

ANDREW BLAKE TRIBUTE AWARD

Validating the use of box training as a refinement to rabbit handling

ALICIA KINALLY, LUCY ONIONS and SARAH GLENN

University of Leicester

Correspondence: ak716@leicester.ac.uk

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Introduction

In the wild, rabbits are prey animals and so the act of being picked up and handled can be extremely stressful for them. Previous studies in pet rabbits have shown that during the act of lifting, rabbits show signs of struggling and aggression due to fear; consequently, pet owners are recommended to avoid directly lifting animals where possible.¹ The most commonly used method of handling laboratory rabbits is to grasp the animal by the scruff, while placing your other hand to support the rear, lifting the animal free of the cage/pen. The animal is then held against the body, with the bodyweight being supported on the arm.² We wanted to introduce a new method of lifting and carrying the rabbit that would reduce stress, similar to the already recognised and widely used method of tunnel handling mice.³

In our facility the rabbits are handled multiple times a week, due to them spending time in specifically designed playpens for weekly exercise or undergoing weekly weighing and health checks. The rabbits we house are used for cardiovascular models of Myocardial Infarction and so it is important they are not experiencing a lot of stress to ensure validity in data produced. We believe less invasive handling will also affect the overall temperament of the rabbits and subsequently they will be more likely to positively respond to human interaction rather than be avoidant as they have previously been when being scruffed regularly.

Our rabbit models

Housing and acclimatisation



Figure 1. Standard rabbit housing at the University of Leicester.

Once our intact male New Zealand white rabbits (all 2.25-2.5kg on arrival) came into the unit from Envigo, they were singularly housed in an Allentown double rabbit rack (see figure 1). One side of the rack trays were lined with a paper tray liner and filled with a thick layer of aspen chips to allow natural behaviour such as digging. The rabbits also received boxes, tubes, chew blocks and play balls as environmental enrichment. They were acclimatised here for one week; The rabbit holding room is on a 12/12-hour light cycle with lights gradually fading to off at 7pm and gradually turning back on at 7am. They are housed at an average temperature of approximately 19°C which is monitored daily and they are fed the Teklad™ Global Rabbit Diet 2030 in their hopper. Their housing always had a water bottle on both sides of their cage due to the rabbits having access to both sides of their accommodation.

During this time, no handling took place and they were allocated into either of two groups – the first group were to be trained with a carrying box containing a shared piece of Vetbed and the second group were given their own Vetbed. Because the size of the batches of rabbits arriving at one time was so small, the first batch was allocated randomly to either of the two groups. A month later when the next group arrived, they were allocated to the remaining group. We wanted to use rabbits that were going to be staying with us for an extended period of time and so we chose to use our MI/SHAM rabbits as they would be with us for around 10 weeks (2 weeks before and 8 weeks post-surgery). This gave us plenty of time to collect data for the training and allowed us to explore whether the surgery affected the training in any way. The control rabbits we house typically are only with us for two weeks and so whilst it is possible to box train them in that time, we would not have been able to collect much data from them.

As the rabbits were already here and being used for surgery and the handling study was not an intrusive project, we did not require any ethical approval to undertake it.

Incentives

Whilst trialling the carrying box training, we wanted to see if there was something that would be a good incentive for the rabbits that would encourage them into the box quite quickly. We had previously tried treats and clicker training to help encourage them into the boxes but our rabbits did not seem very interested.

We took the idea of the Vetbed from the fact that in their playpen already, the carrying box was being used as a 'hiding house' and it contained a piece of Vetbed that everyone shared whilst in there (see figure 2) so with this, we created 2 groups.

Shared Vetbed rabbits (2 rabbits) – A piece of Vetbed was placed in the carrying box that was not changed



Figure 2. Rabbit carrying box with Vetbed *in situ*.

or washed in between every rabbit and therefore picked up all of the rabbits' scent (washed only when very dirty, typically once a week). This was the same as the routine used in the playpen – the rabbits are used to smelling each other only in the playpen and otherwise do not have access to other rabbits' scent. The thought process was that the shared Vetbed may smell interesting to the rabbit due to everyone else's scent being on there and therefore he may want to jump in and investigate the scent.

