Adapting and using a desktop computer numerical controlled (CNC) milling machine for automated cranial microsurgeries in mice

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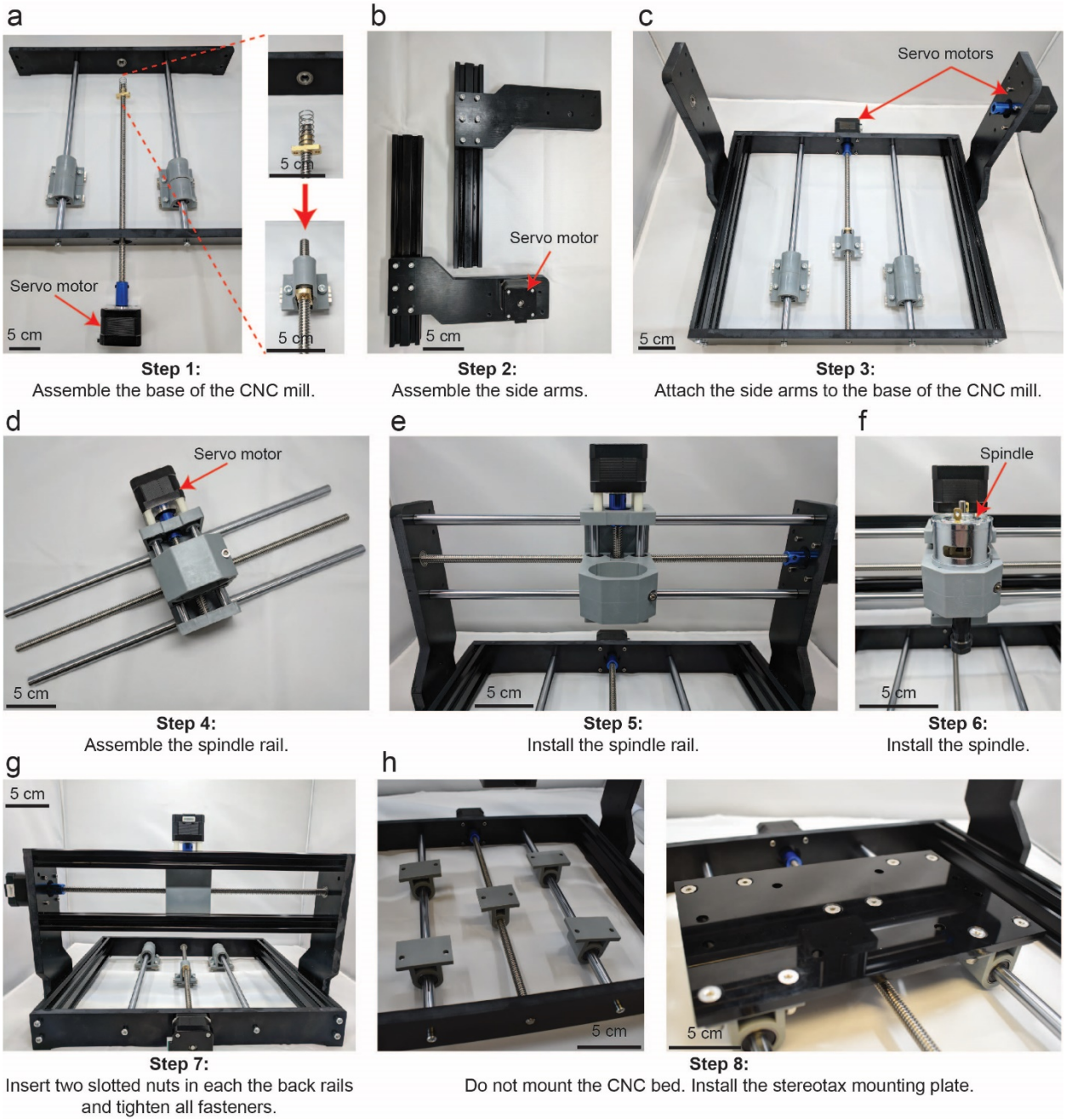
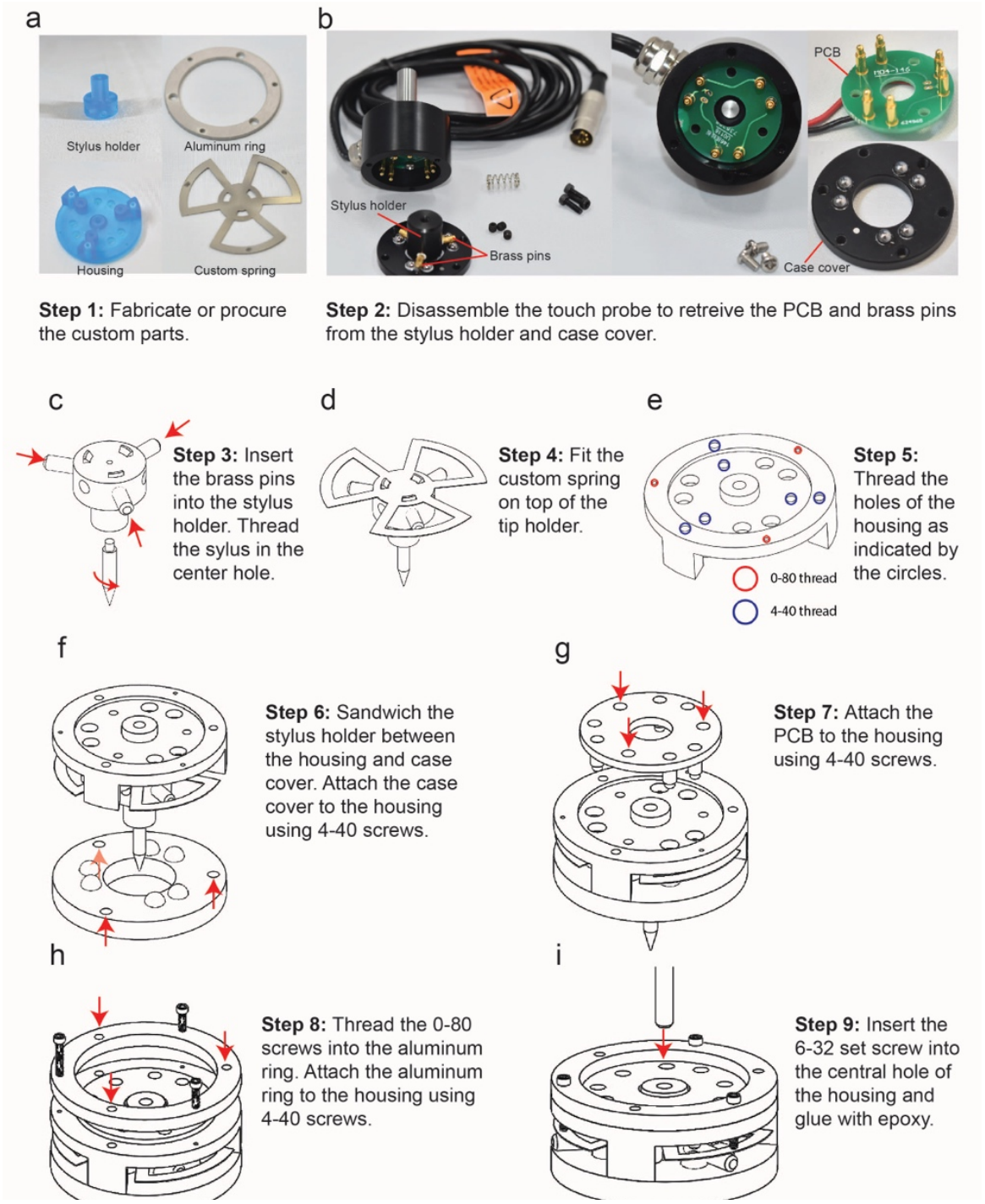
Supplementary Figures and Notes

