In vivo photopharmacology enabled by multifunctional fibers

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SUPPORTING INFORMATION

SUPPORTING FIGURES

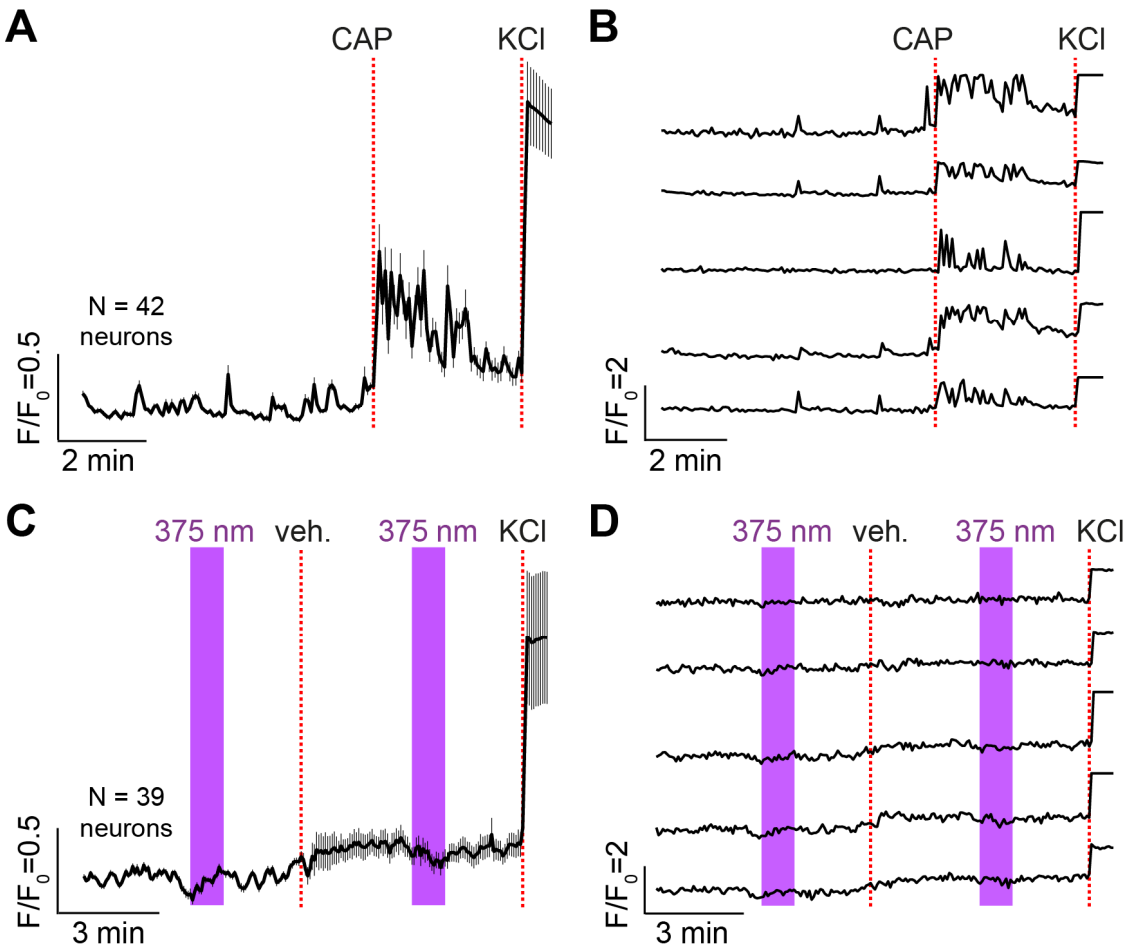

Figure S1. *red*-AzCA vehicle control experiments in cultured neurons. Fluo-4 fluorescence was recorded in cultured rat hippocampal neurons that had been transduced with LentiCaMKIIα::TRPV1-p2A-mCherry virus. (**A**,**B**) CAP addition (20 nM) increased intracellular Ca^{2+} levels. Displayed as (**A**) the average normalized fluorescence level (N = 42 neurons from 2 experiments) and (**B**) 5 traces from representative neurons (**C**,**D**) Addition of a vehicle control (0.1% DMSO, addn.) did not affect Ca^{2+} levels before or after 375 nm irradiation. KCl (25 mM) addition still increased Ca^{2+} levels. Displayed as (**C**) the average normalized fluorescence level (N = 39 neurons from 2 experiments) and (**D**) 5 traces from representative neurons. Error bars = mean ± S.E.M.

