

Lifting laboratory rats: A survey of methods, handlers' reasons and concerns, and rat behavioural responses

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ABSTRACT

Lifting mice by the tail is of animal welfare and scientific concern, but rat lifting methods are little researched, potentially differing from mice. Using an online questionnaire we explored different methods for lifting laboratory rats, alongside handlers' reasons and concerns, and rat behavioural responses. We received 249 valid responses from self-selected rat handlers across research sectors, job roles, and 26 countries (39% UK). In this sample, eight different lifting methods, plus 'other', were used; Shoulder saddle (39%), Chest-and-bottom support (20%) and Tail lifting (11%) were most common. Regarding respondents' reasons for using their main lifting method, those using Tail lifting selected 'rat comfort' significantly less frequently, and 'quick' more frequently, than did those using other methods. Most respondents had no concerns about their main lifting method, but those using Tail lifting were significantly more concerned about possible 'stress to the rat' than were those using Chest-and-bottom support. Concerns about speed, feasibility and scientific validity were rare. Rats reportedly defecated significantly more, and sniffed the hand less, when Tail lifted than when lifted using Chest-and-bottom support. Respondents who used Cupped hands reported rats to approach, sniff and climb onto their hand significantly more than respondents using certain other methods did. Notwithstanding potential sample bias, the findings suggest that lifting rats by the tail risks unnecessary suffering. It is also potentially concerning that the most common method, Shoulder saddle, showed no significant benefits over Tail lifting. Chest-and-bottom support, Cupping, Tunnel, and some rarer methods, may offer more refined methods for lifting rats.

1. Introduction

The impact of different handling methods on laboratory rats has rarely been considered in any depth (Baturaite et al., 2005), in contrast to the large body of research focusing on handling practices in mice (e.g. Hurst and West, 2010; Clarkson et al., 2018; Gouveia and Hurst, 2019; Henderson et al., 2020b; Sandgren et al., 2021; Novak et al., 2022). Lifting mice by the tail increases measures of anxiety and depression compared with lifting them either in cupped hands, using a tunnel (first reported in Hurst and West, 2010), or using an in-cage ladder (Sandgren et al., 2021). Stressful handling of mice can confound experimental findings due to the resulting physiological and behavioural changes (Maurer et al., 2008; Gouveia and Hurst, 2017; Nakamura and Suzuki, 2018; Hull et al., 2022).

Despite the possibility that inappropriate handling of rats causes similar issues to those observed in mice, and despite literature aimed at pet owners cautioning against tail-lifting of rats (e.g. Himsel, 1991; RSPCA, 2011; Starr, 2021), discussions of how handling methods may

affect laboratory rat welfare and scientific validity have been seldom seen. Advice given to pet owners has not been derived from peer-reviewed empirical evidence, and it may not always be directly applied to research animals mainly because the large numbers of rats usually handled by any one person in research context means that time available to handle each rat is very limited; therefore, different methods may be used for laboratory rats versus pet rats. Whilst rat lifting methods are rarely described in experimental literature, guidance for how to lift laboratory rats suggests methods including the 'Shoulder saddle' (using one hand over a rat's shoulders, e.g. Waynforth et al., 1998; Flecknell, 2001; Koolhaas, 2010), or lifting by the base of the tail (e.g. Wolfensohn and Lloyd, 2002; Pritchett and Corning, 2004; Bogdanske et al., 2010; Koolhaas, 2010). Guidance for pet rats, conversely often advises the 'Sandwich' method, sliding one hand underneath their belly to lift them up, while placing the other hand on their back to steady them (e.g. Himsel, 1991; Fox, 1997; Hill, 1998).

There are very few studies comparing different rat handling methods. Early laboratory studies, unsurprisingly, showed that gentle

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handling of rats, especially early in life, reduced measures of fear and anxiety. For example, a ‘gentle handling’ treatment in young rats increased handleability and reduced measures of fear in tests such as the open field and the novel object approach, compared with either ‘rough handling’, combined rough and gentle handling, or no handling (Bells, 1961). Of relevance, the rough handling treatment in that study included lifting rats by the tail, but also abusive actions, such as pinching the rats, throwing them in the air, and rubbing crisp packets in their faces, so the increased fear cannot be solely attributed to tail-lifting. More recently, Baturaite et al. (2005) compared the cardiovascular responses of seven rats to four handling and lifting methods: scruffing, encircling, using a plastic cone, and lifting and holding by the tail on the arm. All four methods significantly increased heart rate and blood pressure compared with baseline measures (Baturaite et al., 2005). In a further experiment, when rats were lifted using the Sandwich method, they produced both more ultrasonic chirps (suggesting positive welfare) and audible squeaks (possibly suggesting negative welfare) than when lifted by the tail (Burn, 2006); both lifting methods caused an increase in stress-related chromodacryorrhoea compared with before handling. Those results are difficult to interpret fully, but suggest that both methods were stressful to some degree.

Rats are routinely handled as part of their management, for example for cage-cleaning, as well as for experimental manipulation (Kemp, 2000; Deacon, 2006). Consequently, the methods used to lift rats are likely to have significant implications for their welfare throughout their lives. Before any refinement of laboratory rat handling can be achieved to optimise welfare, it is critical to understand what methods are currently used in practice, the reasons for the choice of method and concerns associated with them, as well as the behavioural responses of rats to these different methods of lifting.

1.1. Aims and hypotheses

Using a questionnaire, we aimed to describe the range and relative sample prevalence of methods used to lift laboratory rats, in an international population. We also aimed to collate information on the sources from which rat handlers learned how to lift rats, their reasons for using their main method, any concerns they may have had about it, and the behaviours they observed in rats when they lifted them up. The results could be used to underpin further research into refining handling methods for rats.

We hypothesised that, if tail handling or any other lifting method is aversive to rats, respondents would be more concerned about rat welfare with that method, and more rat behaviours indicative of fear or distress would be reported by respondents using that method to lift rats. Also, if the literature about tail handling of mice is impacting rat handling, then respondents who were more familiar with the mouse handling literature would be significantly less likely to lift rats by the tail, and more likely to use tunnel handling and cupping, than those who were less familiar with that literature.

2. Materials and methods

2.1. Survey creation and distribution

An online questionnaire was created using the Alchemer platform (formerly SurveyGizmo; Alchemer®, Louisville, USA). Before release, it was pilot tested by Royal Veterinary College (RVC) laboratory staff who work with rats, including an animal technician, an animal unit manager (also a NACWO and NTCO), a researcher, and a Named Veterinary Surgeon. Their feedback was used to improve the wording and content of the questionnaire. The survey received ethical approval from the RVC’s Social Sciences Research Ethical Review Board (URN 2020 0283).

The questionnaire was live between 11th of January and 23rd of February 2021. It was distributed via:

- notices to members of the Animal Welfare Management Discussion Group, HOLTIFs, the Lab Animal Vet list, LAVA, Norecopa, and VOLE;
- emails to the professional contacts of the authors (TC and CB) in industry, academia, charities, and regulators, including contacts in the USA, Canada and Switzerland;
- posts on the authors’ (TC and CB) social media accounts, including Facebook™ and LinkedIn®; and
- Tweets by AWERB hub, EARA, LASA, Learning Curve Development, NC3Rs, RSPCA Science Department, UAR, and a 3Rs coordinator in Switzerland, plus retweets.

