The Old School, Brewhouse Hill, Wheathampstead,
Hertfordshire AL4 8AN, UK

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Housing conditions affect self-administration of anxiolytic by laboratory mice

CM Sherwin*† and IAS Olsson†

* Centre for Behavioural Biology, Department of Clinical Veterinary Science, University of Bristol, Bristol BS40 5DU, UK
† Institute for Molecular and Cell Biology (IBMC), Rua Campo Alegre 823, 4150-180 Porto, Portugal
* Correspondence: chris.sherwin@bristol.ac.uk

Abstract

Tests of emotionality conducted outside the home-cage show that rodents from standard laboratory housing are more anxious than animals from enriched housing; however, it is not known if this also indicates increased anxiety within the home-cage. We used a novel method, recording the self-administration of a psychoactive anxiolytic, to examine home-cage anxiety levels of laboratory mice (three per cage) in Standard (n = 10 cages), Unpredictable (n = 10 cages) and Enriched (n = 6 cages) housing. The mice were given a choice of drinking either non-drugged water or a solution of the benzodiazepine Midazolam. There was a significant effect of housing on the proportion of total fluid consumed from the bottle containing Midazolam solution (P = 0.02). Mice from Standard and Unpredictable cages drank a greater proportion than mice from Enriched cages. This indicates that mice from the Standard and Unpredictable laboratory caging may have been experiencing greater anxiety than mice from the Enriched cages. There was also a significant effect of bottle position (P = 0.002). Mice from the Standard and Unpredictable cages drank a greater proportion of total fluid from the bottle containing Midazolam solution when this was toward the rear of the cage rack, ie in a location that was less susceptible to extraneous disturbance. Monitoring self-administration of psychoactive drugs as a method of welfare assessment could be applied to a wide variety of housing conditions, husbandry practices, or experimental procedures that putatively induce negative mental states. The major finding is that standard cages for laboratory rodents may induce greater anxiety than enriched cages. This is discussed in terms of animal welfare and the validity of data from animals housed in minimalistic environments.

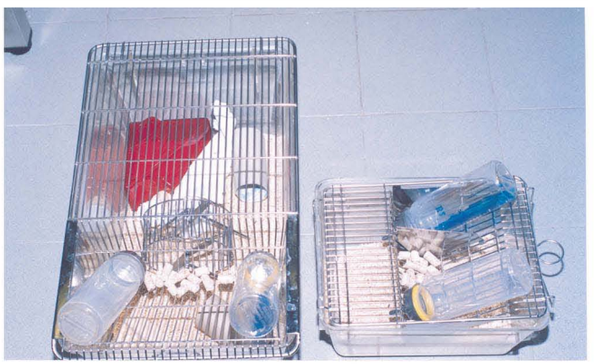
Keywords: *animal welfare, anxiolytic, enrichment, mice, self-administration*

Introduction

Standard housing for laboratory rodents is highly minimalistic with respect to both the quantity and quality of the environment it provides. Typically, cage dimensions are small (less than 4–5 body-lengths of the animal) and the cage contains nothing other than food, water, litter material and perhaps cage-mates. It is widely argued that standard housing and husbandry compromises animal welfare (eg Jennings *et al* 1998; Würbel 2001; Olsson & Dahlborn 2002; Sherwin 2002; Olsson *et al* 2003). One consequence of standard housing can be an influence on the mental states of the animals. For example, in tests of emotionality (eg elevated plus maze, open field, shuttle box), rodents from standard laboratory housing behave in a manner indicative of being more anxious or fearful than animals from enriched housing systems (Chamove 1989; van de Weerd *et al* 1994; Prior & Sachser 1995; Chapillon *et al* 1999; Roy *et al* 2001; Schrijver *et al* 2002). Although these studies clearly demonstrated the influence of housing on mental states, these tests were conducted on animals outside their home-cage. It is important for two reasons to examine the possibility that housing influences mental states within the home-cage. First, if standard laboratory housing causes animals to become anxious within the home-cage, this indicates that such housing systems might routinely induce a negative mental state that compromises welfare; this might persist for a large proportion of the animals' lives. Second, anxiety or fearfulness can substantially influence a range of physiological and behavioural responses (eg Broom & Johnson 1993; Moberg & Mench 2000). This would, in many circumstances, reduce the validity of the research for which the animals are being housed and thus negate the reason for their being housed in the first instance.