Own Vetbed rabbits (3 rabbits) – A piece of Vetbed that was only for the individual rabbit was provided for these rabbits. It was not shared among the group. The Vetbed would only smell like the rabbit who it was assigned to and was only washed if really dirty. The idea behind this was that the rabbit might have felt more comfortable to jump into the box if the box smelled of his own scent.

Introduction of the box

After a week the rabbits arrived into the unit (usually the following Monday) the carrier box was introduced to the rabbits at the front of the rabbit cage where we were to hold it steadily at the opening waiting for the rabbit to fully jump in (see figure 3). They were timed for 3 minutes using a timer and after 3 minutes, if they had not fully jumped into the box (all 4 legs and body) they were scooped up and were carried to the playpen by hand. If they did jump into the box, the entrance to the box was covered using our arms to ensure the rabbit could not fall or jump out and the rabbit was carried to either the playpen or scales where the box was placed down and the rabbit could jump out on his own (see figures 4 and 5).



Figure 3.

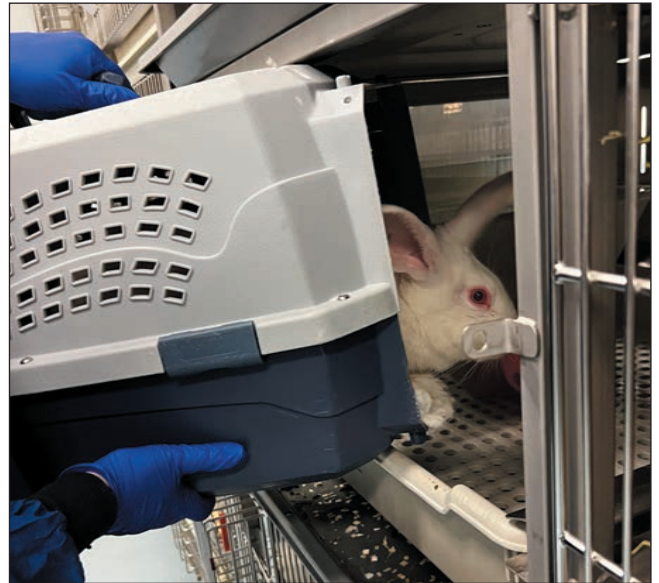


Figure 6.



Figure 4.



Figure 5.

The timings were recorded for each rabbit on a spreadsheet so we could monitor how long the rabbits took to jump in.

After the rabbits had their allocated time in the playpens or had been weighed on the scales, they were gently encouraged back into the box by lightly guiding them in from behind. They were then taken back to their cage the same way as before, with the entrance covered. Once they arrived back at their cage, the box was placed back in the same position in front of the cage as before and the rabbits were able to hop back into the cage in their own time (see figure 6). They were not forced out of the box and most rabbits jumped back into their cage straight away.

Positive association

When training the rabbits, we wanted to ensure they associated the box as being a positive activity that they would voluntarily do. This again would reduce stress. The way in which we did this was by making sure not to rush the rabbits into the box; whilst waiting for them to jump in, they were not pushed, touched or made to feel like they had to jump in. The same goes for jumping out at their end destination; the rabbits are not forced out of the box or picked up out of the box as it could cause the rabbit to form a negative association with the box and effect the end result of becoming a box trained rabbit.

We also wanted the box training to be repeatable and a positive experience with whoever was in the rabbit room at the time, due to there being different people actively working in the rabbit room during staff annual leave, training, post op care and weekend cover. Each person who was going to be handling rabbits in the room at various time points regularly came into the room to

familiarise themselves with the rabbits and vice versa. It was important that the rabbits were comfortable with a range of people so that they felt confident jumping into the box regardless of who would be handling them. This involved giving them some extra attention during the day or at end of day sign-offs such as encouraging them to play, talking to and stroking them.

We also ensured everyone knew exactly how to undertake the box training, how to time it and how to record the data. We did this by carrying out training sessions with everyone who was involved. For it to have successfully worked, the handling method needed to be carried out in the same way by everyone so that the rabbits were confident enough to jump in regardless of who was handling them.