2.2. Survey structure and content

The questionnaire (Supplementary file S1) covered lifting and restraint methods for both rats and guinea pigs within research settings, but only the rat lifting methods are within the scope of the current paper. In the introductory text, our stated aim was ‘to understand which handling methods were the most widely used on both rats and guinea pigs’, and why they were used.

We asked for participants who ‘currently handle rats or guinea pigs in a research setting’ and who were at least 18 years of age. The anonymous survey was estimated to take 10–15 min to complete and comprised four main sections: (A) general demographics and workplace type, (B) handling methods for rats and their strengths and weaknesses [3 pages], (C) handling methods for guinea pigs and their strengths and weaknesses [3 pages], (D) concluding questions.

The questions included multiple choice formats that required respondents to choose either one answer only, or to tick all that applied. For each question, ‘Unsure’ was offered as an answer so as not to force non-representative answers. There were also free text boxes that allowed respondents to expand on their answers if they wished. For answer lists that had no natural order, the order of the answers was randomised for each participant, to avoid bias arising from order effects. The only compulsory question was ‘Which rodents do you currently handle as part of your job?’, and this determined which pages of the survey the respondents would be offered to complete (e.g. they would not see the rat handling pages if they only ticked ‘guinea pigs’). Summaries of the sections relevant to the current study are as follows.

- The ‘General demographics and workplace type’ section included questions asking:
 - o whether respondents handled rats, guinea-pigs, both or none as part of their job;
 - o respondent gender, age and country of work;
 - o the industry type representing their work, their main job role, and the name of their institution (only to inform us ‘which respondents are independent of each other and how many organisations we have managed to survey. The institution name will NOT be included in any reports or communications.’); and
 - o the source from which respondents learned to handle laboratory rodents, and for what purposes they restrained them (e.g. health checks, restraint for procedures, educational purposes, or ‘I never restrain these animals’).
- The ‘Handling methods for rats and their strengths and weaknesses’ section included questions asking about use of alternative lifting methods:
 - o ‘Which rat lifting methods most closely match how people at your workplace lift/pick rats up?’ (a tick-all-that-apply question with 12 options, including ‘other’; Fig. 1);
 - o ‘Which best describes the single main lifting method that YOU personally use for juvenile rats (from weaning to about 100 g)?’; and
 - o ‘Which best describes the single main lifting method that YOU personally use for adult rats (weighing more than about 250 g)?’.
 Other questions in this section included the following.

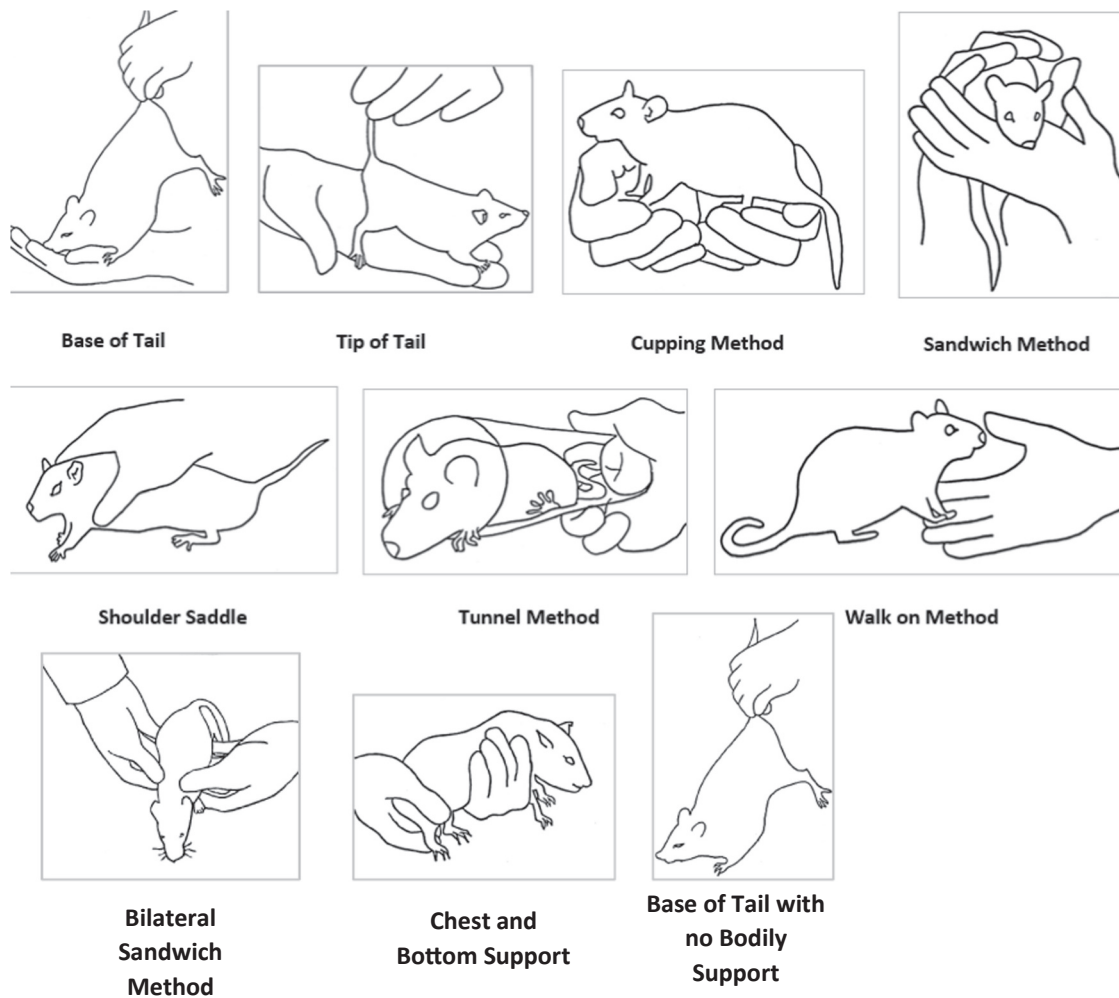


Fig. 1. Diagrams and names of alternative rat lifting methods, as shown in the questionnaire. The order in which the alternative methods were shown was randomised between respondents and between the three questions that covered rat lifting methods. It was explained that all diagrams show the rat as if viewed from the side, except for Bilateral sandwich, which was as if viewed from above. There were two other answer options that were not presented with a diagram, which were lifting using forceps or other tool and 'other'. The diagrams were drawn by T. Camacho for the purposes of this questionnaire.