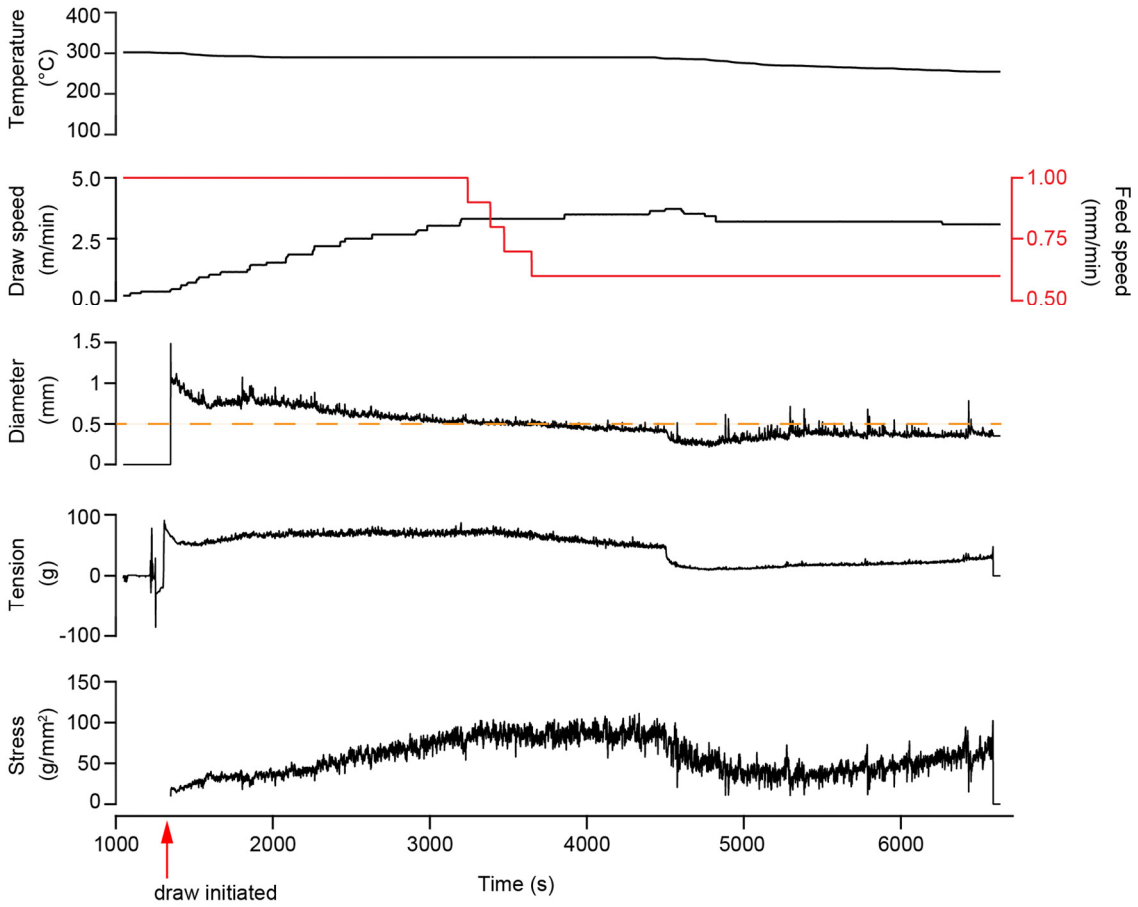

Figure S2. Drawing parameters and the thermal drawing process. The thermal drawing process was utilized to draw the macroscopic preform into a microscopic fiber. Displayed are the oven temperature, the draw/feed speeds, the final fiber diameter, and the resulting tension and stress values. The fiber used in this study was taken between t = 5000-6000 s.

