HardwareX 13 (2023) e00391

Contents lists available at ScienceDirect

HardwareX

journal homepage: www.elsevier.com/locate/ohx

Hardware Article

An open-source head-fixation and implant-protection system for mice

Isaac A. Weaver [a], S. Aryana Yousefzadeh [b], Michael R. Tadross [a,*]

[a] *Department of Biomedical Engineering, Duke University, USA*
[b] *Department of Psychology and Neuroscience, Duke University, USA*

A R T I C L E I N F O

Article history:
Received 1 July 2022
Received in revised form 21 November 2022
Accepted 26 December 2022
Available online xxxx

Keywords:
Neuroscience
Neural implant
Headfix
Mouse
3D printed

A B S T R A C T

Mice are widely used in neuroscience experiments, which often require head-fixation and attachment of skull-mounted hardware. For many experiments, these components must remain intact over weeks to months, ideally with animals group housed. Many labs have designed *ad-hoc* head-fixation systems, which is an inefficient process. For example, when reinventing these solutions in our lab, we faced challenges with group housing, wherein mice would chew and damage implanted cannulas and electrodes of their cage mates. We performed several non-trivial design iterations to solve this problem, and present the most successful designs as an open-source collection. The designs include a standard mounting headbar compatible with most skull-mounted hardware, a snap-on protective mouse hat (headhat) to prevent mice from chewing the hardware, and a head-fixation station to facilitate common experimental procedures. We provide 3D-printing files, detail vendors and software used to make the components of the system, and provide editable design files for maximum flexibility to individual lab requirements.

Specifications table

Hardware name	*Mouse Headfix Hardware*
Subject area	• Neuroscience
Hardware type	• Imaging tools • Measuring physical properties and in-lab sensors • Other [Animal Handling and Head-Immobilization] • Other [Mouse Surgery Tools]
Closest commercial analog	There is no direct commercial analog to the hardware described here given the additional functionality our designs provide but the closest commercial analog for our head-bar would be “Head Plates” from Neurotar which retail for $225.50+: https://www.neurotar.com/product/headplates/

(*continued on next page*)

* Corresponding author.
E-mail address: michael.tadross@duke.edu (M.R. Tadross).

https://doi.org/10.1016/j.ohx.2022.e00391
2468-0672/

(continued)

Hardware name	Mouse Headfix Hardware
	Additionally, the closest commercial analog for our headfix setup is the "HeadFix" from Neurotar which retails for $9999.00:https://www.neurotar.com/product/headfix/
Open source license	CERN-OHL-P
Cost of hardware	$43.52 per headfix setup + cost of 3D printed components (variable)
Source file repository	https://doi.org/10.17632/tgggwcmhyz.2

Hardware in context

Head fixation is a common practice in neuroscience studies involving mice, as it facilitates several procedures [1]. Perhaps the most common reason for head-fixation is to facilitate optical or electrical access to neurons, while minimizing electrical noise or motion artifacts [2]. In addition, head-fixation is key to several behavioral paradigms that benefit from restricting the number of behavioral degrees of freedom [3]. These include behaviors involving defined motor skills [4], reward-associated learning [5], tasks that involve licking, eye movements, lever pulling, and virtual reality [6–8].

However, despite the broad usage of head fixation in neuroscience and related fields, no standardized head fixation system exists. Commercially available hardware is expensive and proprietary, making it unsuitable for customization or modification (e.g. Amuza, LabeoTech, Neurotar). The process of head fixing an animal requires an attachment to the head of the animal (here, referred to as a headbar) and an external apparatus that immobilizes the headbar and the head of the animal along with it. Many existing head fixation solutions have headbars that are wider than the body of the animal, which can impede their natural movements and make handling the animal difficult. To attach to these somewhat bulky headbars, often large platforms that have a clamping mechanism are employed. The complexity of these headfixing platforms can take longer than desired to headfix the animal.

In addition to animal immobilization, there is a need for a system to protect skull-mounted hardware. In particular, we have found that implanted cannulas and electrodes are often damaged by cage-mates. In many cases, this concern has necessitated single-housing mice to protect their hardware. However, single housing imposes social isolation stress and substantially increases cage costs. To our knowledge, no head fixation system with implant protection has been published.