- 'Why do you use your main lifting method for adult rats?' (10 options, e.g. quick, comfortable for the rat, reduces bites/scratches, or has little effect on scientific results);
- 'Do you have any concerns about your main lifting method for adult rats and, if so, what are they?' (11 options, e.g. no concerns, pain/stress for the rat, risks bites/scratches, slowness, or affects scientific results);
- 'Do you believe the positives outweigh the negatives of using this handling method, given the alternatives?'; and
- 'What do some of the adult rats do when you lift them using your main method? Tick all that apply to at least a few of the rats you would usually pick up, e.g. anything from all the rats doing it, down to about 1 in 20 of them doing it. Do not tick very rare behaviours.' (20 options, e.g. urinate, defecate, struggle, squeak, attempt to bite, or avoid the hand, or approach the hand).
- The 'Concluding questions' section included questions asking:
 - o the extent to which respondents were familiar with the research comparing the effects of lifting of mice by the tail, in cupped hands or in a tunnel;
 - o which methods staff used to lift mice at their workplace (no mice at the workplace, tail handling, cupping, tunnel, other (please explain), or unsure);
 - o whether respondents believed rat handling methods needed to be improved; and

o any further comments.

2.3. Criteria for exclusion

Prior to analysis the raw survey data were screened to ensure that responses data were eligible for inclusion. Respondents were excluded from the dataset for the current paper if they did not currently handle adult rats (or had not handled them within the two years prior to completing the survey), if they were less than 18 years of age, or had left their age category blank.

2.4. Statistical analysis

Data were exported from the Alchemer platform into Microsoft Office Excel for screening and cleaning prior to analysis which was conducted in IBM SPSS Statistics (version 28.0). Descriptive analyses were used to explore the distribution of survey responses, with a focus on how they related to each lifting method. The three variants of tail handling were described separately and then combined for statistical analysis. With regards to familiarity with research about mouse handling, 'very familiar' and 'familiar' were combined and compared against a combined category of 'slightly familiar' and 'unfamiliar'. Fishers Exact tests for independence were used to test for associations between lifting methods and why they were chosen, concerns the respondent had

regarding their use, and the behaviour of the rats exposed to each method. Statistical comparisons were only conducted for handling methods selected as the main method for lifting adult rats by more than 12 respondents, which totalled 112 comparisons. A two-tailed P-value of < 0.050 was the threshold for statistical significance. Formal statistical comparisons were not used for the remaining methods because they were too rare for analysis.

3. Results

3.1. Sample demographics

There were 541 responses to the survey, of which 249 were usable responses that met the criteria. The majority of respondents handled rats at the time the survey was completed (96% $n = 239$); those who no longer handled rats (4% $n = 10$) were included in the final dataset if they had only recently stopped handling rats as determined by their response to the free text question. Respondents worked in 26 countries, most being located in the UK (39% $n = 96$), the USA (12% $n = 29$) and Canada (11% $n = 27$) (Table 1). Nearly three-quarters of respondents were female (72% $n = 178$), with 27% ($n = 68$) being male. The median age of respondents was 35–44 years, with all age categories represented in the sample.

Respondents were asked to give the names of their employer so that

Table 1

Distribution of responses to the demographic questions. Unless the variable was ordinal, categorical responses are ordered from most to least common. $n = 249$ respondents.

Demographic Feature	Category selected	Number of responses	Percentage (%)
Gender	Female	178	71.8
	Male	68	27.3
	Non-binary	1	0.4
	Prefer not to say	1	0.4
Age group (years)	18–24	15	6.0
	25–34	74	29.7
	35–44	79	31.7
	45–54	52	20.9
	55–64	28	11.2
	65 +	1	0.4
	TOTAL ^a		
Country of work	UK	96	38.6
	European Union (EU)	76	30.5
	USA	29	11.6
	Canada	27	10.8
	Other TOTAL ^b	22	8.8
	TOTAL		
Main industry type of workplace	Academic research institution	168	67.5
	Pharmaceutical industry	34	13.7
	Government scientific research institution	21	8.4
	Contract research organisation	12	4.8
	Commercial laboratory animal breeder	10	4.0
	Other	4	1.6
	Animal technician	91	36.5
	Scientist	46	18.5
Main job role	Research animal veterinarian	41	16.5
	Animal unit manager	30	12.0
	Husbandry and procedures advisor	18	7.2
	Other	12	4.8
	Research student	11	4.4
	TOTAL		

^a EU comprised of: Spain ($n = 17$), Germany ($n = 13$), Netherlands ($n = 10$), Denmark ($n = 8$), Finland ($n = 7$), Ireland ($n = 5$), Sweden ($n = 5$), Romania ($n = 3$), Slovenia ($n = 3$), Czech Republic ($n = 2$), Greece ($n = 2$), Portugal ($n = 1$)

^b Other comprised of: Turkey ($n = 8$), Norway ($n = 5$), Australia ($n = 2$), Chile ($n = 2$), Colombia ($n = 2$), Argentina ($n = 2$), India ($n = 1$)

independence of the responses could be ascertained, but this was not a compulsory question. In total, 193 (78%) of respondents answered this question, naming 116 different employers. One pharmaceutical company was named by 17 respondents, although these were based in three different countries (the UK $n = 6$, the EU $n = 3$, and the USA $n = 8$). The remaining employers were represented by 1–9 respondents, with an overall mean of 1.7 respondents per named employer.

Two-thirds of respondents worked in an academic research institution (68% $n = 168$), with the remainder in four further industry types, plus ‘other’ (Table 1). Animal technicians were the most highly represented job role (37% $n = 91$), followed by five other roles, plus ‘other’; the 12 respondents who selected the ‘other’ option described this as including training related and managerial roles.

The majority of respondents were familiar with the research comparing the effects of lifting mice by the tail, in cupped hands or in a tunnel to some extent. Of the 188 respondents who answered this question, 46% ($n = 87$) were very familiar with this research, 30% ($n = 56$) were familiar, 13% ($n = 24$) were slightly familiar and 11% ($n = 21$) were unfamiliar with it.

3.2. Rat lifting methods used

3.2.1. Methods observed in the workplace

The methods that respondents had observed being used to lift rats by people at the same workplace as themselves encompassed all methods listed, with the exception of handling using forceps or another tool, which was not used at all (Fig. 2). The most common methods reported as being observed within the workplace were the Shoulder saddle (77%; $n = 192$), Cupping (65%; $n = 163$), and Chest-and-bottom support (59%; $n = 148$). Lifting by the tail tip was the least used (6%; $n = 15$), but when all variants of lifting by the tail were combined, Tail lifting was observed by over half of respondents (53%; $n = 132$). The ‘other’ methods described by respondents were typically expanded versions of those listed in the survey, although one respondent reported that rats were typically guided into a transport cage to avoid restraint.

When broken down by workplace, Tail lifting methods were used in 39% ($n = 98$) of workplaces. Of these, 72% ($n = 71$) used only one method of tail lifting, 22% ($n = 22$) used two tail lifting methods and 5% ($n = 5$) used all three tail lifting methods listed. For the remainder of the analysis the three tail lifting methods were grouped together.