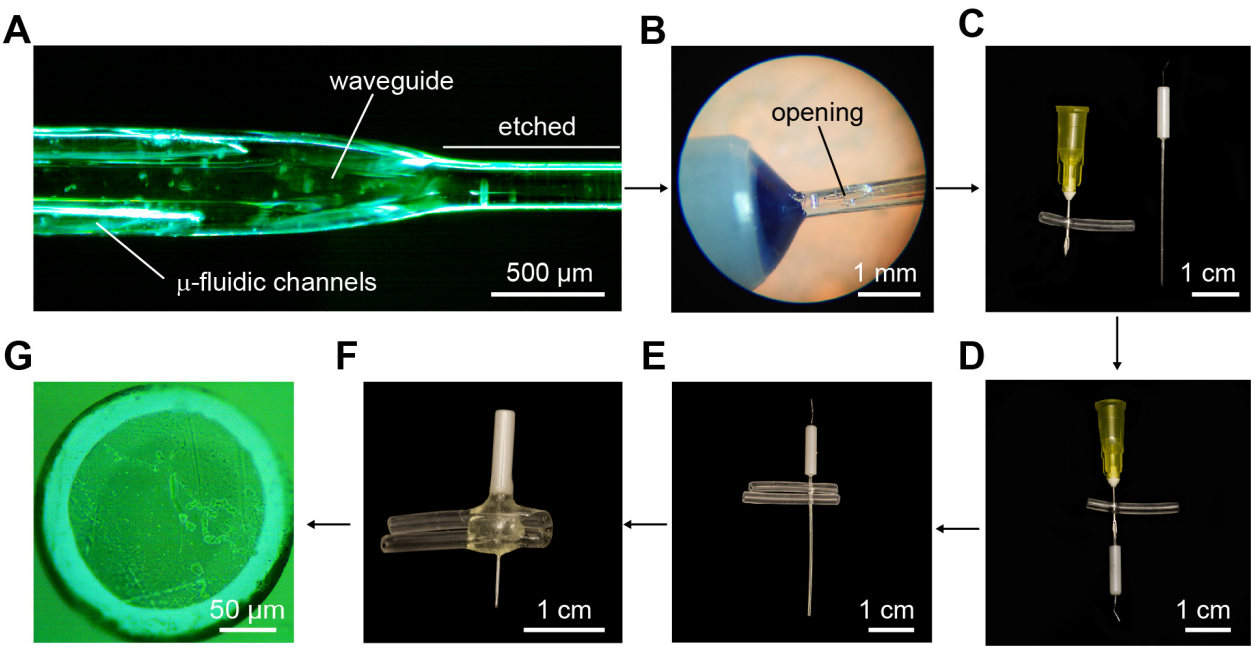

Figure S3. Fiber connectorization process. (**A**) One end of the fiber tip was etched in dichloromethane for insertion into the optical ferrule. (**B**) After gluing into an optical ferrule, the microfluidic channels were manually opened just below the ferrule. (**C**,**D**) A piece of tubing was pierced with a needle, through which the fiber was inserted to slide into the tubing. (**E**) Two pieces of tubing were positioned above the hole for each microfluidic channel. (**F**) The tubing and fiber were sealed with epoxy and (**G**) the ferrule was polished to afford a fully connectorized device (as shown in Figure 1C).

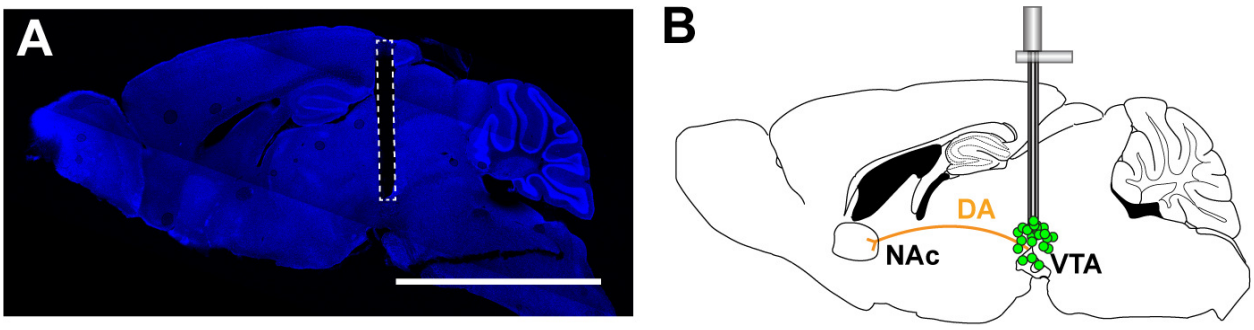

Figure S4. Implantation locations. (**A**) A representative mosaic fluorescence micrograph of a mouse sagittal brain slice (ML = 0.5 mm, 60 μm thick) stained with DAPI. The fiber implantation location in the VTA is depicted by the dotted line. Scale bar = 5 mm. (**B**) Schematic of all injection positions for the c-Fos immunofluorescence experiments, depicted as colored dots.

