Moving forward with standardization: The need for parametric analyses of apparatus design (and test procedure) to identify factors that influence test validity

Richard E. Brown
Psychology Department
Dalhousie University
Halifax, Nova Scotia, Canada, B3H 4R2
rebrown@dal.ca
Abstract

A behavioural phenotype is the observable characteristic of humans or mice with a particular genetic or chromosomal disorder. The behavioural phenotype of a transgenic mouse model will differ from the phenotype of the wild-type control, reflecting the neuro-genetic changes in the genetically altered mouse. In order to determine reliable and valid behavioural phenotypes, we must ensure that the tests we use measure what we think they measure (Schellinck et al. 2010, Advances in the Study of Behavior, 41, 255-366). I will discuss two examples of how the design of an apparatus affects the results. First, Forwood et al (2005, Hippocampus, 15, 347-55) showed that the design of the test apparatus determines the roles of the hippocampus versus the parietal cortex in studies of object recognition in the rat. Second, we have shown that the apparatus and procedure for testing mice in the Barnes maze (O’Leary & Brown, 2011, Journal of Neuroscience Methods, in press) determines what they learn and remember. Thus in any “standardized” test one must be sure of what is being measured in order to make conclusions about the neural and/or genetic mechanisms underlying the learning and memory process under study. I will also discuss how preclinical assessment of novel drugs in mouse models of neurodegenerative diseases is impeded when differences between transgenic and wildtype mice are unreliable due to differences in apparatus design.
10 types of hammers... Which one is right for you?

Hammers come in many different shapes and forms. Using the wrong hammer can make a project go sour very quickly. It’s a good idea to know what hammers are available and when to use them.

- Sledgehammer
- Mason’s hammer
- Mallets
- Deadblow
- Framing hammer
- Claw hammer
- Tack hammer
- Ball pein hammer
- Roofing hammer
- Drywall hammer
Psychology as the behaviorist views it is a purely objective experimental branch of natural science. Its theoretical goal is the prediction and control of behavior. Introspection forms no essential part of its methods, nor is the scientific value of its data dependent upon the readiness with which they lend themselves to interpretation in terms of consciousness. The behaviorist, in his efforts to get a unitary scheme of animal response, recognizes no dividing line between man and brute. The behavior of man, with all of its refinement and complexity, forms only a part of the behaviorist's total scheme of investigation.
The importance of apparatus in the scientific study of Behaviour

John B. Watson argued that psychology should be an objective experimental branch of natural science whose theoretical goal is not the understanding of mind but the prediction and control of behaviour. Watson and earlier behaviourists used objective, experimental methods in the study of behaviour in order to separate their research from the method of introspection.

For the early behaviourists, the methods for the analysis of leaning and memory involved objective observations of behavioural data varying as a function of the experimental manipulation and control of stimulus conditions.

Lubbock (1882) introduced apparatus and quantification into the study of animal behaviour to provide precise, detailed, quantitative descriptions of the conditions of observation.

By 1894 experimental apparatus was used in behavioural experiments for stimulus control and quantification of responses.

This resulted in the rapid development of objective methods for experimental control, quantification and registration of behaviour, measurement, and research design.

Thorndike (1898) developed the puzzle box apparatus for the observation of animal learning and employed it in systematic laboratory research. Thorndike's general experimental technique was:

- **objective**: it minimized the influence of the observer...
- **quantitative**: the course of learning could be measured accurately by recording the time to show a correct response on each trial.
- **reproducible**: the work of one investigator could be repeated and verified by others.
- **flexible**: the responses required could be varied in kind and complexity.
- **natural**: not too remote from the animal's ordinary course of life...
- **convenient**: a large enough sample of animals could be studied to provide a representative picture of each of a variety of species.

By 1913, the tools for an objective and scientific psychology focused on what Watson described as "the scientific determination of modes of behavior...(using) an objective standard of interpretation...without mentioning consciousness or deviating from a (wide) biological point of view."

