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Stereotaxic viral injections and array implantation



In 1 collection

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ABSTRACT

We have developed a new micro-fiber array approach capable of chronically measuring and optogenetically manipulating local dynamics across over 100 targeted locations simultaneously in head-fixed and freely moving mice, enabling investigation of cell-type and neurotransmitter-specific signals over arbitrary 3-D volumes. This protocol includes the stereotaxic viral injections and array implantation steps. Please contact us (mwhowe@bu.edu) if you are interested in using this technique.

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viral injection

- 1 Mice were anesthetized under isoflurane (1-3%) and placed in a stereotaxic frame.
- 2 A large craniotomy was performed with a surgical drill (Midwest Tradition 790044, Avtec Dental RMWT) to expose the cortical surface above the striatum.
- 3 Mice were injected with AAVs to express genetically encoded proteins for optical measurements and manipulations. Virus (~200-800nL per site) was injected stereotaxically through a pulled glass pipette (diameter 30-50µm) at a rate of ~100nL/min.
 - **3.1** For striatum expression, virus was injected at 10-40 total locations chosen to maximize overlap with fiber positions.
 - For midbrain targeting, for one set of mice, we injected the 4 sites relative to bregma: AP = -3.05, ML = 0.6, DV = -4.6 and DV = -4.25; AP = -3.5, ML = 1.25, DV = -3.9 and DV = -4.5.
 - For a midbrain targeting in another set of mice, we injected at 3.07mm caudal to the bregma at 4 sites: ML = 0.5mm, DV =-4.00 mm and -4.25 mm, ML = 1mm, DV = -4.125mm and ML = 1.5 mm, DV = -3.8 mm below the dura.

array implantation

- The fiber array was then mounted in the stereotaxic and slowly lowered into position until one side contacted the skull surface.
 - **4.1** Precise targeting of the array and rigid placement into the stereotaxic is essential to avoid misplacement of fibers (which may result in tissue distortion) and tissue damage caused by lateral movements of the fibers.
- A thin layer of Kwik-Sil (WPI) was applied to seal off the exposed edges of the craniotomy, and then Metabond (Parkell) was used to secure the plastic frame to the skull.
- After this initial layer of Metabond solidified, a metal headplate and ring(Atlas Tool and Die Works, as in Howe and Dombeck, 2016; Dombeck et al., 2010) were secured to the skull with Metabond and the surface covered with a layer of blackened Metabond (carbon powder, Sigma).
- Finally, a small cylindrical plastic protective cap cut to extend just above the fiber bundle end was secured around the bundle and covered on the inside surface with blackened Metabond.