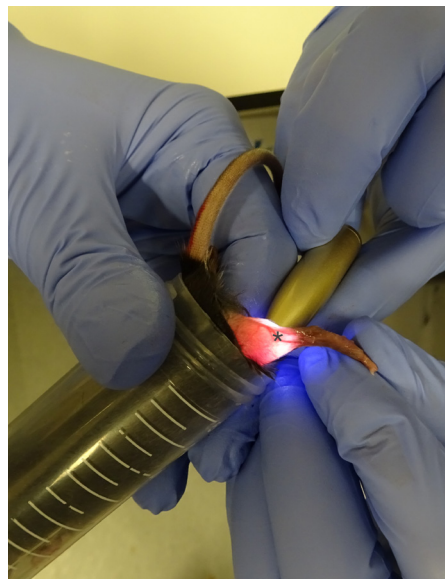
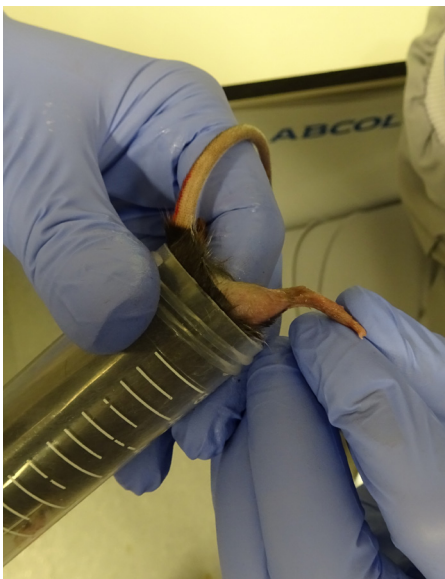
My inspiration for this approach resulted from a happy accident in my home life that gave me a 'lightbulb' moment. While I was fiddling with an LED light, I noticed that the light was strong enough that it shone through my hand, allowing me to see the veins in my finger. This gave me the idea to use an LED light to improve visualisation of the saphenous vein in our mice. To achieve this the mouse is picked up using a refined method [1] and restrained using a restraint tube. The hind limb is immobilised in the extended position and the light is held against the underside of the animal's leg allowing the vein to be located quickly and easily. Once you have located the vein you can proceed to accurately take a blood sample.

If you are using a clamp to hold the light in place, care should be taken to only switch the light on once the mouse's leg is placed over the LED to avoid shining light straight into your eyes. Alternatively, a second person can hold the light in place and point it to the right direction, so it doesn't shine into the eyes of the person taking the blood sample.

### **What was the outcome of the project?**

My method not only has allowed me to reliably and consistently perform saphenous bleeding in mice, but it has also increased the speed in which blood can be collected thus reducing the stress of restraint to the animal. Being able to locate the saphenous vein reliably means that bleeding requires a single needle stick, so unnecessary suffering due to multiple stabs is minimised. I have found that the success rate of saphenous bleeding, measured by the number of samples that the target volume was achieved with a single needle stick, has more than doubled simply by using an LED light. The training required to learn this approach is very short, sometimes it only takes a single session. However, I would make sure people had several sessions of supervision, before being signed off, to ensure full competency of the technique. Once someone masters this technique, saphenous bleeding using the light almost never fails. Blood quality has vastly improved, too; blood collected from the saphenous vein is rarely haemolysed and the plasma samples are clean and consistent. These improvements mean that the number of mice required to collect sufficient and good quality blood samples can be kept to the minimum.

This procedure has been signed off by the training oversight staff of the facility and I have been invited to train colleagues in other facilities who found my method really useful for improving the quality of the blood samples and reducing the time it takes to bleed a mouse, while improving animal welfare.



**1. For details on the refined handling methods used by Sheena, visit our resource on mouse handling: [nc3rs.org.uk/3rs-resources/mouse-handling](https://nc3rs.org.uk/3rs-resources/mouse-handling).**

Left image: View of the mouse leg without light.

Right image: When an LED light is held against the underside of the mouse's leg the saphenous vein (indicated with \*) is clearly visible.



# 3Rs papers of interest

Each issue we feature recent 3Rs papers, providing summaries and links to the full articles for further reading. This issue we focus on refinements for large animal species.