But, how can we objectively assess the negative mental states of animals in their home-cage? One possibility is to allow animals the opportunity to self-administer drugs believed to alleviate negative mental states. Self-administration of drugs as a tool to assess animal welfare has been reported previously. Colpaert *et al* (1980, 1982) used a simple drug choice procedure to show that rats suffering from painful adjuvant-induced arthritis consumed more of an analgesic than did non-arthritic animals. Pickup *et al* (1997) and Danbury *et al* (2000) examined self-administration of analgesic by broiler chickens. These birds often experience leg problems associated with breeding, growth and husbandry, and can develop highly abnormal gaits or

Figure 1



The enriched and standard cages used to measure the effect of housing on self-administration of anxiolytic by laboratory mice.

become completely unable to walk. Lame birds selected significantly more drugged food than non-lame birds and, as the severity of the lameness increased, lame birds consumed a significantly greater proportion of the drugged food (Danbury *et al* 2000). This was interpreted as indicating that lame birds were experiencing the negative mental state of 'pain' and behaved appropriately in the home-pen to alleviate this. The present study followed a similar line of reasoning. We hypothesised that if mice were housed in conditions that caused them to be anxious or fearful, they would self-administer more anxiolytic than animals housed under conditions that are presumed to induce less anxiety.

Midazolam is a benzodiazepine that has a clear anxiolytic effect on rodents. Administration of this drug reduces the rate of ultrasonic distress vocalisations made by mouse pups when separated from their mother and littermates (Fish *et al* 2000), reduces aggression between mice in staged intruder contests without increasing immobility (Lopez & Navarro 1999), decreases anxiety in an elevated plus maze (Nunes-de-Souza *et al* 2000; Rosa *et al* 2000; Bertoglio & Carobrez 2002; Cruz-Morales *et al* 2002), reduces reactivity to predator odours (Dielenberg & McGregor 2001; McGregor *et al* 2002), abolishes the avoidance of an aversively conditioned environment (Harris & Westbrook 1994), and induces preferences for an environment (Pain *et al* 1997). The aim of the present study, therefore, was to determine whether mice in a range of housing systems would differentially self-administer an anxiolytic, Midazolam, indicating different levels of anxiety related to housing conditions.

Materials and methods

Animals and housing

The study was conducted at The Institute for Molecular and Cell Biology, Porto, Portugal. Seventy-eight C57Bl/6J female mice were obtained from a commercial breeder (Harlan Iberica) at three weeks of age. After one week of quarantine, the mice were placed into groups of three and randomly assigned to one of three housing treatments (Figure 1). These were Standard (animals housed in standard cages with standard husbandry, n = 10 cages), Unpredictable (animals housed in standard cages with unpredictable events, n = 10 cages) and Enriched (animals housed in larger cages with an enriched environment, n = 6 cages). The standard cages were standard wire-topped Makrolon II cages (265 mm × 205 mm, 140 mm high). The unpredictable events comprised changing the position of the cage in the rack, or tilting the cage by 7° along the shorter side of the cage either inwards towards the wall or outwards towards the room. This was done two or three times each week for varying periods with a maximum of two days, starting at a randomly selected time between 0900h and 1500h, and was the same event for all cages of the Unpredictable treatment. The enriched cages were standard wire-topped Makrolon III cages (265 mm × 410 mm, 175 mm high) containing a translucent red PVC nest box (MouseHouse; Tecniplast), a 115 mm diameter metal running-wheel, two 100 mm long, 45 mm diameter cardboard tubes, and two sheets (240 mm × 220 mm) of absorbent paper (Renova SA, Torres Novas, Portugal) for nesting material which were replaced once each week.

For all cages, the sawdust bedding material (Harlan Iberica) was changed once each week by transferring the mice and the cage top to a clean cage bottom with fresh bedding. Mice in all cages had *ad libitum* access to standard rodent chow (Harlan Iberica). The room was on a 12h light : 12h dark cycle with the light phase starting at 0500h. Temperature and relative humidity were maintained at 21–23ºC and 70–100%.

