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Open Field

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Protocol status: Working

We use this protocol and it's working



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Abstract

The following protocol is used to assess gross motor and anxiety-related behaviors using the Open Field Test at the University of Alabama at Birmingham's Neuroscience Behavioral Assessment Core.

Guidelines

Use of this protocol needs approval by user's Institutional Animal Care and Use Committee (IACUC) or equivalent ethics committee.

Materials

1. Chlorhexidine
2. Paper towels
3. Open field boxes (appropriately sized for test subject)
4. Ethovision version 17 software

Before start

You will need a test mouse whose behavior is not included in the experiment to calibrate detection settings.

Set Up

- 1 Bring animals into testing room for habituation (at least 30 minutes prior to start of experiment)
- 2 Set up Maze
 - 2.1 Set up the number of open field boxes you want to use (6 fit well under our camera setup)
 - 2.2 Clean the maze
 - spray with chlorhexidine and wipe out
- 3 Plug the Ethovision version 17 software dongle into the computer
 - shines a red light when working
- 4 Open EthoVision version 17 software
- 5 Start a new experiment
 - 5.1 Name the file
 - 5.2 Indicate where the file will be saved
- 6 Experiment Settings
 - 6.1 Select live tracking
 - select the camera to be used
 - center your arena(s) under the camera
 - 6.2 Select center point detection
 - 6.3 Select the number of arenas you will be using

6.4 Units should be cm, s, and deg

7 Arena Settings

7.1 Grab a background image from the camera

- ensure the maze is in its intended area, moving it after this step will affect analysis
- mark placement with tape so the maze can be returned to the exact same location if accidentally moved between trials

7.2 Draw scale

- measure the width of one box
- draw a scale bar over the area measured and indicates its length (in cm)

7.3 Click the first arena in the list on the right

- select the subdivided rectangle drawing tool
- trace one box
- divide the rectangle into 4 rows and 4 columns
- drag the arena label inside the box
- repeat for all arenas

7.4 Create cumulative zones

- Inside - add the 4 boxes in the middle of the subdivided rectangle to this cumulative zone
- Outside - add the 12 boxes around the edge of the subdivided rectangle to this cumulative zone

7.5 Validate setup

- If you have any overlapping zones, they will be considered invalid and must be redrawn

8 Trial Control Settings

8.1 Begin condition should be on the start button with the condition of center point being in the arena

8.2 Stop track with the condition being after 5 minutes

9 Detection Settings



9.1 Click advanced settings

9.2 Dynamic response

9.3 Indicate if subject is darker or lighter than background

9.4 Place a test subject in the maze

9.5 Click and drag to create a box around the subject

9.6 The software will create an automatic threshold around the subject that moves with it

9.7 Finetune it with the scrub bar to make it fit perfectly around the subject

10 Trial list

10.1 Add the desired number of trials

Running the Experiment

11 Acquisition

11.1 Software will load the first trial

11.2 Put the subject(s) in starting arena(s)

- in the center of the arena
- keep starting area consistent across subjects



- 11.3 Press record
 - the program will run for 5 minutes and automatically stop
- 11.4 Remove subject(s)
- 11.5 Clean enclosure with chlorhexidine
- 11.6 Repeat for all trials

Results and Analysis

- 12 Data profile
- 12.1 Ensure that the software is pulling data from all trials
- 13 Analysis Profile
- 13.1 Distance traveled
 - Arena
 - Statistic - total only
- 13.2 Velocity
 - Arena
 - Averaging interval - 1
 - Statistics - mean
- 13.3 Acceleration
 - Arena
 - Statistics - mean
- 13.4 In zone
 - Select the inside and outside cumulative zones
 - Frequency
 - Latency to first
 - Cumulative duration
- 14 Results



- 14.1 Statistics and charts
- 14.2 Calculate to generate a file with the analysis parameters chosen
- 14.3 Export and save
- 15 Ensure the video files are exported and saved
 - analysis can be repeated from video files if a problem occurs

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