Results

During the first week of box training, the rabbits took a while to jump into the box or did not jump in at all after 3 minutes of timing them. The box was new to them at this point and so they were hesitant to jump in, this was shown by them either running to the other side of their cage or sitting looking at the box not wanting to interact with it at all. After the first minute however, most showed interest in the box either sniffing it or rubbing their chins on the front of the box. If they did not jump in the box, they were picked up and carried by hand and the box was left in the playpen with them as normal.

After one week of exposure to the box, the rabbits overall became quicker jumping into the box and they seemed to be more comfortable with it and the effect of the different Vetbed also started to show (see figure 7).

The shared Vetbed was increasingly preferred during the second week and the rabbits were more interested

to jump in due to the mixed scent. We found they were sniffing and rubbing their chins on it a lot more in the initial few seconds of being presented with the box and were much less wary of its presence. They also did jump in a lot quicker, in almost half the time than in the first week. It looked as though they knew what to do and were excited to explore the smells on the Vetbed.

Although the own Vetbed group did jump into the box quicker than the first week, they did not continue to improve and slightly lost interest in the box after the first 2 weeks. There was less chin rubbing and sniffing when the rabbits were presented with the box and this was probably due to the Vetbed not being covered in the strong scent of the other rabbits, so it was not exciting enough for them. They did however jump in eventually.

These results seemed to be quite consistent through the 6 week period except for week 3 and 5 of the own Vetbed group and week 5 of the shared Vetbed group where the rabbits had myocardial infarction surgery. During the 3 day postoperative period, they were presented with the box as normal as they needed to be weighed and although we did record their data, it was however, discounted. The rabbits did not really show interest in the box during this time as they were recovering from surgery. We have found that since though, some rabbits are still happy to jump into the box during the post op period however, it is usually on days 2 and 3 of the checks.

By the end of the study, we found that the shared Vetbed rabbits were still very interested in the box every time the box was presented to them and they got quicker overall jumping in (an average of 14.6 seconds by the end of the study compared to an average of 42 seconds the first week). It seemed to become routine for them when they saw the box – they were coming to the front of the cage as soon as they saw us; perhaps in anticipation that they would get to come out into the box.

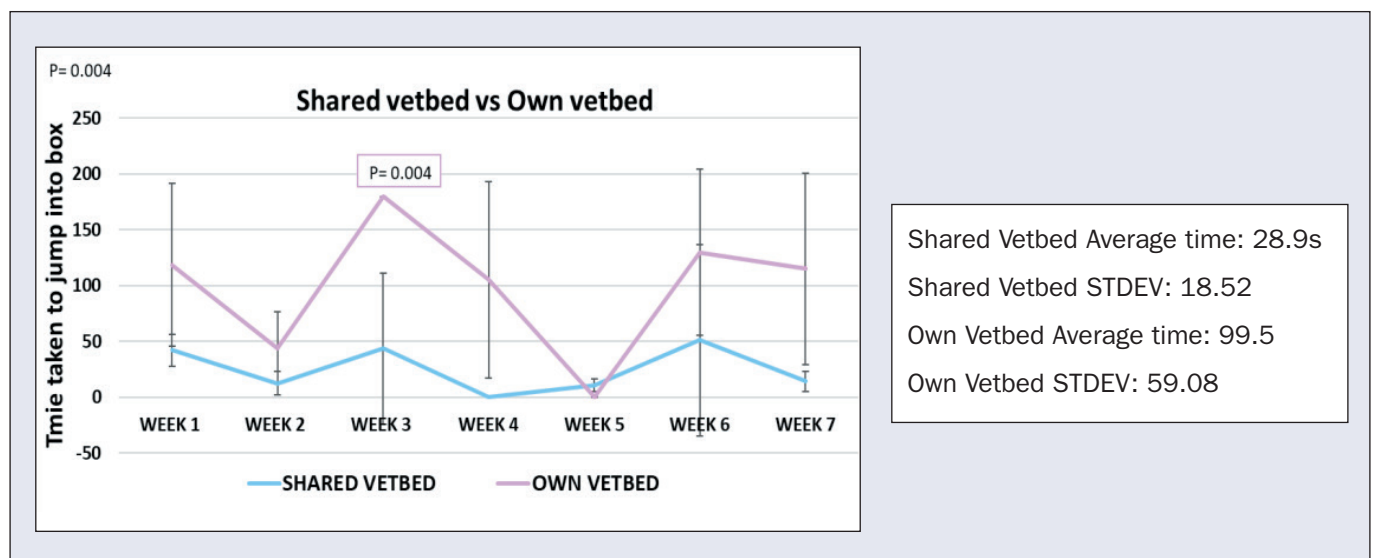


Figure 7.