Anxiolytic and procedure

The top of each cage contained two transparent water bottles, one marked with a blue stripe down the length of the bottle and one unmarked. Each cage had two drinker positions, one to the rear or inside of the rack ('In') and one to the front or outside of the rack ('Out'). Within each cage, the striped bottle was always placed in the same position; however, the bottle positions were balanced so that half of the cages of each treatment had the striped bottle in the 'In' position and half in the 'Out' position. The striped bottle was always placed with the stripe downwards so that it was visible to the mice.

The study comprised four phases:

(1) Control (10 days): both bottles contained only autoclaved tap water.

(2) Self-administration, 0.02 mg ml^{-1} (28 days): Midazolam solution (0.02 mg ml^{-1}) was available in the striped bottle and autoclaved tap water in the non-striped bottle.

(3) Midazolam only (6 days): on alternate days either only Midazolam solution (0.08 mg ml^{-1}) in the striped bottle was available, or only autoclaved tap water in the non-striped bottle.

(4) Self-administration, 0.08 mg ml^{-1} (6 days): Midazolam solution (0.08 mg ml^{-1}) was available in the striped bottle and autoclaved tap water in the non-striped bottle.

Midazolam solution was prepared by dissolving tablets (Dormicum®, Roche Farmacêutica Quimica, Lda) in

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autoclaved tap water. All bottles were weighed and refilled with fresh autoclaved tap water or Midazolam solution twice each week during Phases 1 and 2, and every two days during Phases 3 and 4.

Statistical analysis

The data were normally distributed (Anderson-Darling Normality Test run on MiniTab; A2 = 0.349, $P = 0.468$) and were therefore analysed primarily by ANOVA with differences between means tested by Fisher's Protected Least Significant Difference when appropriate. Where data are expressed as proportions, the raw data were first subjected to arcsine, square-root transformation. Values in figures are non-transformed. The data from Phase 4 were additionally subjected to a single-group *t*-test to determine whether mean consumption of anxiolytic solution for each housing treatment and bottle position differed significantly from 0.5 of the total consumption of fluid.

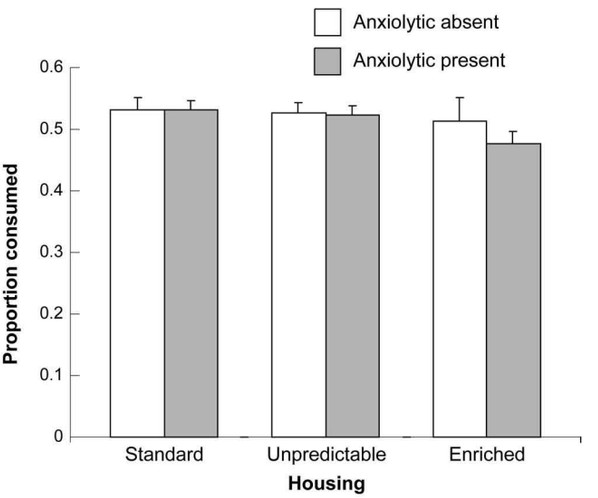
Results

Comparing only data from the Control and Self-administration 0.02 mg ml^{-1} phases, two-way ANOVA showed that there was no significant effect of housing treatment, Midazolam availability or interaction ($F_{2,1,2} = 1.4$, 0.6, 0.4; $P = 0.23$, 0.4, 0.65) on the proportion of total fluid consumed from the striped bottle (Figure 2).

During the Midazolam-only phase, substituting Midazolam solution for tap-water increased total fluid consumption (Figure 3). Two-way ANOVA showed that there was no significant effect of housing treatment on the total volume of fluid consumed, but there was a significant effect of Midazolam availability and interaction ($F_{2,1,2} = 2.3$, 89.3, 3.4; $P = 0.1$, 0.0001, 0.03).