## Clean-catching urine from pigs: A method for collecting quality specimens for urinalysis and microbiological culturing in a laboratory environment

- Urine collected for analysis needs to be clean and free of contaminating microbes. Standard methods of sample collection from pigs include bladder catheterisation or suprapubic bladder aspiration, two invasive procedures that require general anaesthesia.
- In this study, the authors aimed to validate a novel method of non-invasive urine collection from pigs.
- The study collected urine from 41 female pigs (Landrace/Yorkshire, cross-breed), which were housed in groups of up to ten individuals. Mid-stream urine was collected non-invasively using a long-handled pick-up tool (similar to what is used for litter picking) with a 100ml sterile cup attached at the end.
- To avoid time-consuming waits for spontaneous urination, the authors also developed a protocol to encourage urination.
- This was based on the observation that when the pigs were allowed back into their enclosure after it had been cleaned, or after they had been temporarily moved elsewhere in the facility to undergo training or socialisation, they would often immediately go to the designated dunging corners to urinate.
- While in the dunging corner and before urination began, the pigs would express their rooting behaviour, alerting the bystander that they were about to urinate. This observation further facilitated clean-catch collection of urine.
- When compared to urine collected by the standard methods, the authors found minimal contamination of microbes from skin and vaginal surfaces. This refinement improves animal welfare, without compromising data quality.

## Postoperative severity assessment in sheep

- Sheep are commonly used in orthopaedic studies which often involve surgical interventions, such as tendon ablation, making them painful for the animals. Effective pain management is crucial for animal welfare and data quality. However, adequate severity assessment methods for pain in large animal models are often limited.
- The authors of this study used a combined approach of telemetry and the Sheep Grimace Scale (SGS) to assess pain severity in four German black-headed mutton sheep after two surgical interventions, telemetry transmitter implantation and tendon ablation.
- Following tendon ablation, the authors monitored activity and heart rate using telemetry devices, along with SGS analysis from video recordings and classic clinical scoring. Clinical scoring included vocalisation, food intake and general appearance.
- SGS and telemetry data can be effectively used as indicators of pain, unlike clinical scoring. These methods may be useful refinements for pain detection in large animals.

**Stærk K et al. (2023). Clean-catching urine from pigs: A method for collecting quality specimens for urinalysis and microbiological culturing in a laboratory environment. *Laboratory Animals* 57(3),293-303. doi: [10.1177/00236772221133433](https://doi.org/10.1177/00236772221133433)**



**Zentrich E et al. (2023). Postoperative Severity Assessment in Sheep. *European Surgical Research* 64(1), 27-36. doi: [10.1159/000526058](https://doi.org/10.1159/000526058)**



# Updates from the NC3Rs

## Replacing sentinel animals with environmental health monitoring

We have created a new project page on using environmental sampling methods to reduce the number of animals used for colony health surveillance.

Monitoring rodent research colonies for pathogens and parasites is important for animal welfare and conducting high quality science. Traditionally, sentinel animals are maintained within the colony and culled for health surveillance purposes. This may involve buying animals specifically for this purpose, but accurate and cost-effective colony health surveillance is possible without sentinels.

Technological advances mean that molecular analysis of environmental samples (e.g. dust debris and soiled bedding) can replace the use of sentinel animals. We spoke to Chago Bowers (animal technician, University of Chicago) and Dr Lynlee Stevey-Rindenow (veterinarian, University of Southern California) about how they substantially reduced the number of mice and rats culled at their facilities for health monitoring purposes. Chago and Lynlee also shared their perspectives on the time, cost and emotional benefits of replacing sentinel animals.

Visit [nc3rs.org.uk/our-portfolio/replacing-sentinel-animals-environmental-health-monitoring](https://nc3rs.org.uk/our-portfolio/replacing-sentinel-animals-environmental-health-monitoring) to read a past Tech3Rs interview with Chago and listen to a brand new audio interview with Lynlee.

For more information on environmental health monitoring, including how to switch, visit the 3Rs Collaborative's resource: [na3rsc.org/health-monitoring](https://na3rsc.org/health-monitoring)

Have you replaced sentinel animals with environmental health monitoring? If so, we would love to hear from you: [tech3Rs@nc3rs.org.uk](mailto:tech3Rs@nc3rs.org.uk)

## Refining rodent head fixation and fluid control

We convened an expert working group to identify refinements to head fixation and fluid control in neuroscience research using mice. Current practices and recommendations to improve animal wellbeing and data quality, based on a survey of the community, literature reviews, and the expert opinion and practical experience of the working group were published in 2022 (Barkus *et al.* 2022).

We have now created a summary sheet specifically for animal technicians with the main recommendations from this report, which is available to download. The summary sheet also provides links to the relevant sections of the paper as well as other useful resources.