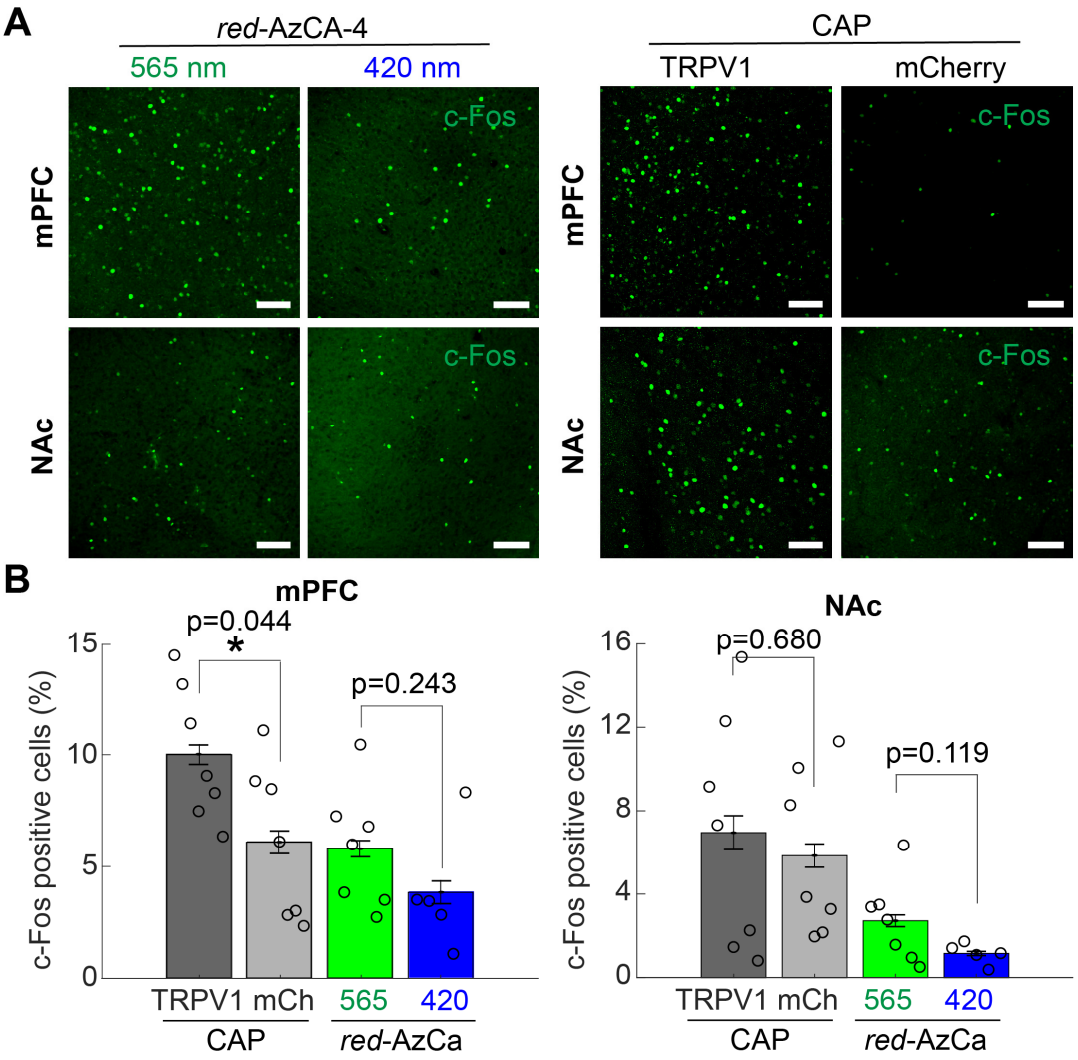

Figure S5. Chemogenetic control of VTA projections. *Red-AzCA-4* (3 μL, 1 μM) and CAP (3 μL, 10 μM) injection in anesthetized mice increased c-Fos expression in the NAc and mPFC. For *red*-AzCA-4, c-Fos was further upregulated in the presence of green (565 nm) light (N=7) compared to blue (420 nm) light (N=5). Similarly, CAP injection into mice expressing TRPV1 (N=7) upregulated c-Fos expression compared to control mice expressing mCherry only (N=7). Displayed as (**A**) representative images from fixed VTA slices, and (**B**) a quantification of the % c-Fos positive cells from multiple animals. Scale bars = 100 μm. Error bars = mean ± S.E.M.

Table S1: Corresponding primary and secondary antibodies for immunohistochemistry

Protein Target	Primary Antibody	Secondary Antibody
NeuN	Anti-NeuN Rabbit mAb; Abcam, #ab177487, lot #: GR3250076-4; 1:300 dilution	Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488; Invitrogen, #A-21206, lot #: 2045215; 1:1000 dilution
c-Fos	Anti-c-Fos (9F6) Rabbit mAb; Cell Signaling Technology, #2250s, lot #: [Ref: 09/2019 Lot:10]; 1:1000 dilution	Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488; Invitrogen, #A-21206, lot #: 1927937; 1:2000 dilution
TRPV1	Anti-Capsaicin Receptor Antibody, NT; Chemicon®, AB5889, lot #: 3022017; 1:1000 dilution	Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488; Invitrogen, #A-21206; 1:2000 dilution