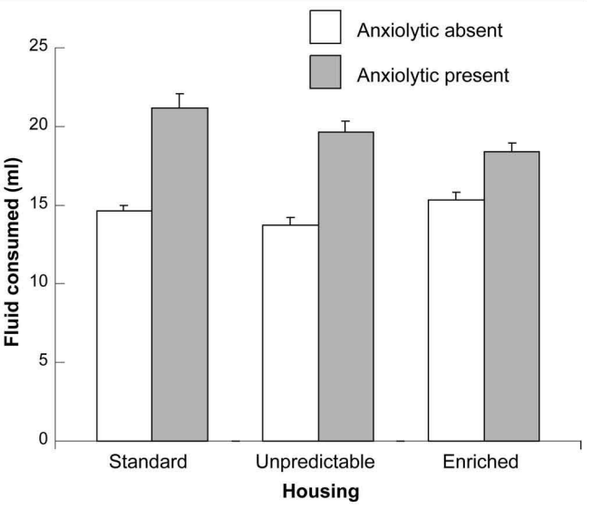
During Phase 4, the mice were able to self-administer Midazolam solution of a greater concentration than during Phase 2, ie 0.08 mg ml^{-1}. Two-way ANOVA showed that during Phase 4, overall there was a significant effect of housing treatment, bottle position and interaction on the proportion of total fluid consumed from the bottle containing Midazolam solution ($F_{2,1,2} = 4.1$, 9.9, 7.6; $P = 0.02$, 0.002, 0.01). Mice from Standard housing drank a significantly greater proportion of anxiolytic than mice from the Enriched housing ($P = 0.007$), and tended to drink a greater proportion than mice from the Unpredictable housing ($P = 0.06$). Because of the significant interaction, the data were re-analysed separately for each housing treatment. One-way ANOVA showed that bottle position had a significant effect on the proportion of total fluid consumed from the bottle containing Midazolam solution for the Standard ($F_1 = 24.9$; $P < 0.0001$) and Unpredictable ($F_1 = 20.8$; $P < 0.0001$) treatments, but not the Enriched treatment ($F_1 = 0.9$; $P = 0.34$). Single-group *t*-tests showed that the mean consumption of Midazolam solution was significantly greater than 0.5 of the total fluid consumption for mice in the Standard housing with the Midazolam bottle in the 'In' position ($t = 10.4$; $df = 14$; $P < 0.0001$) and mice in the Unpredictable housing with the Midazolam in the 'In' position ($t = 4.67$; $df = 14$; $P = 0.0004$). None of the other proportions was significantly different from 0.5 (Figure 4).

Figure 2



The effect of housing conditions on the proportion of total fluid consumed from a marked bottle containing either tap water or Midazolam anxiolytic solution (0.02 mg ml^{-1}). Values represent means + SEM.

Figure 3



The effect of housing conditions on the amount of total fluid consumed when only tap water or Midazolam anxiolytic solution (0.08 mg ml^{-1}) was available. Values represent means + SEM.

Discussion

The major finding of this study is that when mice were able to drink Midazolam (0.08 mg ml^{-1}) solution, overall, mice in standard laboratory housing drank more of the solution than did mice in enriched housing (Figure 4). Midazolam is widely considered to have psychoactive anxiolytic properties for rodents, and the increased consumption supports the hypothesis that mice in standard laboratory housing were

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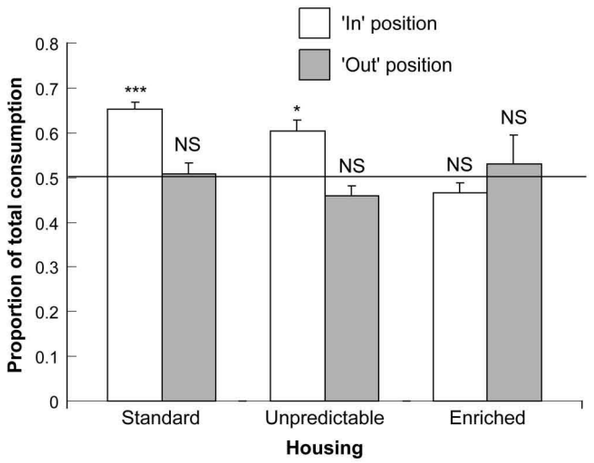
Figure 4



The effect of the interaction between housing conditions and bottle position on the proportion of total fluid consumed from a bottle containing Midazolam anxiolytic solution (0.08 mg ml^{-1}). Values represent means + SEM. ***Significantly different from 0.5 ($P < 0.001$); *significantly different from 0.5 ($P < 0.05$); NS, not significantly different from 0.5 ($P > 0.05$).