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**Download the summary sheet and find further information about the project: [nc3rs.org.uk/3rs-resources/refining-use-head-fixation-and-fluid-control-rodents](https://nc3rs.org.uk/3rs-resources/refining-use-head-fixation-and-fluid-control-rodents)**

## Are you a UK-based Named Information Officer? We want to hear from you!

In collaboration with LASA's Home Office Liaison, Training and Information Forum we are running a survey on the role of the Named Information Officer (NIO).

The survey aims to establish a national picture on how the NIO operates in practice, better understand how the role is supported and identify opportunities to further support those in this important role.

If you are currently in an NIO role working in a UK research establishment (academic, industry, CRO or government funded) please take a few minutes to share your thoughts.

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**Survey closes Monday 18 December 2023: [nc3rs.org.uk/our-portfolio/supporting-named-information-officer-role](https://nc3rs.org.uk/our-portfolio/supporting-named-information-officer-role)**

## External event: 'Technicians' welfare workshop

15 January 2024

The Biological Services Facility at the University of Manchester is hosting a free online event to discuss how wellbeing can be improved within the industry and learn about the latest advancements in technician wellbeing.

Speakers include Penny Hawkins (RSPCA), Haley Daniels (University of York) and Jonathon Wood (Datesand). There will also be an opportunity to ask questions.

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**Registration: [www.eventbrite.co.uk/e/technician-wellbeing-workshop-tickets-746961220117](https://www.eventbrite.co.uk/e/technician-wellbeing-workshop-tickets-746961220117)**

# Celebrating our 20-year anniversary

We are excited to be celebrating our 20-year anniversary throughout 2024. Further details on how we will be marking this occasion will be released shortly on our website, but we would love for animal technicians to get involved in our activities.

## Feature in our anniversary edition of Tech3Rs

What are your success stories over the last 20 years? Have you led on, or been part of a 3Rs project that has improved the welfare of animals?

Have you changed practices or implemented a refinement after reading Tech3Rs?

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We would love to hear from you, get in touch: [tech3rs@nc3rs.org.uk](mailto:tech3rs@nc3rs.org.uk)

## Apply for a Public Engagement award

If your work advances the 3Rs, regardless of your role you can apply for £2,000 to support public engagement activity in 2024.

Ideas include online engagement activities (e.g. videos and webinars), workshops or visits to other establishments to share your 3Rs ideas.

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Next deadline, 22 January 2024: [nc3rs.org.uk/our-funding-schemes/public-engagement-awards-20-year-anniversary](https://nc3rs.org.uk/our-funding-schemes/public-engagement-awards-20-year-anniversary)

## Call for 3Rs images

Do you have any images of your work you are willing to share with us to help us bring our work to life (credited or anonymous)?

We also have a small budget available next year to commission images for particularly relevant or interesting 3Rs approaches.

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Get in touch to discuss sharing your images or commissioning a photographer: [media@nc3rs.org.uk](mailto:media@nc3rs.org.uk)

## Other news

### Do you administer tamoxifen to your mice? Your input is needed!

Current administration methods for tamoxifen include oral gavage or peritoneal injection, which are invasive, or using tamoxifen-diets, which can cause feed aversion due to the bitter taste of tamoxifen.

Fluid Pharma Ltd have developed a tasteless tamoxifen formulation to improve administration to rodents with the support of NC3Rs CRACK IT Challenge funding. Before making the product fully available, Fluid Pharma Ltd. are undertaking some research to learn more about the size and needs of the market.

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If you are using tamoxifen and you are willing to help, please follow the link to the questionnaire: [bit.ly/3N5CjEN](https://bit.ly/3N5CjEN)

You can find out more about this product on the NC3Rs website: [nc3rs.org.uk/crackit/tat-fit?tab=products](https://nc3rs.org.uk/crackit/tat-fit?tab=products)

### Do you care for laboratory frogs, or are you setting up a *Xenopus* facility?

Wherever you are in the world you can join a community of animal technicians and *Xenopus* scientists sharing questions and advice on frog health, handling and husbandry.

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Email [tech3rs@nc3rs.org.uk](mailto:tech3rs@nc3rs.org.uk) to be connected with the organiser and added to the *Xenopus* virtual mailing list.

The NC3Rs team would like to wish you a restful festive season. Please note our office will be closed from Thursday 21 December and will reopen on Tuesday 2 January 2024.