

Using a runway paradigm to assess the relative strength of rats' motivations for enrichment objects

Louise A Hanmer, Patricia M Riddell & Claire M Williams

School of Psychology & Clinical Language Sciences, University of Reading

Correspondence Address: Dr Claire M Williams,

School of Psychology & Clinical Language Sciences,

University of Reading,

Earley Gate,

Whiteknights,

Reading, RG6 6AL,

UK.

Email: claire.williams@rdg.ac.uk

Phone: 44 (0)118 378 5740

Fax: 44 (0) 118 378 6715

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Abstract

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1 While laboratory animals should be provided with enrichment objects in their cages, it 2 is first necessary to test whether the proposed enrichment objects provide benefits that 3 increase animal welfare. The two main paradigms currently used to assess proposed enrichment objects are the choice test, which is limited to determining relative 4 5 frequency of choice, and consumer demand studies that can indicate the strength of a 6 preference but are complex to design. Here we propose a third methodology, a runway 7 paradigm, that can be used to assess the strength of an animal's motivation for 8 enrichment objects, is simpler to use than consumer demand studies and is faster to 9 complete than typical choice tests. Time spent with objects in a standard choice test 10 was used to rank several enrichment objects in order to compare with the ranking 11 found in our runway paradigm. The rats ran significantly more times, ran faster and 12 interacted for longer with objects with which they had previously spent the most time. 13 It was concluded that this simple methodology is suitable for measuring rats' 14 motivation to reach enrichment objects. This can be used to assess the preference for 15 different types of enrichment objects, or to measure reward system processes.

Introduction

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2 Improving the welfare of captive animals is an important issue, particularly for 3 laboratory animals. A common method used to increase welfare is to provide caged 4 animals with additional enrichment items that allow them to perform important natural behaviours (e.g. chewing, foraging, and nesting). Environmental enrichment 5 6 (EE) implies that the items introduced to the cage will have a positive effect on the 7 welfare of the animals, for instance, by increasing nesting, exploration or play 8 behaviour. Currently, cages may be 'enriched', often with some bedding material 9 (Bradshaw and Poling, 1991; Townsend, 1997; Heizmann et al., 1998; Van deWeerd 10 et al., 1998a,b; Patterson-Kane, 2003) or tubes on which rats can chew (Bradshaw and 11 Poling, 1991; Chmiel and Noonan, 1996; Sorensen et al., 2004). While, in the UK, it 12 is a requirement that Home Office licence holders provide some form of enrichment 13 for caged animals, the degree of provision, and types of objects used are not uniform 14 across laboratories. This is partly due to the lack of information available to determine 15 which provisions are most useful. Hence, a simple methodology that can be used to determine preference for EE objects would be of use in the process of standardising 16 17 EE provision.

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Environmental enrichment research often attempts to assess an animal's motivation for a particular stimulus by measuring either the choice(s) the animal makes when faced with two or more different stimuli, or measuring the time the animal spends with each stimulus (e.g. Chmiel & Noonan, 1996; Heizmann *et al*, 1998; Van de Weerd *et al*, 1998). However, motivational experiments can also be used to judge preference by requiring animals actively to work for objects, for example by requiring the animal to press a lever in order to gain access to the enrichment (e.g. Sherwin &

Nicol, 1997). This assumes that animals will be more motivated to work for some objects, thus enabling a rank order for motivational preference to be established. The current study was designed to determine whether a simple runway paradigm could be used to assess the strength of rats' motivation to run to different enrichment objects, thus providing a measure of preference based on the rats' motivation to work (i.e. run down the runway) for a particular object. This would improve on previous passive measures since these require that rats simply interact with an object that is placed in their environment. Additionally, it would improve on consumer demand designs in being simpler to design and quicker to run.

Typically, in a runway paradigm, a food reward (i.e. sucrose solution) is placed in the reward box at the end of the runway, a rat is placed in the start box and released, at which point it runs to gain access to the palatable drink. Consistent findings show that as the drink is made progressively sweeter, rats run faster to reach it, indicating that it is more rewarding and that rats are more strongly motivated to reach it. When the concentration of the sucrose solution is reduced and therefore less rewarding, rats are found to run more slowly or fewer times indicating a lower motivation to reach it (Flaherty et al, 1973; Burns et al, 1984; Burns & Griner, 1993).

