

LAB_070 Foot Fault/Balance Beam Test for Rodents

I. OBJECTIVE

To describe the procedure for measuring bilateral and lateralised motor deficits underlying neurological function. This is a sensitive test based on foot faults (slips) made by a rodent when traversing either a gradually narrowing beam or a uniformly narrow beam.

II. COMMENTS / RECOMMENDATIONS

- Behavioural assessments are ideally performed in a dedicated behavioural suite.
- The environment should be free from uncontrolled external stimuli that may influence the animal's behaviour such as human traffic, unnecessary noise, and intense lighting.
- Male and female rodents should be tested separately, with one sex in the room at a time. Where possible
 males should be tested first, preferably on separate days but with at least thorough cleaning between the
 sexes. This is unless rodents are already housed within wire top cages or equivalent and both sexes are
 present in the home room.

III. EQUIPMENT

• PPE

Minimum PPE is gloves and gown, additional PPE may be required based on facility or additional risk e.g. working with infectious animals.

- Appropriate trolley for transporting cages.
- Disinfectant and paper towel for cleaning equipment.
- Apparatus often consists of loading and unloading platforms at either end of the beam (where no scoring occurs). The unloading platform can be covered by a dark box or lead to a home cage to encourage the rodent's crossing of the beam. Option 1 Is the use of a tapered beam that gradually narrows and has markings at regular intervals to identify segments with increasing difficulty of beam crossing. Option 2 Is the use of a uniformly narrow beam. The beam can include a ledge along its length on both sides providing a crutch for the rodent to use when a deficit is present (optional). Appropriate sizing for the apparatus is shown in the table below.

Arena dimensions	Mouse	Rats	
Overall length (cm)	130	175	
Loading platform (cm)	Minimum 10 x 3.5	Minimum 15 x 6	
Tapered run (cm)	100 long with taper from 3.5 to 0.5	135 long with taper from 6 to 2	
Even Run (cm)	100 long x 0.5-1.0	135 long x 2-2.5	
Ledge along length (cm) 1		2	
Unloading platform (cm)	20 x 20	20 x 20	

• The beam can be supported between any open space

Conditions:

- Investigators named in an animal ethics application, relative to this SOP, must be competent to implement the SOP
- Any variation to this SOP must be described in the relevant animal ethics application
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- Video recording equipment connected to a computer for video capturing to be scored manually OR use of an
 automated tapered beam apparatus connected to software in which foot faults onto the side ledge are
 automatically detected in real time.
- A curtain or screened off area for experimenter to be hidden from the rodents during testing, if available.

IV. PREPARATION

- 1. Check AEC approvals to ensure that the correct procedure and personnel are approved for the planned work.
- 2. Prepare equipment items including disinfecting prior to first use.
- 3. Bring rodents into the room (with lighting levels pre-set at the level required for the experiment) for at least 30 mins prior to start of experiment.

Length of habituation time in the testing room should be consistent for all rodents within an experiment.

V. PROCEDURE

1. Record light levels in the middle of the arena or room, for reproducibility and consistency. *Lux range should be between 30-100 LUX and should remain the same for all rodents within an experiment.*

- 2. Start recording and identify subject/s within the camera view and/or set up automatic detection with equipment.
- 3. Handling of rodents as per: <u>LAB_006 Handling and Restraint in Mice and Neonates</u> LAB_039 Handling and Restraint in Rats and Neonates
- 4. Training typically consists of three consecutive days with three trials each day.

All rodents for the study should complete their first trial of the day before starting the second trial to ensure sufficient and consistent time between trials for all rodents.

- 5. After training, rodents can be tested either for a 'once-off' period or can be tested longitudinally. For a 'once-off' trial rodents are tested for seven to ten consecutive days with three trials each day. If wanting to examine chronic drug administration effects or developing pathology then 3 trials a day once a week or fortnight can be utilised instead.
- 6. Rodents are positioned at the loading platform end of the beam and are allowed to freely explore the apparatus for 3 min before being gently nudged by the experimenter towards the dark box at the other end.
- 7. On the first training day, rodents are given 30 to 60 s of time in the dark box at the far end to facilitate the association of traversing the beam with 'safety'.
- 8. At the end of each trial, remove the rodent and return them to the home cage.

Ensure access to food and water.

- 9. Stop recording and make sure to save the video file or automatic tracking results.
- 10. Remove scat and thoroughly disinfect the arena and allow to dry completely.
- 11. Analysis can be made manually or output from the automatic detection from the sensors.

VI. ANALYSIS

- For the tapered balance beam the number of foot faults obtained for the different segments along the beam can be scored. These segments indicate the levels of difficulty.
- Time to cross the full length of the beam can also be analysed.
- Repeated measures across trials or days can be analysed, or analysis can be done before and after treatment or longitudinally with disease progression.

If analysis is done manually, the rater should be blinded to treatment.

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VII. REFERENCES

- Ardesch DJ, Balbi M, & Murphy TH. Automated touch sensing in the mouse tapered beam test using Raspberry Pi. J Neurosci Meth. 2017, (291) 221-226. <u>https://www.sciencedirect.com/science/article/pii/S0165027017303175#fig0010</u>
- Schallert T, Woodlee MT, Fleming SM. *Disentangling multiple types of recovery from brain injury*. Institute for Neuroscience UT - Austin. In Pharmacology of Cerebral Ischemia, J. Krieglstein, S. Klumpp (Eds). 2002, pp 201-215. <u>https://www.scienceopen.com/document?vid=fd1486f4-dbda-468c-878e-f12323910ae6</u>

Version #	Reviewing AEC (note: all other relevant AECs ratify the approval)	AEC Review Date	Approval To Date
1	LBM	06/10/2022	06/10/2025

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