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Unilateral intranigral AAV alpha synuclein mouse model

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ABSTRACT

This protocol details methods for the unilateral intranigral AAV alpha-synuclein injection

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KEYWORDS

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MATERIALS TEXT

Virus and reagents:

1. AAV alpha-synuclein, dilute the stock to 7.8×10^{12} gc/ml in sterile PBS. Viruses can be kept at 4C for 2 weeks and -80C for a very long time. Avoid freeze-thaw from -80C.
2. Avertin-HCl (2% 2,2,2-tribromoethanol and 1% amyl alcohol, at 4C, shielded from light), 0.15 ml/10 g body weight.

MOUSE SN injection

From λ AP +0.09 cmML +0.12 cm DV -0.44 cm

Mouse#	AP +0.09	ML +0.12	DV -0.44	Δ breg/ λ	Note

- 1 Clean all equipment and surgical tools with 70% ethanol.
Turn on the heating pad and cover it with a paper towel.
- 2 Clean and prepare injector needle.
Rinse 3x with 70% ethanol.
Rinse 3x with saline.
- 3 Animals: adult mice.
Coordinators: from lambda AP +0.09 cm, ML+0.12 cm, DV -0.44 cm
Injection rate: 12 μ l/hour, total 2 μ l in 10 min.
- 4 Anesthesia: IP animals with Avertin solution
15 ml/10 g body weight. After approximately 3-5 minutes animal should be completely anesthetized. Verify this by observing no response to tail pinch.
- 5 Fill the glass micropipette needle with virus solution (20 μ l) by drawing the syringe plunger slowly. You can see a line between mineral oil and the solution. Adjust the rate to 12 μ l/hour, volume to 2 μ l. Pre-run the micro pump until you see a drop of liquid in the needle tip. Resume

the volume to 2 μ l.

- 6 Make a midline scalp incision and identify both the bregma and lambda coronal sutures. Place animal on platform in prone position and place head in ear bars (ear bars should be aligned with the area just anterior to external ear). Place snout in incisor bar and tighten loosely. Equilibrate bregma and lambda by measuring height of each using the stereotaxic arm to lower cannula until it contacts the exposed skull surface. A difference of less than 0.02 cm is acceptable. Adjust by raising or lowering snout height using incisor bar (be careful not to tighten snout holder since it will force wrong head angle).
- 7 From lambda dial in the following coordinates: AP: +0.09 cm, ML: +0.12 cm, lower needle tip to exposed dura and dial in: DV: -0.44 cm.
- 8 Infuse AAV alpha synuclein, at a rate of 12 μ l/hour for 10 min (total 2 μ L). Wait 2 minute and slowly withdraw needle 1 minutes after the end of the infusion
- 9 Suture scalp and place animal in incubator for recovery from anesthesia.