The runway paradigm was adapted to assess rats' relative motivation to run for enrichment objects instead of food, by placing an enrichment object at one end of the runway, and the rat at the other end. The rat then had to 'work', by running the length of the runway in order to be able to interact with the object. It was assumed that the rats would be more highly motivated to reach an object that they had previously ranked top, as opposed to an object that had been ranked bottom. This would be

indicated by the rat running more often, running faster, and/or interacting longer with the top ranked object. Due to the nature of the runway paradigm, the rats were only allowed to interact with the enrichment object for a short time upon reaching it (5 seconds in Study 1 and 10 seconds in Study 2). This was in order to maintain the rats' motivation to gain access to the object, since if the rat was given as much time as it liked to interact with the object on the first encounter, it might be less motivated to run on subsequent trials. Despite the short length of time permitted to interact with the object, rats still had the chance to choose to interact with some objects for longer than others over the course of the experiment, providing a ranking for the objects.

In the first study, two objects which had previously been ranked top and bottom in an open field paradigm were tested. We predicted that rats would run more often, and faster, to the top ranked object compared to the bottom ranked object, and that they would interact with the top ranked object for longer when they reached it.

Study 1

In order to determine whether the runway paradigm was suitable for testing motivational preferences for enrichment objects, we used objects for which a rank order had previously been determined by measuring the time spent with the object in an open field (Williams, Hanmer & Riddell, 2008). In the first study, objects with which rats had previously spent a lot of time (top ranked), or very little time (bottom ranked), were selected for assessment in the runway. This resulted in two sets of objects, one pair consisting of a top ranked plastic house and a bottom-ranked cardboard tube (EE) and the other set a top ranked large block made from plastic Lego® and a bottom ranked single Lego® brick covered in synthetic fur (Lego®).

Two replications were carried out with each set of objects in order to thoroughly check that this methodology provided consistent rankings and to determine whether they were the same as had previously been recorded using time spent with the object. This confirmed that rats would run faster, and more often to a top ranked object, but ceiling effects prevented demonstration that rats would also interact with the top ranked object for longer. In Study 2, we increased the length of time available to interact with a set of objects, and demonstrated that this parameter also differentiated between top and bottom ranked objects, with rats interacting for longer with a top ranked object.

1 Methodology for Study 1

2	Animals
3	Adult male Lister-Hooded rats (N=18), weighing 460-600g, were group housed in
4	standard cages under standard conditions (n = 4 rats per cage). Each cage measured
5	23cm (H) x 32cm (W) x 52cm (L) giving a total available floor space of 1664cm ²
6	Cages were solid-bottomed and contained both sawdust and a large cardboard tube
7	The cages were kept in a temperature- (21 \pm 1 _o C) and humidity- (55 \pm 10%)
8	controlled environment under a reversed 12:12 hour light:dark cycle (lights off at
9	10:00). Animals had ad libitum access to food (PCD Mod C; Special Diet Services
10	Witham, UK) and water, except during testing. All testing was conducted during the
11	dark phase under red-light.
12	
13	Apparatus
14	A runway, constructed from wood, measuring 185 cm long, with a 26 x 24 cm reward
15	area at one end, was used (Figure 1). The first 39 cm formed the start box, separated
16	from the main runway by a plastic door. The walls of the entire apparatus were 19 cm
17	high and were covered with a clear Perspex lid to prevent the rats from climbing out.
18	
19	Figure 1 about here
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21	Objects
22	One object was used to train the rats and collect baseline data. This was a plastic ball-
23	shaped object (36cm circumference), with holes around the sides (5cm in diameter
24	see Figure 2a). The two sets of objects used in testing consisted of one top ranked and

- 1 one bottom ranked object, as determined by previously published experimental data
- 2 (Williams, et al., 2008). Two sets of objects were prepared for this study:
- a) Lego® set: objects made from Lego® (Figure 2b). This set consisted of a large
- 4 plastic block, made from Lego® bricks (9.5x6x5cm), which had been the top ranked
- 5 object in the previous experiment, and an individual Lego® brick (4x2x2cm) covered
- 6 in soft synthetic fur, which had been the bottom ranked object (Williams et al., 2008).
- 7 b) EE set: objects typically chosen as enrichment objects for rat home cages (Figure
- 8 2c). The "EE" top ranked object was a plastic house made from flat plastic
- 9 connectable shapes (14cm wide, 19 cm long at longest point and 11cm high at tallest
- 10 point) and the EE bottom ranked object was a small cardboard tube (measuring
- 11 12.5cm long, 5.5cm diameter cm: Hanmer, 2008).