The own Vetbed group still showed interest; however, it was lost after a minute or so, most likely due to the rabbits only smelling their own scent and therefore not as interesting. The rabbits would just walk away from the box and no longer interact with it after spending some time sniffing it. This meant the majority of the times they had passed the 3-minute time limit and the rabbits were carried by hand to their destination. At the start of the study the average time it took for them to jump into the box was 118 seconds, at the end it was 133 seconds.

Conclusions

Overall, we now know that rabbits can successfully accept the box training as a less invasive and more positive method of handling. The rabbits seem to really enjoy it and are often sat ready and waiting when they can see the box is coming towards them. We also know that the shared Vetbed helps to encourage the rabbits to jump into the carrier and it is a positive incentive for them whilst training.

We found that after a couple of weeks of training the rabbits with the shared Vetbed, when we offered freshly washed ones at the start of the week, still jumped in and were interested in scenting the box themselves. The rabbits appeared to have learned to jump into the box regardless of whether it was scented or not after getting used to the shared one for a while.

The rabbits typically will not jump into the box the first time but after two or three attempts during the first week will usually have learned what to do. This completely removes the need to physically pick up the rabbit as once the rabbit has jumped into the box, the rabbit can then be carried to the destination e.g., scales, playpen, etc., and back. It was easy to implement into the facility as a primary method of handling and typically, the rabbits will jump into the box, once they have learned the method, regardless of who is handling the box.

In our facility, we have multiple members of staff working in each room as well as routine visits by our vet and so it is an important point that they are able to jump in regardless of the member of staff handling them. A good example of successful box handling with unfamiliar people was when a group of visitors from a different facility came in to view the box training in action and were able to box the rabbits themselves with no problems. The rabbits jumped in almost straight away. Similarly, we have had our Named Veterinary Surgeon (NVS) come in recently to check on one of our rabbits and she commented on how easy it was to get the rabbit out of the cage to be taken to the scales and how she was impressed how the rabbit jumped into the box for her swiftly when she had never met that rabbit before.

Resource:	Quantity	Cost – each (£)
Animal carrier	4	27.11
Vetbed	2	40
Total:		67.11

Budget

We have broken down the budget of the box training. Both the Vetbed and carriers were relatively cheap and have lasted us a long time.

Box training female rabbits

Over the last few weeks, we have had the opportunity to trial the box handling with a few female rabbits we were housing. We were interested to see if they were as responsive as the males to the Vetbed/box with the same scent-based incentive.

We undertook all the same steps from acclimatisation to the box handling, however we were only able to trial a shared Vetbed due to the limited number of rabbits we had housed. The trial also only lasted 2 weeks as the females were all experimental controls. The training took place every day and as there was no playpen to be taken to in the box, the rabbits were walked around the room in the box for a couple of minutes.

We found that the female rabbits actually were just as responsive to the box and followed the same pattern during the training. At first, they were quite wary of the box but eventually came out from the back of their cage to explore the box. During the first 2 days, the box was placed inside of the cage rather than held at the front as the females were a lot more timid than the males and we wanted to let them explore in their cage where they felt safe. After those first days though, the box was held as normal at the front of the cage and the rabbits started to jump in relatively quickly after it was presented to them, all jumping in before the 3-minute time limit. We noticed they did not show the same sniffing, chin rubbing and scenting behaviours as the males did however it seemed as though they were jumping in to find a nice comfy place to hide.

We hope to continue to collect data from female rabbits in the future if we have the chance to house them again but it is encouraging to see that so far, they have taken well to the box handling.

Acknowledgements

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