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Barnes Maze

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Protocol status: Working

We use this protocol and it's working

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Abstract

This protocol provides a detailed, step-by-step guide for conducting the Barnes maze test, a well-established behavioral assay for assessing spatial learning and memory in mice and rats. The maze utilizes a rodent's innate aversion to open, brightly lit spaces and their drive to seek a hidden, dark escape box. The procedure covers pre-experimental habituation, multi-day acquisition training trials to evaluate learning, and a final probe trial to assess long-term spatial memory retention. This method is a robust, dry-land alternative to the Morris water maze, minimizing stress associated with water immersion.

Setup

- 1 Have a clear open space in the middle of your behaviour room to work with.
- 2 Tear napkin pieces into small squares to prepare for escape box covering
- 3 Organize images (cross, triangle and circle) on the walls adjacent to maze apparatus. Ensure they are all at the same x and y plane.
- 4 Be sure to secure maze to ensure it does not shift during experiment
- 5 Obtain a cup that is wrapped in napkin or paper to allow for a dark enclosure for mice.
- 6 *Allow for one hour period for mice in the testing room before each phase of the test each day.
*Cover home cages with trash bags or a blanket to keep it dark, leaving the front of the cage exposed to light.
- 7

HABITUATION PHASE, Day 1 (no time limit, use home cage bedding):

- 8 Check positioning of escape box on barnes maze apparatus and place home cage bedding inside.
- 9 Place. Mouse in center of maze covered by dark cup.
- 10 10 seconds after placement, allow mouse to freely roam the maze for an undefined amount of time until they find and independently enter the escape box.
- 11 If mice do not find the escape box, gently guide mouse into the box and allow to sit in there for approx 2 min
- 12 Upon entry, cover the escape box hole with a napkin for at least 15 seconds.



- 13 When removing, carefully and quietly remove the escape box with mouse from the apparatus and place the box in the cage to allow mouse to re-enter home cage.
- 14 Continue the above steps for each of your trials. Clean apparatus and box with 2% chlorohexadine disinfectant.

ACQUISITION PHASE, Days 2-6 (5 days)

- 15 After each trial in this phase: include clean, fresh bedding in the escape box to encourage entry into the tunnel, without them having to rely on a scent.
- 16 For each trial, there will be one, 5-minute session: place mouse in center of apparatus covered by dark cup.
- 17 If mouse does not find nor enter the escape box after 5 minutes, gently guide it towards the direction of the tunnel where they should enter on their own.
- 18 Upon entry, cover the escape box hole with a napkin for at least 15 seconds.
- 19 Carefully remove escape box from maze apparatus and gently place in the home cage of the mouse to allow it to re-enter cage.
- 20 Continue the above steps for each of your trials. Clean apparatus and box with 2% chlorohexadine disinfectant.

PROBE TRIAL, Day 9 (1 day, after two rest/off days)

- 21 Ensure maze is in position so that the holes are in the same position as during the training/acquisition days.
- 22 Remove escape hole from apparatus for the duration of the trials.
- 23 For each trial, there will be one, 90second session: Place mouse in center of apparatus and allow to roam for duration of session.



- 24 Return mouse to home cage.
- 25 Continue the above steps for each of your trials. Clean apparatus and box with 2% chlorohexadine disinfectant.