13 ---Figure 2 about here---

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- All of the apparatus was cleaned using 50% ethanol solution between each rat's trials,
- 16 except for the small fur covered object. Five of these were made to allow each rat to
- have one each, at the end of the day these objects were sprayed with a deodoriser
- 18 (Shaw's Pet Stain and Odour Eliminator) in order to eliminate olfactory cues.

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Familiarisation & Training

- 21 Before testing, all of the rats were habituated to, and trained in using, the runway. A
- 22 plastic ball was used as the object at the end of the runway, only in the habituation
- and training phases, as an incentive to motivate the rats to run along the runway.

- 1 Familiarisation: Initially, pairs of rats were exposed to the runway and plastic ball, for
- 2 five minutes, to minimise stress and encourage them to investigate the apparatus. This
- 3 was repeated on two consecutive days. On the following three days, individual rats
- 4 were exposed to the runway and plastic ball for five minutes individually.
- 5 Training: Following familiarisation to the apparatus, on each of the next five days, the
- 6 rats were individually placed in the start box, the door opened and timing started. The
- 7 rat then had 30 seconds in which to leave the start box, run the length of the runway,
- 8 and interact with the object for 5 seconds. If they were successful, then they were
- 9 placed back in the start box for another trial. They were unsuccessful if they failed to
- leave the start box, failed to reach the object within 30 seconds of the start box door
- being opened or failed to interact with the object for at least two seconds once in the
- 12 reward box. Three unsuccessful trials in a row resulted in termination of the session
- for that rat. Eighteen rats were trained on this procedure, and the data was inspected at
- the end of the 5 day training period. Only rats that had run for at least three successful
- trials in each of the five training session were included in the testing phase (10 rats).

Runway Testing

- 17 Two replications of testing were carried out with each of the two pairs of test objects.
- 18 Each replication consisted of counterbalanced days in which the rats either saw the
- 19 familiar plastic ball (baseline), or the novel top ranked, or the novel bottom ranked
- 20 object, in the reward box. The baseline days, using the familiar plastic ball, were
- 21 included to check that the rats were maintaining their performance levels as testing
- progressed over the week. These confirmed that no rat fell below the minimum three
- runs to the plastic ball on each day. Thus, reduction in motivation in the runway
- 24 cannot explain differences in performance for the test objects. The criteria used to
- determine whether a trial was passed or failed on each attempt was the same as those

used in training. In each session, the rat was placed in the start box, with an object in the reward box. The door to the start box was lifted and timing started. The rat then had 30 seconds in which to reach and interact with the object. The rats were allowed to interact with the object for a maximum of 5 seconds upon reaching it before being placed back in the start box. This process continued until the rats had failed three trials in succession. The time taken to reach the reward object, the number of trials successfully completed in each testing session and the time they spent interacting with

Ethical note

11 All testing was performed in accordance with the United Kingdom Animals

12 (Scientific Procedures) Act of 1986.

the object were recorded.

Statistical Analysis

After the two replications had been completed with the same objects, a one-way repeated measures ANOVA was conducted on data from each set of objects. This was to determine whether there were significant differences in the data collected in each replication in order to collapse the data across replications.

Two-way repeated measures ANOVAs were conducted for the number of runs, the time it took to reach the objects, and the time spent interacting with the objects. Trial 1 was removed from the analysis of both the number of runs made and the time taken to reach objects, since on trial 1 the rats could not have known what object they were running towards. Repeated measures Bonferroni t-tests were conducted to explore significant interactions.

