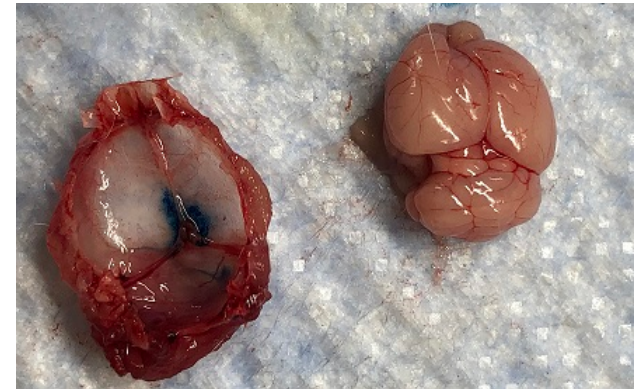
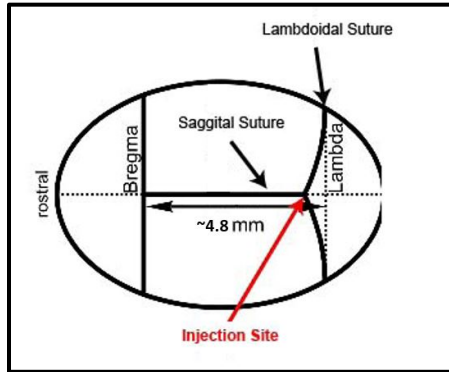
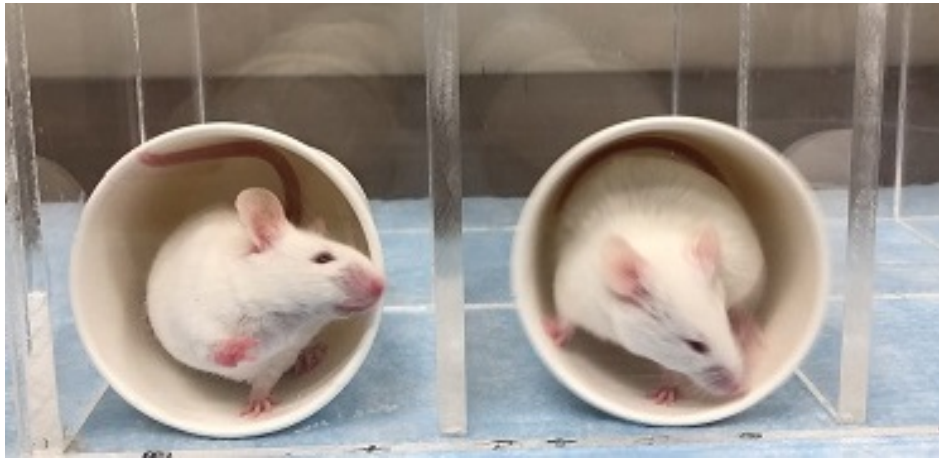
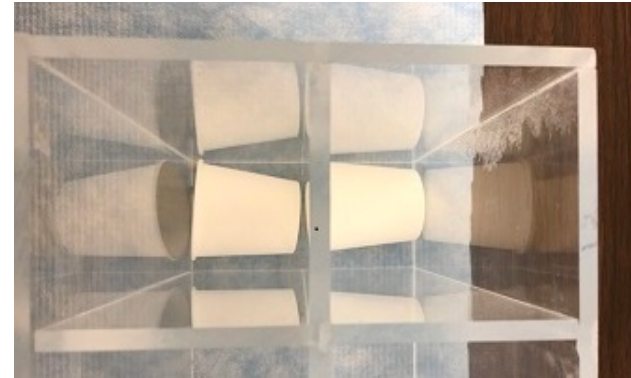
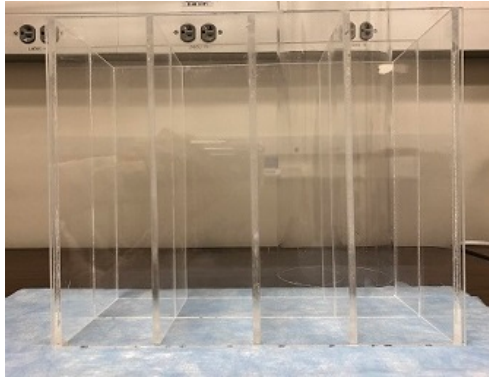




