

Elevated Plus Maze

Take caution not to move the maze, as the tracking software has been precisely calibrated according to its position.

See manual (**Biobserve Viewer Manual.pdf**) for further information



Setup:

- 1) Switch on computer.
- 2) Open Viewer II.
- 3) Check that the “Current Configuration” displayed in the top right corner of the screen is **EPM_final.vcd**. If this is not the case, select the menu option *Configuration* → *Open Configuration...* and select the file.
- 4) Under the tab *Configuration* → *Experiment*, fill in the necessary information for the current experiment and subject (Rat ID# should go in *Animal No.*).
- 5) Specify the recording length under *Time controlled execution*, as well as whether you want a delayed start (Recording will begin *x* seconds after you click record).
- 6) To ensure the tracking zones and settings are correct, click the tab *Filters and Objects*. You should see a live video image of the maze, possibly with some small “false-positive” spots being tracked. If you see large false-positive spots (i.e. larger than the rat), the maze position or software parameters may need to be adjusted. See Troubleshooting for more information.

Recording:

- 1) Under the tab *Acquisition*, click the green play button. Recording will start and end according to the configuration settings you specified, unless you click the red stop button.
- 2) Once recording stops, a file save dialog will appear and prompt you for a save location.
- 3) After saving, you will automatically be brought to the *Data Analysis* tab, where you can view the data and save it as an excel file by clicking the corresponding button (See Fig. 1).

Analysis:

A data summary is produced automatically after each recording session. However, should you wish to reanalyze a previous recording using different parameters, you can do the following:

- 1) Ensure that the correct configuration is displayed in the upper right-hand corner of the screen. If the correct configuration is not displayed, select *Configuration* → *Open Configuration...* and select the file.
- 2) Fill in the information under the *Experiment* tab as described in steps 4-5 of Setup.
- 3) Under the *Camera* tab, check the box marked “Use movie” and click “Open...” to select the video file you wish to reanalyze.
- 4) Follow the steps above in “Recording” to analyze the file the same way you would analyze a live recording.

Clean-up:

- 1) Remove any feces and solid debris from the maze with a paper towel and deposit it in a waste bin.
- 2) Spray the maze with 30% isopropyl alcohol and dry it thoroughly with a paper towel.

Troubleshooting

Large “false-positive” spots are detected by the software

This is caused by oversensitive tracking parameters. In the “Filters and Objects” tab, decrease the “Background filter” sensitivity by moving the slider towards the left until the “false-positive” spots disappear or become sufficiently small. Be sure not to make it undersensitive or it will not detect the rat.

Although the software may continue to detect false-positive spots, as long as the number attached to the spot is not fluctuating, it will not recognize it as the rat. This number is the size in pixels of the tracked spot. If the size of the spot decreases below the minimum animal size, the software will stop measuring the size of the spot since it has concluded that it is too small to be the rat.

The software loses track of the rat and produces large “jumps” in the rat’s path

The tracking parameters may be undersensitive, particularly when the rat rears and presents a smaller profile to the camera. In the “Filters and Objects” tab, increase the “Background filter” sensitivity by moving the slider towards the right until the greatest amount of the rat’s body is detected without creating large false-positive spots. The minimum animal size can also be adjusted such that the software detects small profiles more easily. In “Animal filter”, decrease the minimum animal size by clicking the arrows or typing in a value. Use the aforementioned size in pixels attached to the rat’s detection spot to help you determine the minimum animal size. The best results can be achieved by adjusting the minimum animal size in concert with the background sensitivity.