




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🌐 Handling and behavioral training protocol for in vivo electrophysiological and optogenetic manipulation of basal ganglia neurons in awake head-fixed mice

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ABSTRACT

This protocol details habituation of animals and behavioral training of animals for in vivo electrophysiology or optogenetic experiments.

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Habituation

- 1 1 week prior to in vivo electrophysiological recording, mice should be habituated to the experimental environment and the treadmill apparatus.
- 2 It is recommended that all behavioral experiments be conducted during the dark phase of the light-dark cycle.
- 3 Habituation proceeds in five sessions, typically on consecutive days. Each session lasts for 60 min except for the 1st day which lasts for 30 min. Headplate fixation and placement onto treadmill apparatus
- 4 Before and after any procedures, spray down working lab surfaces and equipment with 5% Nolvasan to disinfect the surgical area.
- 5 Place the mouse in an anesthetic induction chamber and lightly anesthetize with 2-3% isoflurane.
- 6 Transfer the lightly anesthetized mouse to the SpeedBelt linear treadmill apparatus. The treadmill consists of a fabric belt (60cm length x
- 7 5cm width) wrapped around ball bearing mounted pulleys that ensure low friction gliding support for the animal.
- 8 Gently raising the animal's head, firmly screw and secure the animal's headplate into the headpost holder brackets. The head-restraint assembly is custom made using components from Thorlabs and Luigs & Neumann for maximum stability and durability. The headplate should be raised approximately 2.5 cm (per 25g mouse) above the treadmill belt.

Habituation over 5 days

- 9 On day 1, allow the mouse to acclimate to the treadmill apparatus for 30 min. They will attempt frequent postural adjustments and the hindlimbs may slip off the side of the treadmill. If so, use your hands to gently reposition the mouse back onto the center of the treadmill. Some mice may continue to do this over the first couple of sessions, but it will diminish over time. Over the course of the 30 minutes, mice will calm down and may or may not attempt to ambulate on the treadmill.

- 10 On day 2-5, allow the mouse to acclimate to the treadmill apparatus for 60 min. Over these sessions the mice should transition from more random to organized and controlled movements. Early in habituation sessions mice may ambulate backwards. When they do this, pull the belt forward lightly until the mouse transitions to forward ambulating behavior. Some mice may continue to do this over the first couple of sessions, but it will diminish over time

Removal from treadmill apparatus

- 11 After the conclusion of each habituation session, unscrew the animal's headplate from the headpost holder brackets and gently place the mouse back in the home cage.
- 12 Clean the treadmill belt with soap and water
- 13 Replace treadmill belts regularly