A CIRCULAR MAZE WITH CAMERA LUCIDA ATTACHMENT

JOHN B. WATSON

Figure 1. General view illustrating camera-lucida maze

How Many Ways Can Mouse Behavioral Experiments Go Wrong? Confounding Variables in Mouse Models of Neurodegenerative Diseases and How to Control Them

HEATHER M. SCHELLINCK, DAVID P. CYR, and RICHARD E. BROWN

DEPARTMENT OF PSYCHOLOGY & NEUROSCIENCE INSTITUTE,
DALHOUSIE UNIVERSITY, HALIFAX, NOVA SCOTIA, CANADA B3H 4J1

But Mouse, thou art no thy-lane,
In proving foresight may be vain;
The best laid schemes o' Mice an' Men,
Gang aft agley,
An' les'e us nought but grief an' pain,
For promis'd joy!
Still, thou art biest, compar'd wi' me!
The present only toucheth thee:
But Och! I backward cast my e'e,
On prospects dear!
An' forward, tho' I canna see,
I guess an' fear!

From "To a Mouse"
Robert Burns, 1785

In this chapter, we provide an overview of the effects of genetic manipulation on the brain and behavior of the mouse, point out some criticisms of these studies, and then examine in detail the number of confounds and control procedures that must be considered before one can conclude that a particular genetic manipulation is the cause of a particular behavioral change. Our goal in this paper is to provide the information necessary to conduct the controlled analysis of the effects of genetic manipulation on mouse behavior.
What is a Behavioural Phenotype?

Annotation: Behavioural Phenotypes: A Window Onto the Biology of Behaviour

Jonathan Flint

John Radcliffe Hospital, Oxford, U.K.

Characteristic behavioural patterns (including cognitive processes and social interactions) have been reported in a number of syndromes arising from genetic or chromosomal abnormalities, suggesting that molecular analysis of the underlying defect could reveal the biological basis of the behavioural phenotype. Because of the rarity of many of the syndromes, and the complexity of their genetic basis, there are great difficulties in establishing the validity of the association between syndrome and behavioural phenotype. Nevertheless, evidence from animal studies with relevance to human behavioural phenotypes shows that the pathway from genotype to phenotype may be accessible by careful delineation of behavioural phenotypes.

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Keywords: Behavioural phenotype, behavioural genetics, dysmorphic syndromes, animal models
Behavioural Phenotyping of Transgenic Mice

Richard E. Brown, Dalhousie University

Abstract This paper reviews the current work on mouse genetics, brain, and behaviour in my laboratory. It starts with a historical account of our research and shows how certain research themes, such as olfaction, learning, social behaviour, and environmental effects in rodents have led to our current research on behavioural phenotyping of inbred, mutant, knockout, and transgenic mice. We are concerned with finding neural and behavioural sequelae to genetic manipulations in mice and use a battery of tests to detect behaviours that are altered in genetically modified mice. In this way we are working to dissociate neural and behavioural effects of different gene manipulations in mouse models of neurodegenerative diseases. Sensory, motor, cognitive, affective, and social behaviours may all be affected by gene manipulation, thus careful behavioural techniques, with attention to the mice themselves, the apparatus, and procedure, experimenter variables, and environmental effects are necessary in order to determine a reliable and valid mouse behavioural phenotype. As both the genome and the environment have significant effects on the behavioural phenotype, our future research will utilize an epigenetic approach to examine how environmental cues modulate gene expression in the behavioural phenotyping of transgenic mice.

This paper gives an overview of our research on the behavioural phenotyping of mouse models of neurodegenerative disease. Because this research grew out of previous projects, this paper begins with an historical account of our research program. This shows how one research project grows from another in unexpected ways, the relatedness of seemingly unrelated projects, and the importance of many colleagues and students who have contributed and continue to contribute to our work.
## Identifying Behaviour Phenotypes in Mice

