

Avoidance behavior in mice

Refining behavioral analysis for genetic dissection

Mood and anxiety disorders are the most prevalent of all psychiatric disorders. Our research project focuses on the genetic dissection of certain behavioural phenotypes, specifically avoidance behaviour, with the aim to search for more selective pharmacological treatments of neurobehavioral disorders. The nature of standard behavioral tests makes it hard to interpret the behavioural variation observed in different mouse strains [1]. The individual locomotor activity levels, short duration of the experiment, experimenter interference and novelty responsiveness of the animal can interfere with the identification of genotype –phenotype relationships. Therefore, we combined a system for automated multi-day recordings (PhenoTyper® PT10S/P /N Version 1.01) with a designed home cage environment (HCE, figure 1), assessing the animals' reduced preference for exposed areas independent of motor activity levels. In the HCE, the animal can be monitored for avoidance and approach behaviour for several days without experimenter interference. By evaluating three days of continuous recording, measures for novelty as well as baseline behaviour can be obtained.

Recently, a novel method to examine the genetic influences on behaviour has been introduced [2]. A panel of chromosome substitution strains (CSS) was developed, derived from the C57BL/6J host strain and the A/J donor strain. Each of these substitution strains has a single chromosome from the donor strain substituting for the corresponding chromosome in the host strain. This has proven to be a powerful way to detect QTL's on the mouse chromosome [3,4]. With our home cage environ-

ment combined with this novel genetic strategie, we will attempt to show distinct phenotypes by dissecting the behaviour in refined behavioural phenotypes, taking into account novelty-induced and baseline behaviour, as well as circadian variation in behaviour as a function of genetic background.

MATERIAL AND METHODS

Male and female CSS mice were housed in groups, with tap water and chow provided ad libitum. A minimum of two weeks prior to the start of the experiment the animals were moved from the stables to the adaptation room next to the experimental room with a shifted 12:12 hr dark: light cycle (lights off at 1 PM).

The home cage environment (HCE) was used in these experiments, containing a shelter, drinking spout and two different feeding platforms. One hour prior to the dark period, mice were placed individually in the HCE for 73 hours. No experimenter interference occurred during these three days. At the end of the experiment body weight change and food- and water intake were measured. The distance moved, enter frequency and duration spent in the several zones were registered using PhenoTyper [5] and the data were analysed using EthoVision® 3.0 [6]. Infrared sensors placed in front of the food- and water supplies measured frequency and duration of feeding and drinking.

RESULTS

Following behavioral testing of the full panel of chromosome substitution strains in the home cage

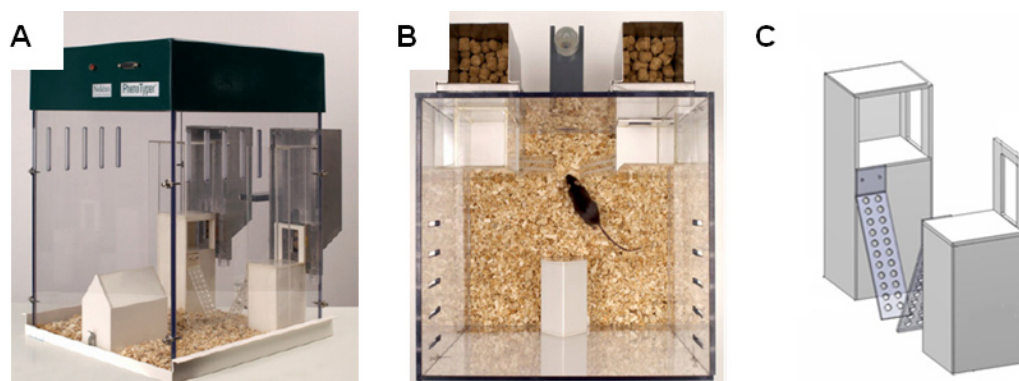


Figure 1: A) The home cage environment with the PhenoTyper top-unit. B) Top-view as recorded by camera. C) Feeding platforms as used in the home cage environment; one providing sheltered feeding, on exposed to the environment.

environment, we were able to select particular strains of interest. Chromosome 1 was found to be involved in both baseline and novelty induced motor activity levels in both males and females, and chromosome 15 and 19 were found to influence baseline avoidance behavior in females. In follow up studies, we were able to map new, distinct genetic loci specific for motor activity levels and avoidance behaviour. For chromosome 15, different genetic regions on this chromosome were identified influencing either novelty induced motor activity levels, novelty induced avoidance behavior and baseline avoidance behavior.

DISCUSSION

The refinement of behavioral testing by using automated longitudinal monitoring of mouse behavior as described here has proven to be very successful. Combining this designed living environment with the testing of a sensitive genetic-mapping panel has allowed the dissociation of complex strategies both at a genetic and behavioral level in mice. As possible relevant translational results have been obtained by using the home cage environment, it holds great potential for future research on genetics and pharmacology.

REFERENCES

1. Kas, M.J.; Van Ree, J.M. (2004). Dissecting complex behaviours in the post-genomic era. *Trends Neurosci.*, **27**, 366-369.
2. Nadeau, J.H.; Singer, J.B.; Matin, A.; Lander, E.S. (2000). Analysing complex genetic traits with chromosome substitution strains. *Nat. Genet.*, **24**, 221-225.
3. Singer, J.B.; Hill, A.E.; Burrage, L.C.; Olszens, K.R.; Song, J.; Justice, M.; O'Brien, W.E.; Conti, D.V.; Witte, J.S.; Lander, E.S.; Nadeau, J.H. (2004). Genetic dissection of complex traits with chromosome substitution strains of mice. *Science*, **304**, 445-448.
4. Singer, J.B.; Hill, A.E.; Nadeau, J.H.; Lander, E.S. (2005). Mapping quantitative trait Loci for anxiety in chromosome substitution strains of mice. *Genetics*, **169**, 855-862.
5. Visser, L.; Bos, R.; Spruijt, B.M. (2005). Automated home cage observations as a tool to measure the effects of wheel running on cage floor locomotion. *Behav. Brain Res.*, **160**, 382-388.
6. Noldus, L.P.; Spink, A.J.; Tegelenbosch, R.A. (2001). EthoVision: a versatile video tracking system for automation of behavioral experiments. *Behav. Res. Methods Instrum. Comput.*, **33**, 398-414.

CONTACT INFORMATION

J.G. Mooij-van Malsen¹, B. Olivier^{1,2} and M.J.H. Kas¹

¹ Rudolf Magnus Institute of Neuroscience, University Medical Centre Utrecht, The Netherlands

² Department of Psychopharmacology, University Utrecht, The Netherlands