3.2.2. Main methods used for juvenile and adult rats

The main methods that the respondents reported personally using to lift juvenile and adult rats spanned all answer options, except for forceps or another tool (Fig. 3). Nearly a quarter of respondents ($n = 55$) did not handle juvenile rats. Shoulder saddle (37% $n = 69$) was the most used method for juveniles, followed by Tail lifting (14% $n = 26$), Chest-and-bottom support (13% $n = 24$) and Cupping (13% $n = 24$). For adult rats, the most used method was again the Shoulder saddle (39% $n = 96$), followed by Chest-and-bottom support (20% $n = 50$), and Tail lifting (overall 11% $n = 27$; base of tail with forelimb support $n = 15$; base of tail only $n = 11$; tail tip $n = 1$). Four respondents did not answer this question.

3.3. Reasons for using each adult rat lifting method

The sources from which respondents learned to lift rats was explored in a tick-all-that-apply question. Animal technicians were the most frequently selected source (78%; $n = 193$), followed by experience of handling pet rodents (32%; $n = 79$) and scientific researchers (31% $n = 77$) (Fig. 4). The professional websites reported included those by NC3Rs ($n = 7$), AALAS ($n = 3$) and Procedures With Care ($n = 2$), and one respondent mentioned the website of an academic institution [urls not given]. Thirty-two respondents expanded on the ‘other’ ways they learned to handle rats. Common answers included during their undergraduate veterinary or veterinary nurse training ($n = 12$), from

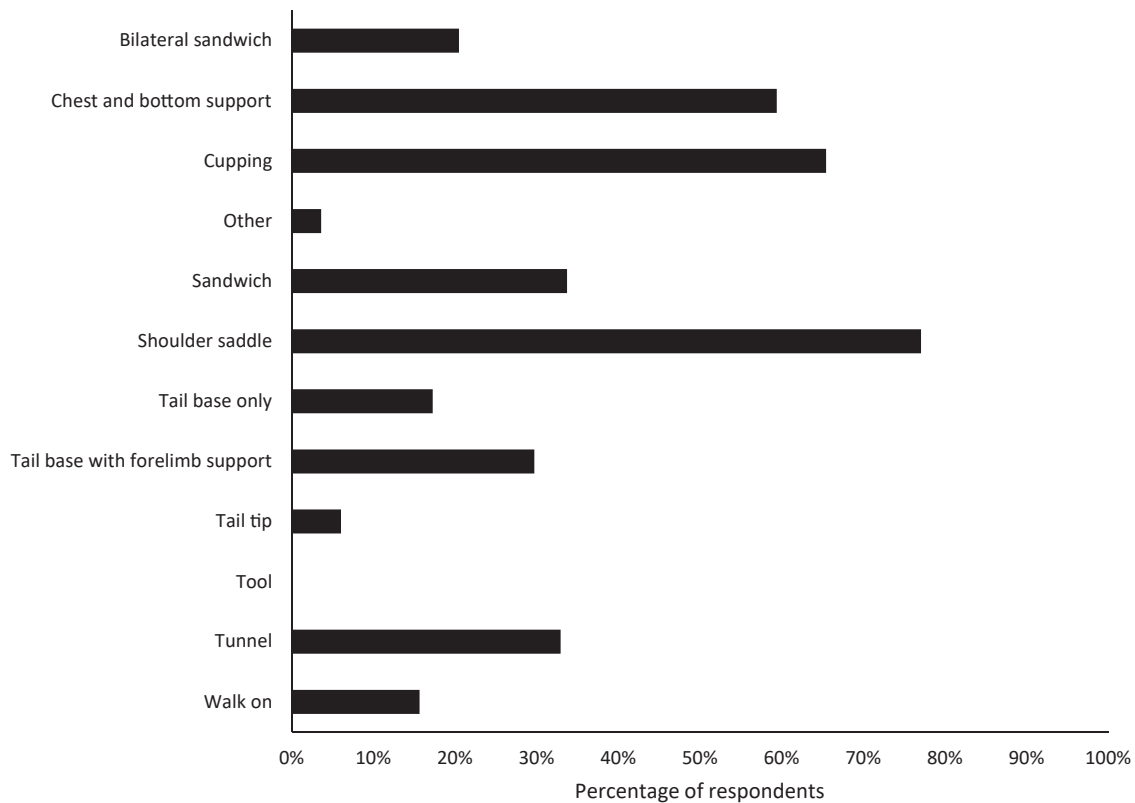


Fig. 2. Percentage (%) of respondents reporting each method that they had observed being used by people to lift adult rats in their workplace. Respondents could select multiple options. The 249 respondents selected 900 options in total. Rat lifting methods are arranged in alphabetical order.

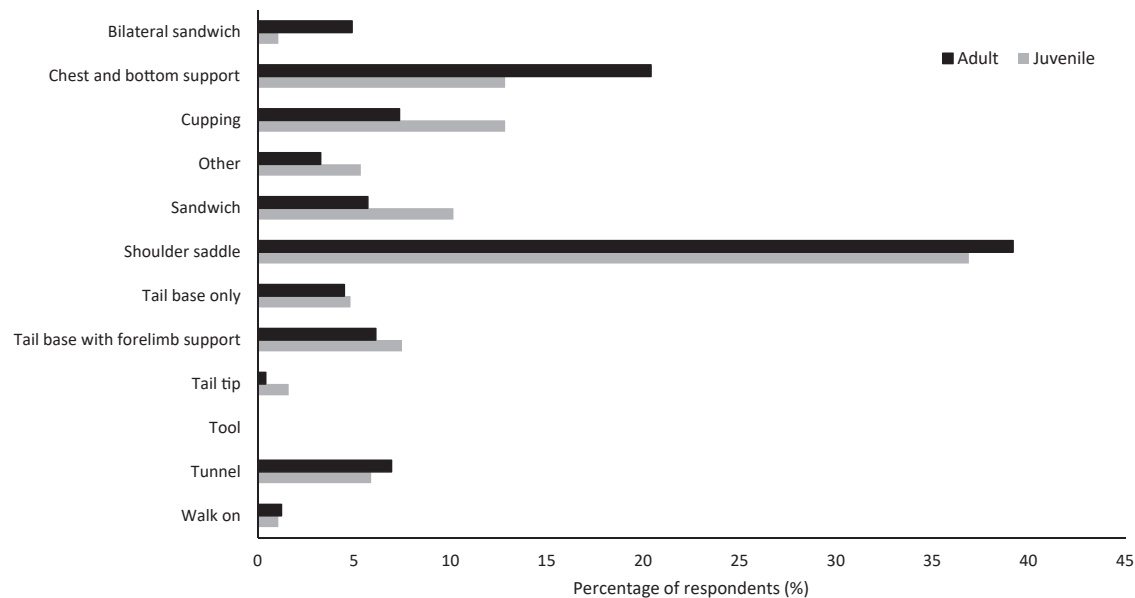


Fig. 3. Percentage (%) of respondents choosing each method as their main method of lifting juvenile (grey; $n = 187$) and adult (black; $n = 245$) rats. Respondents could select one method only.

veterinarians ($n = 7$) and from in-house trainers ($n = 7$).