### Box 1: Experimental Approaches to Behavioral Assessment

<table>
<thead>
<tr>
<th>Approach</th>
<th>Advantages</th>
<th>Disadvantages</th>
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<tbody>
<tr>
<td><strong>Standard tests</strong></td>
<td>• Extensive pharmacological validation available</td>
<td>• Risk of data misinterpretation</td>
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<td></td>
<td>• Relatively simple and rapid to administer</td>
<td>• Limited construct and face validity</td>
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<td></td>
<td></td>
<td>• Certain assays have low throughput</td>
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<td></td>
<td></td>
<td>• Serial tests have very low-throughput and require large numbers of animals</td>
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<tr>
<td><strong>Batteries of standard tests</strong></td>
<td>• Pharmacological validation available</td>
<td>• Limited construct and face validity</td>
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<tr>
<td></td>
<td>• Reduced numbers of animals required</td>
<td>• Certain assays have low throughput</td>
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<tr>
<td></td>
<td>• Data interpretation improved by assessing diverse behaviors</td>
<td>• Test order may confound results</td>
</tr>
<tr>
<td><strong>Naturalistic assays</strong></td>
<td>• Behaviors assessed in a more ethologically valid context</td>
<td>• Low-throughput</td>
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<td>• Reduces misinterpretations of behavioral responses</td>
<td>• May be time-consuming to establish assay conditions for certain behavioral processes</td>
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<td></td>
<td>• Wide range of complex species-characteristic behaviors may be elicited</td>
<td>• Lack of widespread experience with these approaches for mice</td>
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<tr>
<td><strong>High-throughput screens</strong></td>
<td>• Capable of very high throughput</td>
<td>• Requires extensive pharmacological validation</td>
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<td></td>
<td>• Potential for detecting novel behaviors</td>
<td></td>
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<tr>
<td><strong>Quantitative home cage behavior analysis</strong></td>
<td>• Effects of handling and novelty minimized</td>
<td>• Limited construct or face validity</td>
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<td>• Assesses multiple behaviors over long time intervals</td>
<td>• Some rely on limited tests, increasing incidence of false positives and false negatives</td>
</tr>
<tr>
<td></td>
<td>• Potential for detecting novel behaviors</td>
<td>• Requires extensive pharmacological validation</td>
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<td></td>
<td>• Relatively low labor costs</td>
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</table>

What is a standard(ized) test?

1. Have an established apparatus and procedure
2. Have specified procedures for administration and recording
3. Present the same tasks and require the same response from all test takers
4. Administered under standardized or controlled conditions
5. Standardized tests should meet acceptable standards for technical qualities on construction, administration and use
6. Provide a systematic, quantitative procedure for describing behaviours
7. The test design is derived from experiment or observation, rather than theory
8. Provides norms to which the scores of test takers can be compared in order to ascertain their relative standing

“The primary strengths of standardized tests, if they are properly designed and properly used, are that they can eliminate biases in assessment and they provide data that can be aggregated, permitting comparisons of groups to a standard.”
Are behavioural tests reliable & valid?

A valid test – measures what it is supposed to (eg. Spatial memory)
A reliable test – gives the same results in repeated trials
An unreliable test can not be valid.

How sensitive is a test for detecting differences between wildtype and genetically modified mice?
Does object recognition memory involve the hippocampus?

Impaired Recognition Memory in Rats after Damage to the Hippocampus

Robert E. Clark,1 Stuart M. Zola,1,2 and Larry R. Squire1,2

1Veterans Affairs Medical Center, San Diego, California 92161, and 2University of California San Diego, La Jolla, California 92093

Rats with radio-frequency or ibotenic acid lesions of the hippocampus and rats with radio-frequency lesions of the fornix were tested on the visual paired comparison task (VPC), a test of recognition memory. Memory was assessed at five different delay intervals ranging from 10 sec to 24 hr. All operated groups performed normally at the shorter delays (10 sec and 1 min). Across longer delays, the two groups with hippocampal damage were impaired. Rats with fornix lesions performed well on the VPC task but were impaired on a spatial task (spontaneous alternation). The results show that the hippocampus is essential for normal recognition memory. Moreover, fornix lesions need not mimic the effects of direct damage to hippocampal tissue. The findings are discussed in the context of the contribution of the hippocampus to recognition memory.

Key words: hippocampus; rats; visual paired comparison; fornix; ibotenic acid lesions; radio-frequency lesions; spontaneous alternation
“Standard” apparatus for novel object recognition memory

The Novel Object Recognition Test is based on the premise that rodents will explore a novel object more than a familiar one if the animal is able to remember the familiar one.

Following the training period, the rodent is removed from the environment for a delay period which can range from 5 minutes to 24 hours, depending on the type of memory being tested. After the delay, the rodent is returned to the arena, where one of the original objects has been replaced by a new, dissimilar Novel Object. The amount of time the rodent spends exploring each object provides a powerful measurement of memory integrity and attention.

The flexibility of the Novel Object Recognition Test allows for testing of short- or long-term memory, and can be used to selectively test the effects of an acute drug treatment on a specific stage of memory formation.