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Results

- 3 The replications with each set of objects did not differ significantly for either the
- 4 Lego® objects $(F_{(1,9)}=3.714, P=0.086)$, or the EE objects $(F_{(1,9)}=0.938, P=0.358)$ and
- 5 there were no interactions. Therefore, it was assumed that these results show a
- 6 genuinely replicable effect, and that the methodology is reliable. The results were
- 7 collapsed across replications for the remaining analyses.

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Number of runs to objects

- 10 Figure 3 shows the number of times the rats' ran to reach the objects. There was a
- significant interaction between object type and ranking ($F_{(1,9)}$ =5.18, P=0.049). The
- rats ran significantly more times to the top ranked objects ($F_{(1.9)}$ =24.87, P=0.001)
- compared with the bottom ranked objects. Post-hoc t-tests revealed that there were
- significant differences between top and bottom ranked objects for both object types
- 15 (Lego®: $t_{(9)}$ =4.56, P=0.001; EE: $t_{(9)}$ =3.64, P=0.005). In addition to the main effect of
- ranking, there was also a simple effect for the bottom ranked objects. There was a
- significant difference between the Lego® and EE bottom ranked objects ($t_{(9)}=2.449$,
- 18 P=0.037) showing that, for bottom ranked objects, the rats ran significantly more
- 19 times to reach the EE object than the Lego® object. There was no significant
- 20 difference in the number of times rats' ran to the Lego® and EE objects overall.

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22 --- Figure 3 about here ---

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Time taken to reach objects

1 2	Table 1 about here
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4	This data was analysed twice; firstly excluding trials in which the rats failed to reach
5	or interact with the objects and therefore did not record a time (Table 1: columns 2 &
6	3) and secondly using a technique employed by other researchers (e.g. Nencini et al,
7	1991; Ikemoto & Panksepp, 1996) in which the maximum time of 30 seconds was
8	recorded for failed trials (Table 1: columns 4 & 5).
9	
10	Removing failed trials from the data resulted in excluding one rat. Table 1 (columns 2
11	& 3) show the means (with SEM) for the time taken to reach the objects using this
12	method of analysis. A two-way repeated measures ANOVA was conducted on the
13	data from the remaining nine rats, to determine whether rats' ran faster to reach the
14	preferred objects compared with the non-preferred objects and also whether they
15	differed between object sets. There were no significant differences between top and
16	bottom ranked objects for either object set with failed trials removed. Table 1
17	(columns 4 & 5) shows the mean time taken to reach the objects when a maximum
18	time of 30 seconds was recorded in place of a failed trial. The ANOVA showed that
19	the rats' took significantly longer to reach the bottom ranked objects compared with
20	the top ranked objects using this method of analysis, $(F_{(1,9)}=7.39, P=0.024)$. No other
21	significant effects were found.
22	
1 2	Time enout interesting with abjects
23	Time spent interacting with objects
24	Figure 4 shows the average time the rats spent interacting with the objects once they

had reached them. A two-way repeated measures ANOVA revealed there was a

significant interaction between ranking and object set $(F_{(1,9)}=27.60, P<0.001)$, a significant difference between the Lego® and EE objects (F_(1,9)=26.12, P<0.001), and also a significant difference between the top and bottom ranked objects $(F_{(1,9)}=37.88,$ P<0.001). Subsequent t-tests revealed that the interaction resulted from the rats spending less time interacting with the bottom ranked Lego® object compared to all other objects. There was a significant difference between the Lego® and EE bottom ranked objects ($t_{(9)}$ =5.19, P=0.001), with rats spending longer with the EE bottom ranked object than with the Lego® bottom ranked object. There was a significant difference between the Lego® top and bottom ranked objects ($t_{(9)}$ =6.16, P<0.001), with rats spending longer with the top ranked object as predicted.

--- Figure 4 about here ---

Discussion- Study 1

Overall, this study demonstrated that the active runway paradigm is a successful tool for assessing rats' motivational preferences. Clear differences were found between top and bottom ranked object types for the number of times the rats ran, the time they took to reach the object (when 30 seconds was recorded for failed trials), and the time they spent interacting with the objects (Lego® only). However, ceiling effects prevented differences in interaction time for the EE objects from reaching significance. This could be prevented by allowing rats to interact with the objects for longer (10 seconds).

