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Open Field Behavior with head-mount V.1

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ABSTRACT

Mouse behavior testing open field protocol and calibration information for Tang et al 2023.

GUIDELINES

Full Calibration Information:

Calibration. To ensure sensor stability within sessions, several approaches were employed.First, a coated
mating sleeve was attached to the dual optic fiber cannula that sits immediately posterior to the sensor. The
sleeve was thickened with black tape to a desired outer diameter such that it stabilized the sensor in the
anterior-posterior direction. Second, the metal pins in the 4-position connector glued to the sensor were thickened with solder to stabilize their fit inside the receptacle connector in the skull cap. This protects against displacement in all directions. Third, stretchable black tape was wound around the base of the attached sensor and sleeve-covered cannula, further protecting against shifts in sensor positioning.

To control for possible variation in sensor positioning across sessions, a calibration approach was developed. Wireless inertial sensor was attached to individual isoflurane-anesthetized mice and the sensor was secured with the above strategies. Next, individual mice was placed in a custom-made calibration rig. The essential element of the rig is a vertical stainless-steel pole suspended above a stably secured table. In the setup used, the vertical pole was fixed to the horizontal edge of a vertically reversed "L" shape, stainless steel post assembly mounted on a breadboard (Thorlabs). The space between the lower end of the vertical pole and the table is enough for an individual mouse to slide underneath. The lower end of the vertical pole is fixed to a custom-made connector that resembles the connecting

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end of the fiberoptic patchcord. To perform calibration, individual isoflurane-anesthetized mice was securely attached to the vertical pole via a mating sleeve bridging the connection to the mouse's cannula implant. Next, replicate readings of the immobilized inertial sensor were made on Bonsai. Next, mice were attached to the experimental patchcord and allowed to recover in home cage for 20 minutes or until individual mice are clearly recovered and behaviorally active. Individual mice were then placed in open-field box for experimentation.

Calibration involves rotating all accelerometer and gyroscope readings from the inertial sensor by a rotation matrix such that the final gravitational field vector of the stationary sensor, when mounted on the mouse and fixed to the calibration rig, is in a universal frame of reference whereby there is zero vertical tilt. In other words, the only non-zero acceleration is on the universal z-axis (pointing down). To accomplish this, the accelerometer pitch and roll orientation angles of the fixed stationary accelerometer were determined and then applied to calculate the rotation matrix.The rotation matrix is multiplied by the sensor accelerometer and gyroscope readings to remove the stationary vertical tilt from the sensor. To account for possible drift in gyroscope baseline over time, a daily reading of stationary gyroscope baseline was made with a mock cement skull cap attached to the sensor just before the start of each experimental day. The baseline gyroscope readings were subtracted from all gyroscope values before the rotation matrix is applied to sensor data.

Habituation

1 One-month post-surgery, mice were habituated to head-mounted equipment over 2 days in their home cages.

2 On day 1, an actual or mock wireless inertial sensor (~2.5 cm H x 1 cm L x 0.5 cm W with ~ 2.5-3.0 cm antennae, ~1.8 g weight) glued to the 4-position connector (Harwin Inc., M52-040023V0445) was attached to the implanted receptacle connector on the skull cap.

Individual mice roamed freely in the home cage for 1 hour.

3 On day 2, an actual wireless inertial sensor and mono fiberoptic patchcord (200/220 µm diameter, 0.22 NA; Doric DFP_200/220/900-0.22_2m_DF0.7-2FC) was attached to the skullcap via a mating sleeve.

Patchcords were attached to 1x2 fiber-optic rotary joint (intensity division, 0.22 NA; Doric, FRJ_1x2i_FC-2FC) and mice roam freelyin home cage for 1 hour.

Open Field Recording Day

4 Sensor/patchcord habituated mice were anesthetized by isoflurane, attached to equipment, subjected to calibration protocol (see guidelines)

5 Individual mice were placed in an open field box inside a sound insulated chamber and allowed to behave freely inside the box for 75 minutes.

5.1 The open field box is made of 410 x 400 mm grey opaque acrylicwalls and a 410 x 400 mm white matte acrylic base.

5.2 The wireless inertial sensor (~1.8 g in weight, WEAR wireless sensor v1.1; Champalimaud Scientific Hardware Platform) conveys motion information sampled at 200 hz (set on WEAR v1.3.2 software; Champalimaud Scientific Hardware Platform) to a receiver base-station (Harp basestation v1.1 or v. 1.2, Assembly v0, Harp v1.4, Firmware v1.5; Champalimaud Scientific Hardware Platform), which conveys the information to the experimental computer running a Bonsai script(Bonsai[45] editor v2.3.1,RRID:SCR_017218) to capture and record motion data and video information. Video was captured with a camera(Flea3 FL3-U3-I3Y3M(17450451), Point Grey Research)coupled to a 1/2" format lens (NMV-6WA, Navitar).