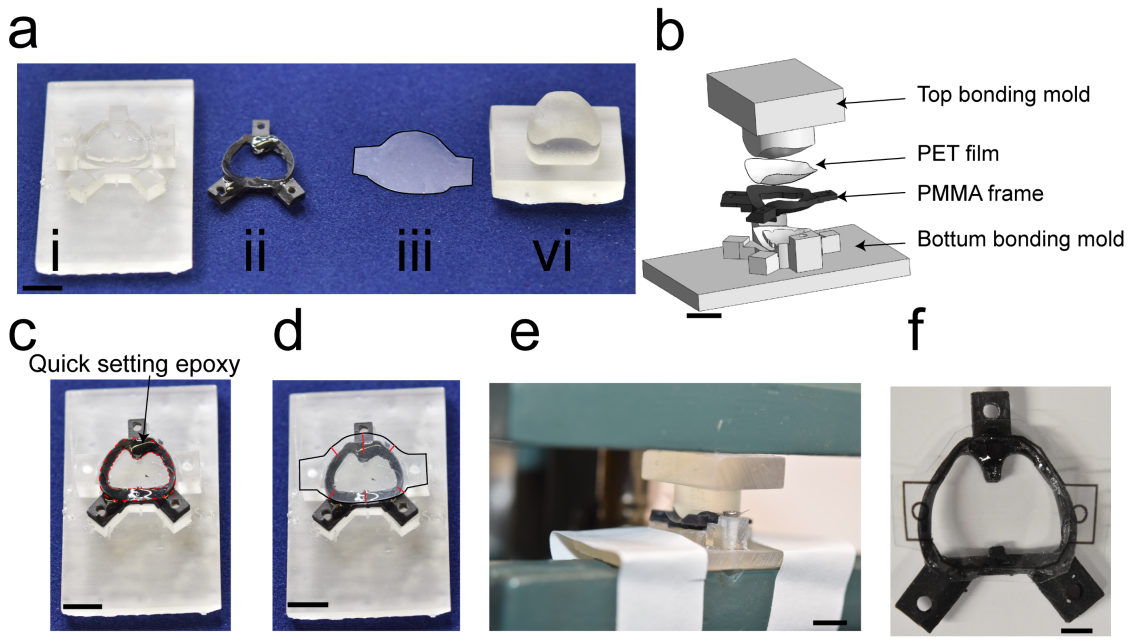


Supplementary Figure 1. Step by step illustration of the CNC mill assembly process. This figure depicts pictographic instructions for assembling the CNC mill and using the stereotax mounting plate. In step 1 **(a)**, the front and back parts of the bottom of the CNC mill containing the y axis are attached using the cylindrical rails and the ¼-20 threaded rods. The grey base plate holders with bushings should be attached to the rails as well. The y axis stepper motor can then be attached to one of the included threaded rods using grub screws and attached as well. The base plate holder should be fitted with an anti-backlash spring before fully assembling the base. In step 2 **(b)**, the vertical arms are attached to the sides of the base. When attaching them, ensure 3.75 cm of space from

the end of the extrusion. In step 3 **(c),** the sides of the base are attached to the base of the CNC mill. In step 4 **(d)**, the rail and threaded rod for the spindle and z-axis is assembled. Make sure to attach the anti-backlash screw the same way it is done for the stereotax holder in **(a)**. In step 5 **(e)**, the spindle rail is attached to the stereotax. The threaded rod should be fastened to the x axis stepper motor with the grub screws as well. In step 6 **(f)**, the spindle is installed in the z axis. In order to increase the working distance, the spindle should be installed in the highest possible position. In step 7 **(g)**, the final pieces of the body holding the vertical arms together are inserted. After ensuring every prior step has been completed correctly, all screws should be tightened. The final step, step 8 **(h)**, involves attaching the stereotax mounting plate.



Supplementary Figure 2. Step-by-step illustration of the surface profiler assembly process.



Supplementary Figure 3. See-Shell assembly and implantation: (a) (i) 3D-printed PMMA bottom bonding mold, (ii) 3D-printed PMMA frame of the See-Shell, (iii) PET film cut using a scissor around an outline roughly matching the PMMA frame, (vi) 3D-printed PMMA top bonding mold **(b)** CAD schematic of the 'sandwich structure' created when bonding the PET film to the See-Shell frame. **(c)** Photograph of the See-Shell frame mounted and the bottom bonding mold and coated with thin layer of epoxy. Dashed red borders highlight the area of epoxy application. **(d)** Photograph of the PET film aligned with the frame prior to bonding. Red lines indicate the location of relief cuts to assist with curving the PET film without strain. **(e)** Photograph of the sandwich structure held within a benchtop clamp during the bonding process. **(f)** Photograph of the See-Shell after bonding. The excess PET film surrounding the implant cut using a razor blade to realize final implant. Scale bars 2 mm. *Reproduced from Ghanbari et al Nature Communications 2019*

Supplementary Note 1

1. Use **Supplementary File 5** to 3D-print the See-Shell structural frame out of PMMA (RS-F2-GPBK-04, Formlabs Inc.) using a desktop stereolithography printer (Form 2, Formlabs Inc.).

2. Use **Supplementary File 5** to 3D-print two molds to assist with bonding the PET to the frame out of PMMA (RS-F2-GPCL-04, Formlabs Inc.).

3. Wash the 3D-printed parts with 100% isopropyl alcohol to remove uncured resin.

4. Tap three holes in the frame using a #0–80 hand tap (# 15J611, Grainger).

5. Clean the PET film using ethanol and low-lint cleaning tissue (KimWipes, Kimtech Inc.).

6. Use an erasable marker to create See-Shell frame outlines on the PET film (MELINEX 462, Dupont Inc.).

7. Cut The PET film along the drawn outline using a fine pair of scissors. Also, generate relief cuts to assist with curving the PET film without strain during bonding. Extension on the sides help with handling the PET film with forceps during assembly.

8. Pierce the PET film at the extended sides and secure it to the bottom molding bond using a #0-80 screw (**Supplementary Fig 3**). With a small piece of lab tape secure the other extended side of the PET to the back of the bottom mold to keep the PET out of the way of the frame.

9. Coat the bottom surface of the frame with quick setting epoxy (Scotch-Weld™ DP100 Plus Clear, 3 M Inc.).

10. Remove excess epoxy by pressing a piece of PET on to the frame and discard the PET film. With a toothpick remove any remaining excess epoxy.

11. Mount the frame on the bottom bonding mold (Supplementary. Fig 3b). Care should be taken to not to contaminate the PET on the side with the epoxy.

12. Secure the arranged components (bottom mold, frame and PET film) to a vice using lab tapes.

13. Gently press the top mold on to the PET to create a sandwich structure as shown in **Supplementary Fig 3**.

14. Close the vice for 5-10 minutes to allow epoxy to cure. Do not tighten the vice too much as that can cause deformation of the implant and damage it.

15. Open the vice and remove the top mold. Take the tapes off from the bottom mold and unscrew the PET film. Then gently remove the See-Shell from the bottom mold.

16. After 2-3 hours, use a pair of fine scissors or razor blades to cut the PET film surrounding edge of the frame.