Additionally, our results show conflicting evidence regarding the usefulness of the measure of time taken to reach the object, depending on how it is calculated. Using

our original data set, with one rat excluded for failing to reach the EE objects, we found no significant differences in the run times to any of the objects. This would suggest that this measure is not a reliable way of differentiating between motivational preferences because the rats run along the runway at the same speed regardless of the object placed in the reward box. Alternatively, when we adapted our design, to include a recording of the maximum 30 seconds for failed trials, as has been previously used in runway methodologies (e.g. Nencini et al, 1991; Ikemoto & Panksepp, 1996), we did find a significant difference in the time rats took to reach top and bottom ranked objects. However, it is possible that this results from recording 30 seconds as the run time for failed trials. Since there were more of these for bottom ranked objects this analysis might result in artificially significant data.

There were also differences in time spent interacting when the bottom ranked object types were compare, but not between top ranked objects. This suggests that while the top ranked Lego® and EE objects seemed to provide the rats with the opportunity to express highly motivated behaviours to an equal extent (since no differences were found between these objects), the non-preferred objects permitted different behaviours. The rats' ran more times and spent longer interacting with the bottom ranked EE object when compared to the bottom ranked Lego® object. One potential explanation for this is that the bottom ranked Lego® object was a small block covered in synthetic fur (a non-preferred texture), which only allows picking up and chewing behaviours, whereas the EE cardboard tube also allowed the rat to put its head inside the object and rear up against it. Since the EE and Lego® objects also differed considerably in other factors (ie. texture, size), these might also have contributed to the differences found between bottom ranked objects.

2 A second study was conducted to determine whether ceiling effects in interaction 3 time could be eliminated by increasing the interaction time from 5 s to 10 s. In 4 addition, a range of objects was used to determine whether the behaviours afforded 5 by the objects, or the physical properties of the objects were more influential in 6 determining object ranking in the runway. To achieve this, two large objects, and two 7 small objects, of identical construction were covererd with either polyester or fur. By 8 using Lego® objects that had been covered in different fabrics, we were able to 9 consider size of object (and therefore behaviour performed) separately from the 10 physical properties of the objects.

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The rats used in the second study were younger than those used in the first experiment. We therefore might expect the rats in this study to demonstrate higher overall levels of motivation for novel objects. However, predictions are based on within subject comparisons for each study, and so differences in overall levels of motivation between were not considered problematic when testing these hypotheses.

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Methodology for Study 2

Animals

- 20 Adult male Lister-Hooded rats (N=14), weighing 350-450g, were group housed in
- standard cages under standard conditions (n = 4 rats per cage). Cage and housing
- 22 environment were maintained as detailed for Study 1.

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Apparatus

25 Apparatus used was identical to that described for Study 1.

1 2 **Objects** 3 ---Figure 5 about here---4 Figure 5 shows the objects tested in this study. In order to test objects with 5 intermediate preference, we used four objects that had been ranked with a range of 6 preferences using open field preference tests from previous work (Hanmer, 2008). 7 We thus used a large polyester covered block made from Lego® bricks (9.5 x 6 x 5 8 cm), a large fur covered block made from Lego® bricks of the same dimensions, an 9 individual Lego® brick covered in polyester (4 x 2 x 2 cm), and a small individual 10 Lego® brick covered in fur of the same dimensions. Of these, the large polyester 11 covered object had been top ranked, the small fur covered object was bottom ranked, 12 and the other two objects had intermediate ranks. 13 14 **Familiarisation, Training and Testing** 15 The experiment was conducted as in Study 1. All animals tested met the criterion for inclusion outlined in Study 1. 16 17 18 **Statistical Analysis** 19 Data was analysed using a one-way repeated measures ANOVA for number of runs, 20 time taken to reach the objects, and time spent interacting with the objects. As above, 21 Trial 1 was removed from the analyses. When analysing the time taken to run to 22 objects, failed trials were replaced with a maximum time of 30 s. Repeated measures 23 Bonferonni corrected t-tests were conducted to explore significant interactions. 24