Across lifting methods, the most commonly selected reasons for use were comfort of the rat, or the method being quick, easy or how the handler was taught. The reasons behind respondents' choice of main handling method for adult rats varied by method (Table 2). Fishers exact tests showed that handlers using Tail lifting were significantly less likely to select the comfort of the rat as a reason for choosing their method than

were handlers who used Chest-and-bottom support, Shoulder saddle, Cupping or Tunnel (Table 2 and Table 3). Accordingly, of the methods represented by 10 or more respondents for this question, the perception that lifting methods were relatively comfortable for the rat was most commonly selected by respondents who used the Tunnel (94%), or Cupping (89%) methods; and it was least commonly reported by those who used Tail lifting (50%; Table 2).

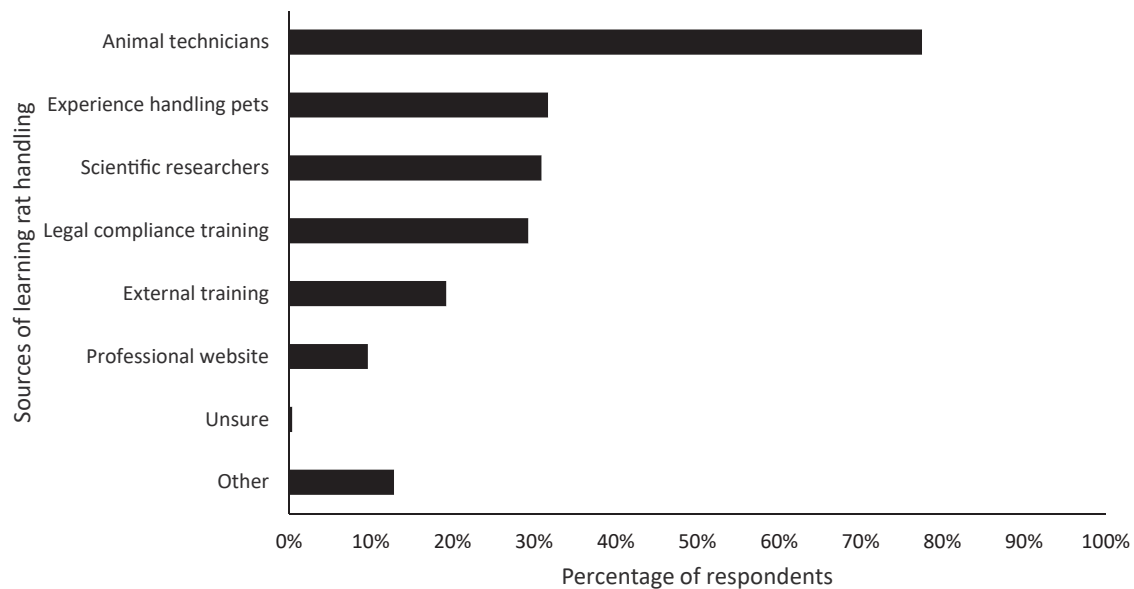


Fig. 4. The sources from which respondents ($n = 249$) learned their rat handling methods. Respondents could select multiple options.

Table 2

Reasons, concerns, and perceptions of whether the positives outweighed the negatives associated with alternative methods for lifting adult rats.

Question	Answer option (n responses / % per option)	Main rat lifting method							
		Bilateral sandwich	Chest-and-bottom support	Cupping	Sandwich	Shoulder saddle	Tail lifting	Tunnel	Walk-on
Reasons	Responses (n)	11	43	18	10	84	24	16	2
	Aids standardisation (%)	9.1	11.6	16.7	10.0	19.0	8.3	25.0	0.0
	Easy (%)	54.5	34.9	55.6	30.0	40.5	37.5	50.0	50.0
	How I was taught (%)	36.4	32.6 ^a	38.9	50.0	54.8 ^b	58.3	25.0	50.0
	Little effect on the results (%)	27.3	18.6	38.9	10.0	27.4	25.0	37.5	100.0
	Normal way to handle rats (%)	36.4	30.2	16.7	50.0	34.5	20.8	25.0	50.0
	Quick (%)	27.3	39.5 ^a	44.4	60.0	50.0	66.7 ^b	37.5	50.0
	Reduces bites and scratches (%)	18.2	23.3	44.4	10.0	35.7	41.7	25.0	50.0
	Relatively comfortable for the rat (%)	90.9	93.0 ^a	88.9 ^a	100.0	81.0 ^a	50.0 ^b	93.8 ^a	100.0
	Other (%)	0.0	4.7	0.0	0.0	2.4	4.2	18.8	50.0
Concerns	Responses (n)	11	40	18	10	81	23	16	2
	Could affect results (%)	0.0	2.5	5.6	0.0	2.5	8.7	0.0	0.0
	Could cause pain (%)	0.0	0.0	11.1	0.0	2.5	13.0	6.3	0.0
	Could stress the rat (%)	8.3	12.5 ^a	16.7	0.0	16.7	39.1 ^b	11.8	0.0
	Handler needs training or practice (%)	8.3	2.0	5.6	0.0	5.2	0.0	5.9	0.0
	Risks biting or scratching the handler (%)	25.0	27.5	11.1	40.0	14.8	8.7	6.3	0.0
	Takes a long time (%)	0.0	2.5	5.6	0.0	0.0	0.0	6.3	0.0
	The rats require training (%)	8.3	12.0	22.2	0.0	12.5	3.7	17.6	33.3
	Unusual so may affect standardisation (%)	0.0	0.0	0.0	0.0	0.0	0.0	5.9	0.0
	No concerns (%)	63.6	60.0	66.7	70.0	56.8	52.2	50.0	50.0
	Unsure (%)	0.0	2.5	0.0	0.0	3.6	4.3	12.5	0.0
	Other (%)	8.3	2.0	0.0	0.0	2.1	0.0	0.0	0.0
	Responses (n)	10	41	18	10	83	24	16	2
Positives outweigh negatives	No (%)	0.0	4.9	0.0	0.0	1.2	8.3	0.0	0.0
	Yes (%)	90.0	82.9	88.9	90.0	75.9	75.0	93.8	100.0
	Unsure (%)	10.0	12.2	11.1	10.0	22.9	16.7	6.3	0.0

The associated n is given for each questionnaire section, because the number of respondents differed in each case (the questions were optional so respondents could skip questions). The answer options and rat lifting methods are ordered alphabetically, with the exceptions of 'Other', 'Unsure' and 'No concerns'. ^{a, b} indicates statistically significant differences in behaviour between alternative lifting methods (Table 3).

The most commonly selected reasons for respondents to use Tail lifting were being 'quick' and 'how [the respondent] was taught' (Table 2). Tail handlers were significantly more likely to select 'it is quick' as a reason for using their method than were those who used Chest-and-bottom support (Table 3). Use of a method because it was how the respondent had been taught was most commonly selected by

respondents using Tail lifting (58%), and least commonly by those using the Tunnel method (25%; Table 2). Handlers who used Chest-and-bottom support were significantly less likely to report that it was how they had been taught than were those who used the Shoulder saddle method (Table 3). For the Walk-on method, statistical comparison was not possible, because there were only two respondents, but 'little impact

Table 3

Statistically significant comparisons between handling methods used to lift adult rats regarding the reasons respondents used those methods, any respondent concerns, and rat behavioural responses.