Objects may vary from those pictured.
Hippocampal Lesions That Abolish Spatial Maze Performance Spare Object Recognition Memory at Delays of up to 48 Hours

S.E. Forwood,* B.D. Winters, and T.J. Bussey

ABSTRACT: The hippocampus is widely considered to be a critical component of a medial temporal lobe memory system, necessary for normal performance on tests of declarative memory. Object recognition memory is thought to be a classic test of declarative memory function. However, previous tests of the effects of hippocampal lesions on object recognition memory have not always supported this view. One possible reason for this inconsistency is that previously reported effects of hippocampal lesions on object recognition memory tasks may have stemmed not from a deficit in object recognition memory per se, but as a result of spatial and contextual confounds in the task. Thus, in the present study, we used a spontaneous object recognition test in a modified apparatus designed to minimize spatial and contextual factors. A group of rats with complete excitotoxic lesions of the hippocampus and a group of control rats were tested on this modified spontaneous object recognition task with retention delays of up to 48 h. These rats were also tested on a spatial nonmatching-to-place task. Spatial memory performance was abolished following hippocampal lesions, whereas performance on the recognition memory task was intact at all delays tested. © 2004 Wiley-Liss, Inc.

KEY WORDS: rat; declarative memory; medial temporal lobe
Hippocampal lesions impair spatial memory

FIGURE 1. Example of a trial in the cross-maze. R and S indicate “reward” and “start” arms, respectively (see text for explanation), a star indicates a reward is present in the arm, a black line indicates the arm is blocked. Left is the sample phase, with the rat placed in the near start arm and the left reward arm baited. The rat must choose the baited arm; the other two arms are blocked. Right is the choice phase; with the rat placed in the far start arm and the right arm baited. The rat can choose either reward arm, a correct response is rewarded.

FIGURE 3. Performance of control and hippocampal lesioned animals on the first four sessions of the cross-maze task at minimal delays (~5 s). Error bars indicate SEM.
Hippocampal lesions do not impair object recognition memory.

But – parietal cortex lesions do impair object recognition memory.
The apparatus determines the results

The shape of the apparatus determines whether or not hippocampal lesions impair object recognition memory.

Which one is a valid and reliable test of the neural basis of object recognition memory?
The Barnes maze for visuo-spatial learning and memory in mice

Adaptation of the circular platform spatial memory task for mice: use in detecting cognitive impairment in the APP\textsubscript{sw} transgenic mouse model for Alzheimer’s disease

Patrick N. Pompl \textsuperscript{a}, Michael J. Mullan \textsuperscript{b}, Kimberly Bjugstad \textsuperscript{a}, Gary W. Arendash \textsuperscript{a,*}

\textsuperscript{a} Alzheimer’s Research Laboratory, Department of Biology, University of South Florida, Tampa, FL 33620, USA
\textsuperscript{b} Roskamp Institute, Department of Psychiatry University of South Florida, Tampa, FL 33613, USA

Abstract

A methodology is described for use of a 16-hole circular platform task to test spatial memory in mice. Both bright light and a fan were used to motivate mice to escape the platform surface through a single hole containing an attached escape box. For each daily trial, three correlated measures (escape latency, number of errors, and error rating) comprehensively evaluated cognitive performance. In an initial study, the ‘spatial’ nature of this task was demonstrated by the much poorer performance of non-transgenic mice when visual cues are removed. Behavioral sensitivity of the circular platform task was then shown through its ability to discern cognitive impairment in 7-month-old transgenic mice, carrying the mutant APP\textsubscript{sw} gene for early-onset Alzheimer’s disease in humans, from non-transgenic litter-mates. Since there are currently only a few tasks available to definitively test cognitive performance in mice, the circular platform task offers a versatile, multiple-measure option with numerous advantages. Particularly in view of the increasing number of genetically manipulated mouse models being produced, the circular platform task should be most useful in providing a sensitive evaluation of cognition in mice. © 1999 Elsevier Science B.V. All rights reserved.

Fig. 1. Illustrative drawing of the circular platform apparatus. Various visual cues are positioned along the curtain enclosure and platform wall. Also note flood lights and high speed fan, present to produce an adverse environment motivating entrance to the escape box located beneath one of the holes of the platform.
How does the design of the Barnes maze influence visuo-spatial learning & memory?