Results

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2 ---Figure 6 about here---3 Figure 6 shows the number of runs (Figure 6a), the time taken to reach the objects 4 (Figure 6b) and the time spent interacting (Figure 6c) for the four objects used in 5 study 2. A one-way repeated measures ANOVA with object as factor, and number of 6 runs, time taken to reach objects, and interaction time as measures was conducted. 7 There was a main effect of object for each of the measures (Number of Runs: $F_{1.16,15.08}$ 8 = 17.59, p = 0.001; Run Time: $F_{1.6, 20.81} = 17.87$, p < 0.0001; Interaction Time: $F_{3,39} =$ 9 14.10, p < 0.0001). 10 11 Post-hoc tests for Number of Runs demonstrated that rats ran more often to the large 12 polyester object than to any other object (p < 0.012). No other comparisons reached 13 significance. When objects were entered in the same rank order as found in our 14 previous work (Hanmer, 2008), there was a significant linear trend ($F_{1,13} = 23.22$, p < 15 0.0001) with rats running more often to the top ranked objects than bottom ranked 16 objects. For time taken to run to objects, there were significant differences between 17 the large polyester object and all other objects (p < 0.03), and also between the small 18 polyester object and the small fur object (p = 0.021), and finally there was a marginal 19 trend for significance between the large and small fur objects (p = 0.051). As with 20 number of runs, there was a significant linear trend in time taken to run to objects 21 $(F_{1,13} = 43.57, p < 0.0001)$. Rats took longer to run for objects of lower ranking. 22 23 For interaction time, post-hoc analysis demonstrated significant differences between 24 the small fur object and all other objects (p < 0.013). No other comparisons reached

- significance. There was, however, a significant linear trend $(F_{1,13} = 49.57, p < 0.0001)$
- with rats interacting for longer with top ranked objects.

4 Discussion- Study 2

- 5 In confirmation of our results in Study 1, this study demonstrated that when objects
- 6 were ranked according to time spent interacting with them in the open field (Hanmer,
- 7 2008), there were significant linear trends with rats running more often and more
- 8 quickly to the higher ranked objects.

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- 10 In the second study, we calculated the time taken to run to the objects by replacing
- failed run time with a maximum 30 s (Nencini et al, 1991; Ikemoto & Panksepp,
- 12 1996). As in Study 1, this resulted in significant differences between our top and
- bottom ranked objects but also between intermediately ranked objects. There was a
- 14 significant linear trend in time taken to reach the objects when these were ranked
- according to time spent in a previous study (Hanmer, 2008). This suggests that time
- taken to reach the objects can reflect subtle differences in motivation when calculated
- in this manner.

18

- 19 In Study 2, we reported differences between top and bottom ranked objects when rats
- were allowed a longer interaction time (10 s in Study 2 compared with 5 s in Study 1).
- 21 We also demonstrated a highly significant linear trend, with rats interacting for
- 22 progressively shorter times according to a descending ranking based on previous
- 23 interaction times in the open field (Hanmer, 2008). Importantly, no ceiling effects
- 24 were found when using this length of interaction time.

1 Across all measures, the highest ranked object was the large polyester block, and the

lowest ranked object was the small fur block. The large fur block and the small

polyester block were intermediate in ranking. Thus, neither the size of the object nor

4 the fabric in which it was covered independently predicted ranking, suggesting that

rats are influenced in their preference decisions by both the behaviours they can

perform with an object, and the physical properties of that object.

preferences.

Overall Discussion

These results reveal that the runway paradigm is a suitable methodology to measure the strength of rats' motivational preferences for enrichment objects. The rats were more highly motivated to reach objects that they had previously chosen to spend more time with compared with objects with which they had previously spent little time (Hanmer, 2008; Williams et al., 2008), as shown by the number of times they ran to gain access to, the time taken to run and the interaction times for the top ranked versus bottom ranked objects in Study 1, and the linear trends in these measures in Study 2. This effect was found to be consistent across replications and with different types of object, so the runway paradigm is a reliable method for comparing motivational

All of the objects used in this experiment had previously been ranked by assessing the amount of time the rats spent interacting with them (Hanmer, 2008; Williams et al, 2008). The current study has reinforced this object ranking by demonstrating that the same ranking occurs when using a different measure of relative motivational preference. Specifically, across Studies 1 and 2, we have demonstrated that the number of runs, the method used previously to calculate time taken to reach the

- objects in which failed trials are replaced by a maximum time (Nencini et al, 1991;
- 2 Ikemoto & Panksepp, 1996) is capable of differentiating the relative ranking of
- 3 objects, and a 10 s interaction time when rats reach the object at the end of the runway
- 4 are suitable parameters to discriminate differences in object ranking.