Question	Answer selected	Majority handling method (% selecting answer out of n respondents) ^a	Minority handling method (% selecting answer out of n respondents) ^a	Odds ratio (95% CI)	P-value
Reasons: “Why do you use your main lifting method for adult rats?”	It is relatively comfortable for the rat	Shoulder saddle (81.0% of 84)	Tail (50.0% of 24)	0.235 (0.093–0.632)	0.004
		Chest-and-bottom support (93.0% of 43)	Tail (50.0% of 24)	13.33 (3.05–47.66)	< 0.001
		Cupping (88.9% of 18)	Tail (50.0% of 24)	8.00 (1.56–39.26)	0.010
		Tunnel (93.8% of 16)	Tail (50.0% of 24)	15.00 (1.81–169.70)	0.005
	It is quick	Tail (66.7% of 24)	Chest-and-bottom support (39.5% of 43)	3.06 (1.05–8.37)	0.043
	It is how I have been taught to lift rats	Shoulder saddle (54.8% of 84)	Chest-and-bottom support (32.6% of 43)	2.51 (1.19–5.28)	0.024
	It could cause stress for the rat				
Concerns: “Do you have any concerns about your main lifting method for adult rats and, if so, what are they?”	It could cause stress for the rat	Tail (39.1% of 23)	Chest-and-bottom support (12.5% of 40)	4.50 (1.38–14.58)	0.026
Rat behaviour: “What do some of the adult rats do when you lift them using your main method?” ^b	Defecate	Tail (26.1% of 23)	Chest-and-bottom support (4.9% of 41)	6.88 (1.44–35.04)	0.021
	Sniff the hand	Chest-and-bottom support (63.4% of 41)	Tail (26.1% of 23)	4.91 (1.50–16.23)	0.009
		Cupping (72.2% of 18)	Tail (26.1% of 23)	7.37 (1.92–32.74)	0.005
	Approach the hand	Cupping (61.1% of 18)	Chest-and-bottom support (24.4% of 41)	4.87 (1.44–16.42)	0.009
	Attempt to crawl onto hand	Cupping (44.4% of 18)	Shoulder saddle (19.8% of 81)	3.25 (1.02–9.49)	0.036
	‘Very familiar’ or ‘Familiar’	Shoulder saddle (81.7% of 71)	Tail (45% of 20)	5.45 (1.96–16.68)	0.003
Familiarity with mouse handling research: “Are you familiar with the research comparing the effects of lifting of mice by the tail, in cupped hands or in a tunnel?”		Chest-and-bottom support (77.8% of 36)	Tail (45% of 20)	4.28 (1.27–13.86)	0.019
		Tunnel (85.7% of 14)	Tail (45% of 20)	7.33 (1.20–37.37)	0.030
		Bilateral Sandwich (90.0% of 10)	Tail (45% of 20)	11.0 (1.37–129.80)	0.024

Respondents could tick all answers that applied to the questions analysed here, other than regarding their familiarity with mouse handling research where they could only tick one answer. The results were calculated using pairwise Fishers Exact tests comparing the five most common handling methods; other handling methods were too rare for formal analysis. In total, 112 comparisons were analysed, but only the 16 that reached statistical significance are reported here.

^an refers to the number of respondents who both answered the question and used the handling method described in the cells.

^bA qualification was added to this question, specifying that respondents should "Tick all that apply to at least a few of the rats you would usually pick up, e.g. anything from all the rats doing it, down to about 1 in 20 of them doing it. Do not tick very rare behaviours."

on the results' was chosen as often as rat comfort was by two of the three respondents using the Walk-on method (Table 2).

'Aiding standardisation' and 'having little effect on scientific results' were relatively rarely selected as reasons for using most handling methods (Table 2). Having little effect on scientific results was most commonly selected as a reason for using Cupping (39%), and Tunnel (38%) methods.

Twenty-five respondents left additional comments to elaborate on their choice of handling method. Some respondents used this space to clarify that the body of the rat was fully supported as soon as possible once lifted. One respondent who routinely used the Shoulder saddle as their main handling method stated that this was because it "avoids the stress of tail handling" and another who used the Tunnel method stated, "It is my belief that these are the least stressful".

3.4. Concerns about using each adult rat lifting method

Respondent concerns about their main method of handling adult rats were explored in a tick-all-that-apply question, with a substantial percentage of respondents for all nine methods reporting that they had no concerns with their choice of main method (Table 2). Tail handlers were significantly more likely to cite causing stress to the rat as a concern than

were those using Chest-and-body support (Table 3). This concern was most frequently reported with Tail lifting (33%), but was never selected for the Sandwich method, although the sample size was too low for formal comparison between these two methods. No other statistically significant differences in concerns between handling methods were found.

Respondents who used Sandwich were the ones who most frequently selected that they had no concerns (70%). The risks of stressing the rat, of scratches or bites to the handler, and/or that the rat required training were the three most common concerns across all methods. Of those methods represented by ten or more respondents, concern about bites and scratches to the handler were most frequently reported for the Sandwich (40%), Chest-and-bottom support (28%) and Bilateral sandwich (27%) methods; it was least commonly reported for the Tunnel (6%) method (Table 2); there were no statistically significant differences. Concerns about the methods taking too long, or affecting scientific results or standardisation were rarely selected by the respondents.

Despite these concerns, most respondents believed that the positives outweighed the negatives of their chosen handling method (Table 2). Only three of the nine handling methods had respondents who disagreed with this statement (Tail lifting (8%), Chest-and-bottom support (5%), and Shoulder saddle (1%)), and eight of the nine methods had

respondents who were unsure whether the positives outweighed the negatives or not; these differences showed no statistical significance.

3.5. Rat behaviour reported when lifted using each method

Respondents were asked to report behaviour displayed by at least some of the adult rats they lifted, with the exception of very rare behaviours (Table 4). Adult rats reportedly approached the hand to some degree with all methods. Respondents who mostly used Cupping reported that rats attempted to crawl onto the arm significantly more than those using Shoulder saddle, and that rats approached the hand significantly more than those who used Chest-and-bottom support (Tables 3 and 4). Sniffing the hand prior to lifting was reported significantly less for Tail lifting than for both Chest-and-bottom support and for Cupping.

Rats were reported to urinate and defecate during handling for all the methods. Defecation was most commonly reported for Tail lifting (22%), and this was significantly more common than when Chest-and-bottom support was used (Tables 3 and 4). No differences in urination reached significance.

Struggling was reported to occur during all methods, but most commonly by respondents who used the Shoulder saddle (21%), and Tail lifting (19%), although there were no significant differences. Attempts to bite the handler were relatively rare, but were reported for five out of the nine handling method (Tail lifting, Shoulder saddle, Tunnel, Sandwich and Bilateral sandwich) (Table 4).

3.6. Familiarity with mouse handling research

Most of the 185 handlers who responded when asked how familiar they were with the existing mouse handling research declared that they were 'very familiar' with it (46.5%); also, 29.2% were 'familiar', 13.0% 'slightly familiar' and just 11.4% 'unfamiliar'. Those respondents who used Tail lifting as their main method were significantly less familiar

with that research than were respondents who used Shoulder saddle, Chest-and-bottom support, Tunnel or Bilateral sandwich methods (Table 3). There was no statistically significant difference in familiarity between those who used Tail lifting versus those using Cupping.