more anxious than mice in enriched conditions. Previous studies have shown that the environment can influence self-administration of anxiolytic. Adams and Oldham (1996) and Rockman and Gibson (1992) housed rats in isolation or in groups in standard cages or enriched environments, and then tested consumption of alcohol after re-allocating the animals to other housing conditions. These studies revealed conflicting evidence of the effects of environment on self-administration of this anxiolytic; however, it should be noted that these studies measured the effect of prior housing conditions on self-administration in a subsequent environment. The present study is the first in which self-administration of an anxiolytic was measured in the home-cage of animals housed in standard, unpredictable, or enriched environments. It has been shown that enriched housing reduces the anxiety levels of animals in behavioural tests conducted outside the cage (Chamove 1989; van de Weerd *et al* 1994; Prior & Sachser 1995; Chapillon *et al* 1999; Roy *et al* 2001; Schrijver *et al* 2002). The present results indicate that enrichment also reduces anxiety within the home-cage. Furthermore, these results can be interpreted as indicating that mice placed into standard laboratory conditions might be exposed routinely to an environment that induces heightened anxiety and thereby compromises animal welfare, regardless of any other procedure that might be performed on the animal.

It is unlikely that taste or other hedonistic properties of the drug caused the differential self-administration during Phase 4. Figure 3 shows that the availability of anxiolytic solution increased the total volume of fluid intake above that of non-drugged water, indicating that the anxiolytic was reinforcing in all three housing systems under some conditions; however, this might easily have been a novelty effect. When the data for Phase 4 were analysed separately for each housing treatment and bottle position (ie ignoring the overall effects shown by ANOVA), most treatment groups drank equal volumes of fluid from both bottles, indicating that they did not differentiate between the anxiolytic solution and non-drugged water (Figure 4). If the Midazolam had hedonistic properties, it might have been expected that the mice would differentially select the anxiolytic solution, irrespective of housing treatment.

There was no significant difference in the consumption of fluid from the striped bottle when this contained tap water during Phase 1 and when it contained 0.02 mg ml^{-1} Midazolam solution during Phase 2 (Figure 2). This was probably due to the mice ingesting insufficient Midazolam to experience the psychoactive effects. We used a concentration of 0.02 mg ml^{-1} based on the calculation of a mouse ingesting 3 mg kg^{-1} in 24 h, as has been widely used previously (Fish *et al* 2000; Pakulska & Czarnecka 2001; Sannerud & Ator 1995; Soderpalm & Hansen 1998; Sun *et al* 2000). However, in these cited studies, the 3 mg kg^{-1} was given as a single dose. In the present study, it is highly unlikely that the mice would have obtained their entire 24 h water requirements in a single drinking bout; therefore, an amount perhaps considerably lower than 3 mg kg^{-1} would have been ingested in any one drinking bout. This would have reduced the anxiolytic effect and negated the opportunity to learn to associate the striped bottle with the psychoactive properties of the drug. Therefore, during the Midazolam-only phase, the concentration of Midazolam was increased to 0.08 mg ml^{-1} and the solution made available on alternate days when the bottle containing tap water was also withdrawn. This ensured that the mice had the opportunity to drink the solution and learn to associate consumption of fluid from the striped bottle with its anxiolytic effects, prior to the second self-administration phase.

The significant effect of bottle position on consumption of Midazolam shows that mice from the Standard and Unpredictable housing drank more Midazolam when this bottle was in the 'In' position. This position was towards the rear of the rack and represents a location where the mice would be less susceptible to extraneous disturbance. This relates to another animal housing welfare issue. Water bottles are typically situated at the front of the cage to facilitate changing, but the present results indicate that for mice in standard housing, the preferred bottle position is at the back of the cage.