- 6 This active runway methodology could also be used to investigate reward processes
- 7 associated with motivational preference. This is because it provides a means to
- 8 investigate both appetitive and consummatory aspects of the reward process. Using
- 9 this paradigm, the number of runs and the time taken to reach the object are both
- 10 measures that can relate to how much the rat 'wants' the object (appetitive
- 11 component), whereas the time the rat spends interacting with the object may relate to
- 12 how much it 'likes' the object (consummatory component). The measurement of both
- phases of the reward process cannot be made with other measures of preference such
- as interaction times since these measure only the consummatory phase. Thus, this
- 15 methodology provides a more comprehensive assessment of motivation.
- 16 Pharmaceutical compounds are known to be selectively active in different
- 17 components of the reward processes (e.g. opioid and cannabinoid agonists and
- antagonists). By administering these compounds when rats are running for previously-
- 19 ranked objects, the activity of different components of the reward system in these
- 20 rankings could be investigated. This would increase our knowledge of the role of the
- 21 reward system in forming motivational preferences for enrichment objects.

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Conclusion

- 24 In conclusion, the runway paradigm is a quick and simple method for collecting
- 25 evidence for motivational preferences for EE objects by determining with which

- 1 objects rats are most motivated to interact. It might be concluded that objects that
- 2 produce higher motivational preferences should be added to the caged environment to
- 3 improve welfare in laboratory rats. This paradigm could also provide a means to
- 4 investigate reward processes related to object preference.

6

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1 **Figure Legends** 2 3 **Figure 1.** The runway apparatus, with dimensions. 4 5 **Figure 2.** Objects used in study 1. a) The plastic ball used to train the rats on using the runway and collect baseline data. 6 7 b) The Lego® objects (Left – large plastic, right – small synthetic fur). 8 c) The EE objects (Left – plastic house-shaped shelter, right – cardboard tube). 9 10 Figure 3. The average number of times the rats' ran to reach the objects. Rats ran 11 significantly more times to reach the top ranked object than the bottom ranked object 12 for both sets. The asterisks indicate that the difference between objects was significant 13 with p < 0.005. 14 15 Figure 4. The average time the rats' spent interacting with the objects. Rats spent 16 significantly less time interacting with the bottom ranked Lego® object compared to 17 all other objects. 18 19 **Figure 5.** Objects used in study 2. The large and small fur-covered blocks are shown 20 on the left, and the large and small polyester-covered blocks are shown on the right. 21 22 Figure 6. Results for study 2. 23 a) The average number of times the rats' ran to reach each of the objects. Rats ran 24 significantly more times to reach the large polyester-covered object more than any 25 other object. 26 b) The time taken to reach the objects. Rats ran significantly faster to the large 27 polyester covered object than any other object, and also ran significantly faster to 28 the small polyester covered object than the small fur covered object. 29 c) The time spent interacting with the objects. Rats spent significantly less time

interacting with the small fur covered object than any other object.

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- 1 Table 1 Mean Run times (with standard error of the mean) for top and bottom
- 2 ranked Lego® and EE objects calculated with failed runs excluded (columns 2 & 3)
- 3 or with failed run times set to 30 secs (columns 4 & 5).

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	Run times (fail	ed runs excluded)	Run times (failed runs= 30 s)		
	Top Ranked	Bottom Ranked	Top Ranked	Bottom Ranked	
Lego®	13.00 (1.76)	12.73 (1.04)	13.40 (2.01)	17.01 (1.63)	
EE	11.38 (0.92)	12.87 (1.18)	13.25 (2.03)	16.79 (1.72)	