4. Discussion

The findings of this survey provide insight into how rats are currently lifted in laboratory settings, the reasons these methods are chosen, respondent concerns associated with them, and how the rats behave prior to and during being lifted using these methods. The sample population was self-selected, so the prevalences and viewpoints presented cannot be interpreted as being completely representative. The demographic composition of the survey sample was very similar to that achieved by a recent international survey on handling laboratory mice (Henderson et al., 2020b). The sample spanned 26 countries and encompassed a range of workplace industry types and laboratory worker roles, with academic research institutions and animal technician the most represented. There were eight methods used to lift rats, plus 'other', and the main methods respondents used to lift rats broadly reflected those they reported being used by others in their workplace.

In this sample population, the three most common methods used to lift adult rats were the Shoulder saddle, Chest-and-bottom support and Tail lifting (either lifting by the tail base with forelimb support, the tail base only, or more rarely the tail tip); Cupping was also commonly observed in facilities. Lifting a rat by the tip of the tail or by the tail base without forelimb support is not recommended due to the stress it is presumed to induce and the risk of the skin detaching (e.g. Kemp, 2000; Deacon, 2006), and further concerns are raised by the current study as discussed below.

Table 4

Percentages of rat handlers who reported adult rat behaviour when using their main lifting method.

Behaviour	Main lifting method (% respondents reporting behaviour)								
	Bilateral sandwich (n = 11)	Chest-and-bottom support (n = 41)	Cupping (n = 18)	Sandwich (n = 9)	Shoulder saddle (n = 81)	Tail lifting (n = 23)	Tunnel (n = 16)	Walk-on (n = 2)	Other (n = 8)
Approach the hand before being lifted	45.5	24.4 ^a	61.1 ^b	66.7	38.3	34.8	37.5	100.0	75.0
Attempt to bite	9.1	0.0	0.0	11.1	2.5	8.7	12.5	0.0	0.0
Attempt to crawl onto handler's arm	27.3	19.5	44.4 ^a	44.4	19.8 ^b	39.1	18.8	100.0	25.0
Attempt to hide as you approach to lift them	36.4	41.5	22.2	33.3	30.9	30.4	18.8	0.0	62.5
Attempt to jump away	0.0	22.0	22.2	55.6	9.9	17.4	12.5	0.0	12.5
Avoid the hand before being lifted	36.4	24.4	16.7	22.2	17.3	26.1	25.0	0.0	37.5
Defecate	18.2	4.9 ^a	16.7	22.2	17.3	26.1 ^b	12.5	0.0	25.0
Move about during lifting	18.2	39.0	22.2	66.7	29.6	39.1	25.0	50.0	37.5
Produce red porphyrin around the nose after lifting	0.0	0.0	0.0	0.0	1.2	0.0	0.0	0.0	0.0
Show the whites of their eyes	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Sniff the hand	36.4	63.4 ^a	72.2 ^a	44.4	46.9	26.1 ^b	43.8	100.0	50.0
Stay still as you approach to lift them	36.4	34.1	33.3	22.2	34.6	39.1	18.8	0.0	37.5
Stay very still during lifting	63.6	24.4	11.1	44.4	30.9	26.1	18.8	0.0	37.5
Struggle	18.2	14.6	5.6	44.4	21.0	21.7	6.3	0.0	25.0
Teeth chatter	0.0	0.0	0.0	11.1	0.0	0.0	6.3	0.0	0.0
Urinate	18.2	9.8	27.8	11.1	17.3	21.7	6.3	0.0	25.0
Vocalise (squeak)	27.3	19.5	27.8	22.2	17.3	21.7	6.3	0.0	25.0
Other	0.0	2.4	0.0	0.0	0.0	0.0	6.3	50.0	0.0
None of these behaviours	0.0	0.0	5.6	0.0	3.7	4.3	13.3	0.0	0.0

The behaviours and rat lifting methods are ordered alphabetically, with the exceptions of 'Other' and 'None'. There were 209 respondents who answered this question.

^a, ^b indicates statistically significant differences in behaviour between alternative lifting methods (Table 3).

4.1. Reasons for using each adult rat lifting method

Previous authors suggested that only a few basic rat handling methods are in common use, and that choice of method reflects tradition and personal preferences, with manual handling techniques used preferentially by experienced handlers, and devices used by inexperienced handlers, although the evidence underlying this was not provided (Baturaite et al., 2005). In contrast, our study shows that many different methods were used, most being manual, and that respondents' choices of lifting method appeared to be driven primarily by their perception of what method was most comfortable for the rat, with the exception of Tail lifting. If it is true that most rat handlers prioritise rat comfort when deciding on a handling method, then this is encouraging for rat welfare, although this finding could be an artefact of the self-selected sample population, e.g. if the questionnaire was mostly answered by handlers who had an interest in animal welfare.

Tail lifting was chosen primarily because it was perceived as quick and because it was the method the respondent was taught to use. In a previous study (Henderson et al., 2020b), time constraints were also commonly cited as a reason for Tail lifting mice rather than using Tunnel handling. However, it is notable that slowness was very rarely considered a concern for any of the lifting methods covered in the current survey, so it is unclear that being quick would offer a genuine advantage of Tail lifting over any other method.

Respondent knowledge about the mouse handling literature (e.g. Hurst and West, 2010; Clarkson et al., 2018; Gouveia and Hurst, 2019; Henderson et al., 2020b; Novak et al., 2022) was also associated with a significantly reduced likelihood of them reporting themselves to lift rats by the tail. This is consistent with findings relating to mouse handler roles, in which researchers were both less aware of the mouse handling research, and more likely to lift mice by the tail, than were animal care staff (Henderson et al., 2020b). The current findings could indicate that the research cautioning against lifting mice by the tail is impacting, not only mouse handling, but rat handling too, with rat handlers assuming that tail lifting would be harmful to rats as it is for mice. This fits with most respondents who used other methods stating that the comfort of the rat was a key consideration in using their main lifting method. Alternatively, because the result is correlational, with uncertain causation, it could be that respondents who avoid lifting rats by the tail happen to be those who attend more training opportunities about rodent husbandry and/or read literature on the topic more. It is interesting to note that a variety of rat lifting methods were used by these more knowledgeable respondents instead of tail handling, and these alternative methods were not restricted to the tunnel and cupping methods that have been recommended for mice (Hurst and West, 2010). Here, Tunnel, but not Cupping, was used significantly more by handlers familiar with the mouse handling literature than by those less familiar with it; and Shoulder saddle, Chest-and-bottom support and Bilateral sandwich were also used by these more informed respondents.