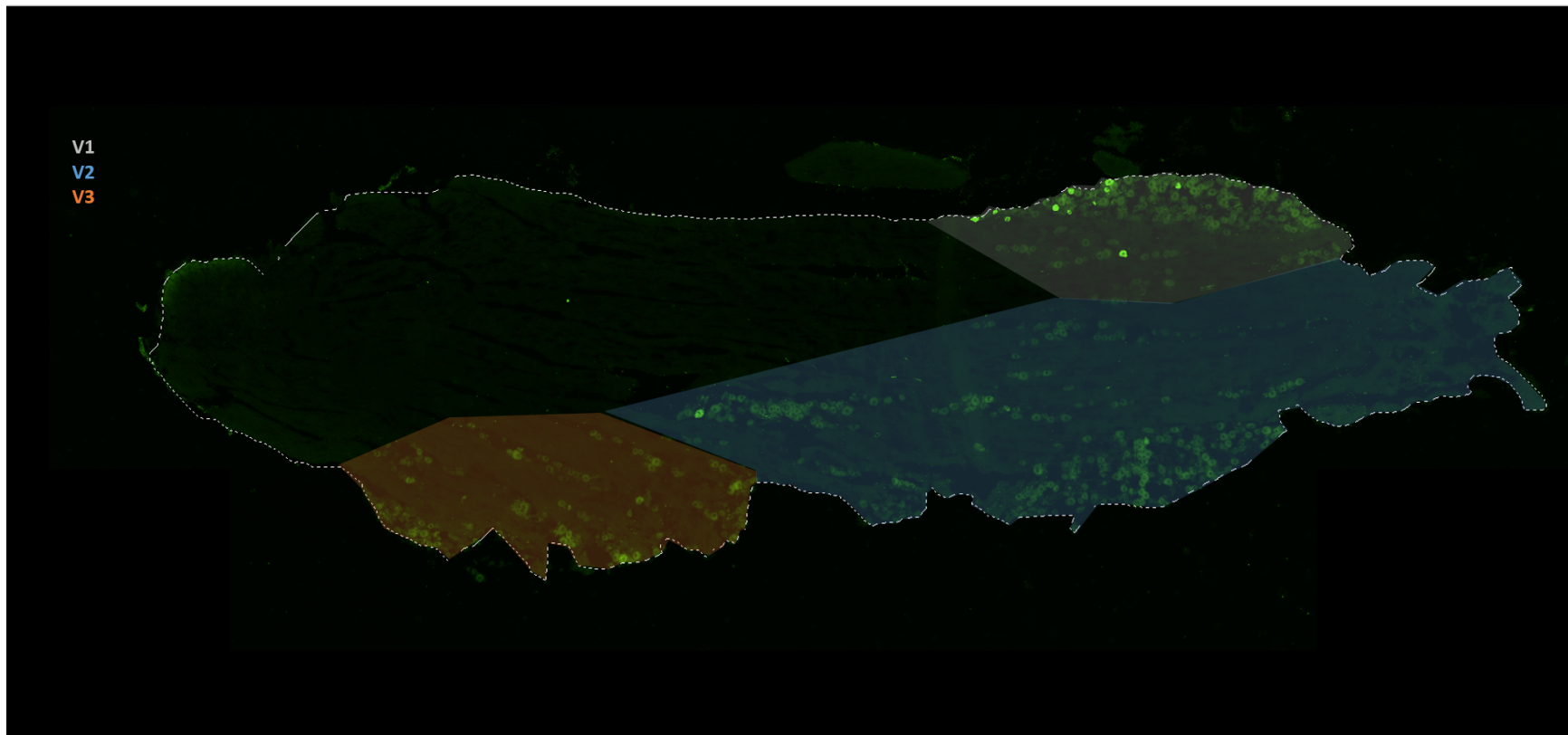
Supplementary Figure 1. Supradural injectors, tubing, and Hamilton syringe. (Left) Mouse dural injectors were modified from commercially available internal cannulas (Invivo1, part #C313I/SPC). The injector projection length (0.5mm to 0.65mm) varied depending upon the weight of each mouse. (Right) The modified injectors were attached to a 10 μ l glass syringe cemented needle (Hamilton Company, 700 series) via tygon tubing (Cole-Palmer, Item # EW-96460-16).



Supplementary Figure 2. Supradural injection placement. (Left) Diagram of the supradural injection site; ~4.8 mm posterior to bregma on the junction of the sagittal and lambdoid sutures. (Middle) Top view of a mouse skull after supradural injection of 5 μ l blue dye. The supradural injection was given under light isoflurane anesthesia to verify proper placement of the injector post-mortem. (Right) A mouse skullcap with blue dye observable on the dura and not on/in the cortex after supradural injection.



Supplementary Figure 3. Habituation chambers, cups, and testing boxes. (Top left and top middle) Side view of mouse habituation chambers (5cm length x 7.6cm width x 23cm height) without and with the paper cups used as facial testing chambers. (Top right) Top view of mouse habituation chambers with paper cups. (Bottom left) Mice habituating to the facial testing chambers (Bottom right) Mice in the facial testing chambers acclimating to suspended Plexiglas chambers (9cm length x 5cm width x 5cm height) with a wire mesh bottom (1 cm²) for mechanical testing.



Supplementary Figure 4. Retrogradely-labeled dural afferents in a trigeminal ganglion slice. 4% Fluorogold solution (Fluorochrome) was applied to the dura of 5-week old female ICR mouse via supradural injection in a volume of 5 μ l. 5-days post injection, animals were perfused with fixative (4% PFA, 12.5% Picric Acid) and trigeminal ganglia dissected and removed. Ganglia were sliced on cryostat in 15 μ m sections for staining. Slices were incubated overnight in primary solution of Anti-Fluorescent Gold Antibody (Millipore AB 153-I) at a dilution of 1:200 in 1% BSA. Secondary 488 anti-rabbit antibody (1:200) was incubated in 1% BSA for 2 hours at 37 degrees celcius. Images were acquired on an Olympus confocal FV1200 using Olympus Fluoview 4.2a software.