<table>
<thead>
<tr>
<th>Small-walled maze (SWBM)</th>
<th>Small maze (SBM)</th>
<th>Large maze (LBM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>69 cm diameter</td>
<td>69 cm diameter</td>
<td>122 cm diameter</td>
</tr>
<tr>
<td>16 holes</td>
<td>16 holes</td>
<td>16 holes</td>
</tr>
<tr>
<td>15 cm high wall</td>
<td></td>
<td></td>
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<tr>
<td>Intramaze cues</td>
<td></td>
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</tbody>
</table>
Does apparatus design influence learning?

Mice on the LBM used the spatial strategy more than the serial strategy

Mice on the SWBM used the serial strategy more than the spatial strategy

Does apparatus design influence memory?

No Curtain

Mice on SBM and LBM spent more time in correct zone, Z1 and Z15 than expected by chance, mice in SWBM did not.

Which design is the most sensitive visuo-spatial test?

η² = .203

η² = .250

η² = .232

η² = .612


Reliability & Validity of Apparatus

Apparatus design influences the learning and memory strategy and the underlying neural pathways involved.
What about measuring anxiety in mice?

Three apparatus commonly used

- Elevated plus-maze (EPM)
- Open field (OF)
- Light/dark transition box (LDB)

Which has the best reliability and validity?
Which is the most sensitive?
A Review of the Validity and Variability of the Elevated Plus-Maze as an Animal Model of Anxiety

SANDY HOGG

Psychopharmacology Research Unit, UMDS Division of Pharmacology, Guy's Hospital Medical School, London SE1 9RT, UK

HOGG, S. A review of the validity and variability of the elevated plus-maze as an animal model of anxiety. PHARMACOL BIOCHEM BEHAV 54(1) 21–30, 1996. —Despite or possibly by virtue of the fact that it is one of the most commonly used animal models of anxiety the Elevated Plus-Maze (EPM) results in a wide range of, often contradictory, results following pharmacological experiments. The responses from a questionnaire distributed to 65 groups that have published studies using the EPM in the past 3 years has, along with reference to published reports, enabled some conclusions regarding the influencing factors to be drawn. Some evidence for differential sensitivities between strains exists, with albino rats being more sensitive to the anxiolytic effects of 5-HT1 receptors agonists and 5-HT2 receptors agonists than pigmented animals. Most important, however, is the manipulation of the animals prior to testing and the aversiveness of the test conditions themselves. Stressing animals before testing (e.g., by moving from holding to test room) or using more aversive test conditions (e.g., elevated light levels) increases sensitivity to potential anxiolytics. Animals that are habituated to gentle handling or tested in less aversive conditions (e.g., EPM with ledges) show reduced likelihood of anxiolytic responses with administration of 5-HT3 antagonists, 5-HT1A agonists, and benzodiazepines.
The open field as a paradigm to measure the effects of drugs on anxiety-like behaviors: a review

Laetitia Prut, Catherine Belzung*

EA3248, Psychobiologie des Émotions, Faculté des Sciences et Techniques, Université François Rabelais, Parc de Grandmont Avenue Monge, 37200 Tours, France

Accepted 10 December 2002

Abstract

The open field is a very popular animal model of anxiety-like behavior. An overview of the literature on the action elicited by effective or putative anxiolytics in animal subjected to this procedure indicates that classical treatments such as benzodiazepine receptor full agonists or 5-HT1A receptor full or partial agonists elicit an anxiolytic-like effect in this procedure in most cases (approximately 2/3). However, compounds (triazolobenzodiazepines such as adinazolam and alprazolam, selective serotonin reuptake inhibitors) that have a different spectrum of therapeutic efficacy in anxiety disorders such as panic attacks, generalized anxiety disorder or obsessive-compulsive disorder were poorly effective as anxiolytics in the open field test, suggesting that this paradigm may not model features of anxiety disorders. The procedure is also relevant for the study of compounds endowed with anxiogenic effects, as such effects were detected after treatments with benzodiazepine receptor inverse agonists or with corticotropin releasing factor (CRF) receptor agonists.