Animal welfare implications

This study has three major implications for animal welfare. First, it describes a method that might be used to assess the welfare of animals in a wide variety of housing conditions, husbandry practices, or experimental procedures that could induce anxiety. The method could be extended to examine the possibility that other negative mental states (eg pain, depression, frustration) are being experienced by animals in other circumstances, which in turn would allow us to investigate potential methods of alleviation. Second, the finding that mice in standard laboratory housing consumed more

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anxiolytic solution than animals in enriched cages adds to the weight of evidence showing that standard laboratory cages can compromise animal welfare. If this finding can be extrapolated to other laboratories and standard caging systems, it indicates that mice are routinely being placed into an environment that evokes heightened anxiety, regardless of any other procedure to which the animal might be subjected. Third, these findings relate to the external validity of research using mice from standard cages. Animal research depends on the assumption that responses of animals from standard conditions are representative and that the study therefore has good external validity. There is evidence that this assumption is not always realised (eg Crabbe *et al* 1999; Würbel 2001; Sherwin 2004). The present study indicates that animals from standard cages might routinely be more anxious than mice from enriched cages. This anxiety is highly likely to have an impact on other aspects of the animals' biology, and will influence responses both within and outside the home-cage in ways that would be difficult or impossible to predict. This might easily reduce the external validity of studies from animals in such housing and thus negate the reason for their being housed in the first instance.

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References

Adams N and Oldham TD 1996 Seminatural housing increases subsequent ethanol intake in male Maudsley reactive rats. *Journal of Studies on Alcohol 57*: 349-351

Bertoglio LJ and Carobrez AP 2002 Prior maze experience required to alter midazolam effects in rats submitted to the elevated plus-maze. *Pharmacology, Biochemistry and Behavior 72*: 449-455

Broom DM and Johnson KG 1993 *Stress and Animal Welfare.* Chapman and Hall: London, UK

Chapillon P, Manneche C, Belzung C and Caston J 1999 Rearing environmental enrichment in two inbred strains of mice: 1: Effects on emotional reactivity. *Behaviour Genetics 29*: 41-46

Chamove AS 1989 Cage design reduces emotionality in mice. *Laboratory Animals 23*: 215-219

Colpaert FC, De Witte P, Mondi AN, Awouters F, Niemegeers CJE and Jansen PA 1980 Self-administration of the analgesic suprofen in arthritic rats: evidence of Mycobacterium butyricum-induced arthritis as an experimental model of chronic pain. *Life Sciences 27*: 921-928

Colpaert FC, Meert Th, De Witte P and Schmitt P 1982 Further evidence validating adjuvant arthritis as an experimental model of chronic pain in the rats. *Life Sciences 31*: 67-75

Crabbe JC, Wahlsten D and Dudek BC 1999 Genetics of mouse behavior: interactions with laboratory environment. *Science 284*: 1670-1672

Cruz-Morales SE, Santos NR and Brandao ML 2002 One-trial tolerance to midazolam is due to enhancement of fear and reduction of anxiolytic sensitive behaviours in the elevated plus-maze retest in the rat. *Pharmacology, Biochemistry and Behaviour 72*: 973-978

Danbury TC, Weeks CA, Chambers JP, Waterman-Pearson AE and Kestin SC 2000 Self-selection of the analgesic drug, carprofen, by lame broiler chickens. *Veterinary Record 146*: 307-311

Dielenberg RA and McGregor IS 2001 Defensive behavior in rats towards predatory odors: a review. *Neuroscience and Behavioral Reviews 25*: 597-609

Fish EW, Sekinda M, Ferrari PF, Dirks A and Miczek KA 2000 Distress vocalizations in maternally separated mouse pups: modulation via 5-HT1A, 5-HT1B and GABA(A) receptors. *Psychopharmacology 149*: 277-285

Harris JA and Westbrook RF 1994 Effects of midazolam and naloxone in rats tested for sensitivity/reactivity to formalin pain in a familiar, novel or aversively conditioned environment. *Psychopharmacology 115*: 65-72

Jennings M, Batchelor GR, Brain PF, Dick A, Elliot H, Francis RJ, Hubrecht RC, Hurst JL, Morton DB, Peters AG, Raymond R, Sales GD, Sherwin CM and West C 1998 Refining rodent husbandry: the mouse. Report of the Rodent Refinement Working Party. *Laboratory Animals 32*: 233-259

Lopez MM and Navarro JF 1999 Effects of midazolam on agonistic behaviour in male mice. *Psicothema 11*: 367-374

McGregor IS, Schrama L, Ambermoon P and Dielenberg RA 2002 Not all 'predator odours' are equal. Cat odour but not 2,4,5 trimethylthiazoline (TMT; fox odour) elicits specific defensive behaviours in rats. *Behavioural Brain Research 129*: 1-16