4.2. Concerns about using each adult rat lifting method

For each of the methods used to lifting adult rats, over half of the respondents in this sample reported having no concerns about their main method. It is possible that handlers who had concerns about their lifting method may be underrepresented in this questionnaire, e.g. due to reluctance to answer questions about a practice they were not comfortable with. Nevertheless, the risk of scratches and bites to the handler was reported as a concern for most methods, especially for some of the two-handed lifting methods: Sandwich, Chest-and-bottom support and Bilateral sandwich. Rats are liable to bite if they are anxious or not handled appropriately (Deacon, 2006); yet, despite the concerns of those working with laboratory rats, unprovoked attacks are rare (Kemp, 2000). The method for which concern about bites and scratches was lowest was Tunnel lifting, perhaps because the tunnel itself forms a barrier between rat and hand. There is also less risk of rats being

inadvertently squeezed or pinched by the hand when lifted in a tunnel, provided that tunnels are large enough to contain rats comfortably, so bites and scratches might be less likely to be provoked compared with when the hands are used directly.

Concerns that the method may stress the rats were also relatively common, and were reported significantly more for Tail lifting than for Chest-and-Bottom support. This concern may be well founded in line with the literature showing that lifting mice by the tail causes stress (e.g. Hurst and West, 2010), if the same is true for rats.

A perception that the rats required training was the third most common concern across most methods, with no statistically significant differences between methods. It has been suggested that handling per se is intrinsically stressful for rats and they may not readily habituate to it (Balcombe et al., 2004). Indeed, mice did not habituate to tail handling and instead showed strong aversion, but habituation to lifting within tunnels, and to a lesser extent cupped hands, occurred rapidly (Gouveia and Hurst, 2013, 2019). It would be important to understand better why training rats to accept handling methods was perceived as a concern in the current study; for example, it could be because some handling methods are difficult to use for fearful rats, or because the training slows handling down. However, both the difficulty of each method and the time taken for handling were very rarely selected as concerns for any of the lifting methods, so the true explanation is unclear.

Most respondents for all methods reported that they felt the positives of their lifting method outweighed the negatives. However, 8% of those using Tail lifting felt the opposite, as did 5% of those using Chest-and-Bottom support, and 1% of those using Shoulder saddle. These differences were not statistically significant.

4.3. Rat behaviour reported when lifted using each method

Reported rat behavioural responses to being lifted varied significantly across methods, with Cupping associated with behaviours potentially indicating a positive response by the rat, including approaching the handler's hand, sniffing their hand before being lifted, and crawling up the handler's arm. Such behaviours suggesting willingness to interact with the handler, and are among the measures by which Cupping and Tunnel handling have been judged as non-aversive lifting methods in laboratory mice (e.g. Hurst and West, 2010). Tunnel lifting showed no significant effects on reported positive handling behaviours here, but this could be because relatively few respondents used that method, resulting in a small sample ($n = 16$). Previously, Tunnel handled mice interacted more with their handler and showed fewer indicators of anxiety than Tail handled mice, even when lifting of both groups was followed by subcutaneous injection (Gouveia and Hurst, 2019), repeated restraint, intraperitoneal injections and anaesthesia (Henderson et al., 2020a). Thus, refining lifting protocols to use non-aversive methods can substantially improve the welfare of laboratory mice beyond each lifting event, and may also have similar benefits in rats.

Here, Tail lifting was associated with significantly more reports of defecation, and less sniffing of the hand, than was Chest-and-bottom support and Cupping. This reinforces that the respondents' concerns about Tail lifting causing stress to the rats may be well founded. Increased defecation can indicate anxiety, fear and/or stress (e.g. Latane, 1969; File and Vellucci, 1979; Ferré et al., 1995; Russo and Parsons, 2021). Mice handled by their tails also show increased defecation, alongside behavioural indices of anxiety, anhedonia, and reduced interactions with the handler in comparison to those handled using a tunnel (Clarkson et al., 2018; Nakamura and Suzuki, 2018; Henderson et al., 2020a). It is possible that our questionnaire results may be biased by respondent expectation bias (Tuytens et al., 2014), if, for example, rat handlers who were aware of the mouse handling findings were more likely to answer the questionnaire than were other handlers, so treatment-blind experimental observations of rat behaviour following handling via different methods may be needed to confirm this

in rats.

Whilst Tail lifting of rats is thus of concern, it is also somewhat concerning that Shoulder saddle was not significantly better than Tail handling in any respect. Shoulder saddle was the most commonly used method, with 96 (39% of) respondents using it as their main method, and therefore should have provided more statistical power to detect differences than any of the other methods used. It is possible that, with a larger sample population, significant differences may have been found. However, even in this sample, less common methods including Chest-and-bottom support and Cupping, did reach statistical significance when compared with Tail lifting, so Shoulder saddle may not offer as much improvement as those methods. Like Tail lifting, but unlike the other methods, Shoulder saddle is a one-handed grip that does not offer support to the rats from underneath; instead the rat is entirely approached from above (e.g. Waynforth et al., 1998; Flecknell; Koolhaas, 2010). The only significant effect on rat behaviour found for Shoulder saddle was that rats were significantly less frequently reported to crawl onto the handlers' arms when lifted this way than by Cupping. These results raise the possibility that Shoulder saddle is an aversive method of lifting rats. This concern urgently needs to be addressed by robust research, given that Shoulder saddle is the most common method for lifting rats, and was reported to be significantly more often taught than Chest-and-Bottom support was.

Despite handler concerns, attempts to bite were rare and only reported for five of the methods: Tail lifting, Shoulder saddle, Tunnel, Sandwich and Bilateral sandwich; these methods did not completely align with those that respondents were most concerned about in terms of risk of bites and scratches.

4.4. Conclusions

This international survey revealed that many different methods of lifting laboratory rats were used by respondents, the most common being Shoulder saddle, Chest-and-bottom support, Tail lifting and Cupping. In this sample, choice of method was largely driven by respondent perception of what was comfortable for the rat, with the exception of Tail lifting which was chosen because it was what was taught and was perceived as quick. Respondents reported concerns with all methods, although they considered the positives of their chosen method to outweigh the negatives. Cupping and Chest-and-Bottom support were associated with more positive rat behaviours, and Tail lifting was associated with increased defecation and less sniffing of the hand. Both positive and negative behaviours were reported for all methods, and further research is required to understand the nuances of the variation seen. Most respondents were familiar with the research on handling mice to some degree, but those who were less familiar with it were more likely to lift rats by their tails.

The results raise concerns for two main reasons. Firstly, rats were still commonly lifted by the tail (11% of handlers, and in 39% of workplaces) and both handler perceptions and reported rat behaviour suggest that it is stressful for rats. Secondly, Shoulder saddle – a one-handed lift without ventral or plantar support – was the most common method in this study, may be commonly taught, and yet did not show significant benefit over Tail lifting in any measure. Empirical research is urgently needed to ascertain whether Shoulder saddle is aversive or not, which alternatives are most humane and feasible, and how to effectively improve the behaviour of rat handlers to support the use of non-aversive handling methods for laboratory rats.

CRediT authorship contribution statement

Charlotte C. Burn, Trinity Camacho: Conception and design of the study, Acquisition of data. **Jo Hockenhull, Charlotte C. Burn:** Analysis and interpretation of data. **Jo Hockenhull, Charlotte C. Burn, Trinity Camacho:** Drafting the article. **Charlotte C. Burn:** Revising it critically.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.applanim.2023.106077.

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