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Keywords: Open field; Benzodiazepine; 5-HT (5-hydroxytryptamine: serotonin); Neuropeptide; Anxiety
The mouse light/dark box test

Michel Bourin*, Martine Hascoët

Faculty of Medicine, EA 3256 Neurobiologie de l’Anxiété et de la Dépression, Faculté de Médecine BP 53508, 44035 Nantes Cedex 1, France
Accepted 10 December 2002

Abstract

The light/dark test is based on the innate aversion of rodents to brightly illuminated areas and on the spontaneous exploratory behaviour of rodents in response to mild stressors, that is, novel environment and light. The test apparatus consists of a small dark safe compartment (one third) and a large illuminated aversive compartment (two thirds). The test was developed with male mice. The strain, weight and age may be crucial factors. The extent to which an anxiolytic compound can facilitate exploratory activity depends on the baseline level in the control group. Differences between the type and severity of external stressors might account for the variable results reported by different laboratories. The light/dark test may be useful to predict anxiolytic-like or anxiogenic-like activity in mice. Transitions have been reported to be an index of activity-exploration because of habituation over time, and the time spent in each compartment to be a reflection of aversion. Classic anxiolytics (benzodiazepines) as well as the newer anxiolytic-like compounds (e.g. serotonergic drugs or drugs acting on neuropeptide receptors) can be detected using this paradigm. It has the advantages of being quick and easy to use, without requiring the prior training of animals.

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Keywords: Antidepressant; Antipsychotic; Anxiolytic; Light/dark test, mouse; Neuropeptide receptor ligand
But – there is the confound of locomotor behaviour...
## Correlational data for mice for the EPM, OF & LDB

From Brown Lab contribution to JAX Phenome Project.

### Measures and Correlations

<table>
<thead>
<tr>
<th>Measure</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
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</thead>
<tbody>
<tr>
<td><strong>Anxiety</strong></td>
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<tr>
<td>1. EPM % time in open arms</td>
<td>---</td>
<td>-.107</td>
<td>.052</td>
<td>-.104</td>
<td>.033</td>
<td>.112*</td>
<td>-.325**</td>
<td>-.169**</td>
<td>-.079</td>
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<tr>
<td>2. OF time in center</td>
<td>---</td>
<td></td>
<td>-3</td>
<td>.048</td>
<td>.043</td>
<td>-.034</td>
<td>.007</td>
<td>.179**</td>
<td>.083</td>
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<tr>
<td>3. LDB % time in light</td>
<td>---</td>
<td></td>
<td></td>
<td>-.167**</td>
<td>-.077</td>
<td>.021</td>
<td>-.035</td>
<td>.037</td>
<td>.161**</td>
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<td>4. EPM defecations</td>
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<td></td>
<td></td>
<td></td>
<td>.484**</td>
<td>.543**</td>
<td>-.044</td>
<td>-.310**</td>
<td>-.323**</td>
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<tr>
<td>5. OF defecations</td>
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<td></td>
<td>.438**</td>
<td>-.035</td>
<td>-.265**</td>
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<td>6. LDB defecations</td>
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<td></td>
<td>-.248**</td>
<td>-.325**</td>
<td>-.336**</td>
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<td><strong>Locomotion</strong></td>
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<td>7. EPM line crosses</td>
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<td></td>
<td>.378**</td>
<td>.287**</td>
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<td>8. OF line crosses</td>
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<td></td>
<td>.738**</td>
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<td>9. LDB line crosses</td>
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</table>

* *p<.05; **p<.01; n = 310-355; 14 strains of inbred mice

**Note that:**
1. measures of locomotion are significantly correlated
2. frequency of defecations are significantly correlated
3. measures of anxiety are poorly correlated
4. within each test, measures of anxiety and locomotion are correlated

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From Brown Lab contribution to JAX Phenome Project.
Animal models of anxiety: do I need multiple tests?

André Ramos

The combination of cutting-edge molecular technology and high-throughput phenotyping tools will not bring the expected contribution to the pre-clinical study of anxiety if not paralleled by an increase in our capacity to interpret behavioral data. Here, previous views about the multidimensional nature of emotional behaviors will be expanded and the psychological meaning and behavioral overlaps of widely used anxiety tests such as the open field, elevated plus maze and light–dark box will be discussed. It is proposed here that short-term, intra-individual variations in emotionality, although normally overlooked, constitute an important factor in the study of anxiety and can lead to unreliable estimates of the similarities between tests. The physical integration of different current tests in one single apparatus, in such a way that the emotional status of an animal becomes assessable through a series of distinct tasks, could contribute to increase reliability, rapidity and comprehensiveness in behavioral testing.