Moberg GP and Mench JA (eds) 2000 *The Biology of Animal Stress: Basic Principles and Implications for Animal Welfare.* CABI Publishing: Wallingford, UK

Nunes-de-Souza RL, Canto-de-Souza A, da-Costa M, Fornari RV, Graeff FG and Pela IR 2000 Anxiety-induced antinociception in mice: effects of systemic and intra-amygdala administration of 8-OH-DPAT and Midazolam. *Psychopharmacology 150*: 300-310

Olsson IAS and Dahlborn K 2002 Improving housing conditions for laboratory mice: a review of 'environmental enrichment'. *Laboratory Animals 36*: 243-270

Olsson IAS, Nevison CM, Patterson-Kane E, Sherwin CM, van de Weerd HA and Würbel H 2003 Understanding behaviour: the relevance of ethological approaches in laboratory animal science. *Applied Animal Behaviour Science 81*: 245 264

Pain L, Oberling P, Sander G and DiScala G 1997 Effect of midazolam on propofol-induced positive affective state assessed by place conditioning in rats. *Anaesthesiology 87*: 935-943

Pakulska W and Czarnecka E 2001 Effect of diazepam and midazolam on the antinociceptive effect of morphine, metamizol and indomethican in mice. *Pharmazie 56*: 89-91

Pickup HE, Cassidy AM, Danbury TC, Weeks CA, Waterman AE and Kestin SC 1997 Self selection of an analgesic by broiler chickens. *British Poultry Science 38*: S12-S13 (Suppl)

Prior H and Sachser N 1995 Effects of enriched housing environment on the behaviour of young male and female mice in four exploratory tasks. *Journal of Experimental Animal Science 37*: 57-68

Rockman GE and Gibson JEM 1992 Effects of duration and timing of environmental enrichment on voluntary ethanol intake in rats. *Pharmacology, Biochemistry and Behavior 41*: 689-693

Rosa VP, Vanderesen N, Calixto AV, Kovaleski DF and

Faria MS 2000 Temporal analysis of the rat's behavior in the plus-maze: effect of midazolam. *Pharmacology, Biochemistry and Behavior 67*: 177-182

Roy V, Belzung C, Delarue C and Chapillon P 2001 Environmental enrichment in BALB/c mice — effects in classical tests of anxiety and exposure to a predatory odour. *Physiology and Behavior 74*: 313-320

Sannerud CA and Ator NA 1995 Drug discrimination analysis of Midazolam under a 3-lever procedure. 2: Differential effects of benzodiazepine receptor agonists. *Journal of Pharmacology and Experimental Therapeutics 275*: 183-193

Schrijver NCA, Bahr NI, Weiss IC and Würbel H 2002 Dissociable effects of isolation rearing and environmental enrichment on exploration, spatial learning and HPA activity in adult rats. *Pharmacology, Biochemistry and Behavior 73*: 209-224

Sherwin CM 2002 Comfortable quarters for mice. In: Reinhardt V and Reinhardt A (eds) *Comfortable Quarters for Animals* pp 6-17. Animal Welfare Institute: Washington, USA

Sherwin CM 2004 The influences of standard laboratory cages on rodents and the validity of research data. In: Kirkwood JK, Roberts EA and Vickery S (eds) *Proceedings of the UFAW International Symposium 'Science in the Service of Animal Welfare', Edinburgh, 2003. Animal Welfare 13* (Suppl): in press

Soderpalm AHV and Hansen S 1998 Benzodiazepines enhance the consumption and palatability of alcohol in the rat. *Psychopharmacology 137*: 215-222

Sun L, Falk JL, Nguyen KN and Lau CE 2000 Post-injection delays in experimental chambers, but not in home cages, produce both sensitization and tolerance of operant behaviour to Midazolam: relation to pharmacokinetics. *Behavioural Pharmacology 11*: 133-142

van de Weerd HA, Baumans V, Koolhaas JM and VanZutphen LFM 1994 Strain-specific behavioural response to environmental enrichment in the mouse. *Journal of Experimental Animal Science 36*: 117-127

Würbel H 2001 Ideal homes? Housing effects on rodent brain and behaviour. *Trends in Neuroscience 24*: 207-211

https://doi.org/10.1017/S0962728600026634 Published online by Cambridge University Press