Figure 3. The integration of three widely used tests of anxiety, the OF, EPM and LDB. The new test physically connects the three individual apparatuses by placing them side by side at the same height and by adding openings at the interface between their respective less aversive areas (OF periphery—EPM closed arms—LDB dark compartment) to encourage transitions between tests. During a 15-min trial, a naïve animal initially placed in the center of the OF can freely explore all areas, thus, providing the classical anxiety- and locomotion-related measures of all tests within the same time window [50].
Measuring anxiety- and locomotion-related behaviours in mice: a new way of using old tests

Leanne M. Fraser · Richard E. Brown · Ahmed Hussin · Mara Fontana · Ashley Whittaker · Timothy P. O’Leary · Lauren Lederle · Andrew Holmes · André Ramos

Abstract

Rationale Batteries of tests that are thought to measure different aspects of anxiety-related behaviour are used to characterise mice after genetic or pharmacological manipulation. However, because of the potentially confounding effects of repeated testing and natural intra-individual variations in behaviour over time, subjecting mice to a succession of tests is not ideal.

Objectives The aim of this study was to investigate, in mice, the utility of an integrated apparatus that combines three classical tests of anxiety, the open field, elevated plus maze (EPM) and light/dark box.

Methods Mice from four different strains (CD-1, BALB/cJ, DBA/2J, C57BL/6J) were used in a series of five experiments where their behaviour was observed for 15 min in the integrated apparatus. Responses to anxiety-modulating drugs and 2-day repeated testing were evaluated.
EPM gives more significant dose-response curves than OF or LDB.

DZP = diazepam
APZ = alprazolam
So is the triple test more reliable and valid than the OF, EPM or LDB for measuring anxiety?

We continue our research on this apparatus.
Drug testing

Mouse models of neurodegenerative diseases

1. Is there a reliable and valid cognitive deficit?
2. Does a drug treatment reverse the deficit?
3. Is the apparatus used a valid and reliable test for the cognitive deficit examined?
4. How can we reliably replicate the size of a deficit without standardization?
Conclusion

If a test apparatus and procedure are to be “standardized” they must undergo rigorous parametric testing to quantify their reliability, validity and power to detect phenotype differences and drug effects.
THE END
Measuring normal and pathological anxiety-like behaviour in mice: a review

Catherine Belzung a,*, Guy Griebel b

a EA 3248 Psychobiologie des émotions, UFR Sciences et Techniques, Parc Grandmont, Avenue Monge, F-37200 Tours, France
b CNS Research Department, Sanofi-Synthelabo, Bagneux, France

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Abstract

Measuring anxiety-like behaviour in mice has been mostly undertaken using a few classical animal models of anxiety such as the elevated plus-maze, the light/dark choice or the open-field tests. All these procedures are based upon the exposure of subjects to unfamiliar aversive places. Anxiety can also be elicited by a range of threats such as predator exposure. Furthermore, the concepts of ‘state’ and ‘trait’ anxiety have been proposed to differentiate anxiety that the subject experiences at a particular moment of time and that is increased by the presence of an anxiogenic stimulus, and anxiety that does not vary from moment to moment and is considered to be an ‘enduring feature of an individual’. Thus, when assessing the behaviour of mice, it is necessary to increase the range of behavioural paradigms used, including animal models of ‘state’ and ‘trait’ anxiety. In the last few years, many mice with targeted mutations have been generated. Among them some have been proposed as animal models of pathological anxiety, since they display high level of anxiety-related behaviours in classical tests. However, it is important to emphasise that such mice are animal models of a single gene dysfunction, rather than models of anxiety, per se. Inbred strains of mice, such as the BALB/c line, which exhibits spontaneously elevated anxiety appear to be a more suitable model of pathological anxiety. © 2001 Elsevier Science B.V. All rights reserved.
Behavioural Phenotype


• A behavior (broadly defined to include cognitive processes and social interaction) consistently associated with, and specific to a syndrome with a chromosomal or genetic aetiology where there is little doubt that the phenotype is a result of the underlying lesion. [Flint J. Annotation: behavioural phenotypes. A window onto the biology of behaviour. J Child Psychol Psychiatry, 1996, 37, 355-67.]