Finally, in the broader category of head-fixation systems, very few examples are open source and those that have been published are not designed for more general neuroscience use—published variants are designed for either in vivo grin-lens imaging, or force detection of head-fixed animals [9,10]. Some neuroscience labs have developed their own head fixation systems but it is inefficient for each lab to re-invent the same technologies, and design idiosyncrasies can be a barrier to inter-lab collaboration.

Here we present a fully open-source 3D-printable head fixation system that affords simple head fixation of mice, along with implant protection. The system described is highly customizable and simple to use.

Hardware description

In contrast to existing head-fixation systems used for immobilizing animals, this system has been designed to minimize the headbar footprint on the head of the mouse and to offer neural implant protection with the headhat. Additionally, the dual headfix attachment forks offer straightforward rapid head fixation of the animal's head, allowing for maximum repeatability in many set-ups.

We offer two headbar-headhat combinations and a simplified headbar for maximum flexibility to the end user. All headbar designs are compatible with the same head fixation station. The system is designed to be low-cost, easy to use, immediately compatible with the majority of experimental workflows, and customizable for cases with particular constraints.

Ideal use cases and applicability to other researchers:

- The compact headbar design is less intrusive than commercially available, larger headbar designs. This allows mice to behave more naturally when not head-fixed, without sacrificing mechanical stability during head fixation.
- The modular headhat design protects skull-mounted hardware (e.g. cannulations or implanted electrodes), enabling mice to be group-housed without risk of implant damage by cage mates.
- The headfix station features a self-centering design via head forks, which facilitates use, and improves reproducible animal positioning.
- The design is scalable and customizable to specific laboratory needs.

The head-fixation system is comprised of three components: the heabar, the headhat, and the headfix station. All headbar designs are attached to the skull during stereotaxic surgery using adhesives described in the Build Instructions section of this article. Additionally, all headbar designs are compatible with the same head fixation forks. All components (including their dimensions) can be found as CAD files or engineering diagrams in the design files repository.

A description of each component is presented here:

- Headbars:
  - Headbar (V1.2): This is a simplified compact headbar design for mice. This design is solely for head fixation and does not have attachment points for a HeadHat. As such, this design offers no implant protection.
  - Headbar (V3.1): This is a compact headbar design with implant protection for mice. It has the same footprint on the skull as V1.2. However, V3.1 differs in that it has attachment points for headhat V3.1. This design is best for small implants that require protection (i.e. cannula guides). See Fig. 1B, 2A.
  - Headbar (V4.0): This is the large headbar design with implant protection for mice. It has a $\sim 20\%$ larger footprint on the skull than the other headbars. V4.0 has attachment points for headhat V4.0. This design is best for larger implants that require protection (i.e. implanted electrodes). See Fig. 1A, 2B.
- Headhats:
  - Headhat (V3.1): This is the implant protection hat compatible with headbar V3.1. The internal cavity of this headhat allows enough room for standard cannula guides (i.e. Cannula guides and tetrodes offered by *P*1 Technologies, Roanoke VA, including the MS333, C235, etc.—shown in Fig. 2C, 2E). Other cannula guides and other implants of similar size will fit under the headhat as well. See Fig. 1D, 2A.
  - Headhat (V4.0): This is the implant protection hat compatible with headbar V3.1. The internal cavity of the this headhat allows enough room for all 64ch probes offered under the NSF NeuroNex program by the Masmanidis lab[11] (shown Fig. 2D, 2F). Additionally, many commercially available electrode designs from NeuroNexus are compatible. See Fig. 1C, 2B.
- Headfix Base:
  - Headfix Mounting Hardware (V1.2): This is the plastic fork-like design in the headfix rig used to immobilize the headbar affixed to the mouse's skull. It is universal and will fit all headbar designs provided here (e.g. V1.2, V3.1, V4.0). See Fig. 3A, 3B.
  - Headfix Base Drill Pattern: This is the template for cutting and drilling the plastic sheet that forms the main structure of the headfix base.

Overall this head fixation system offers speed and ease of attachment. Moreover, all components are easily customizable to the needs of individual experiments.

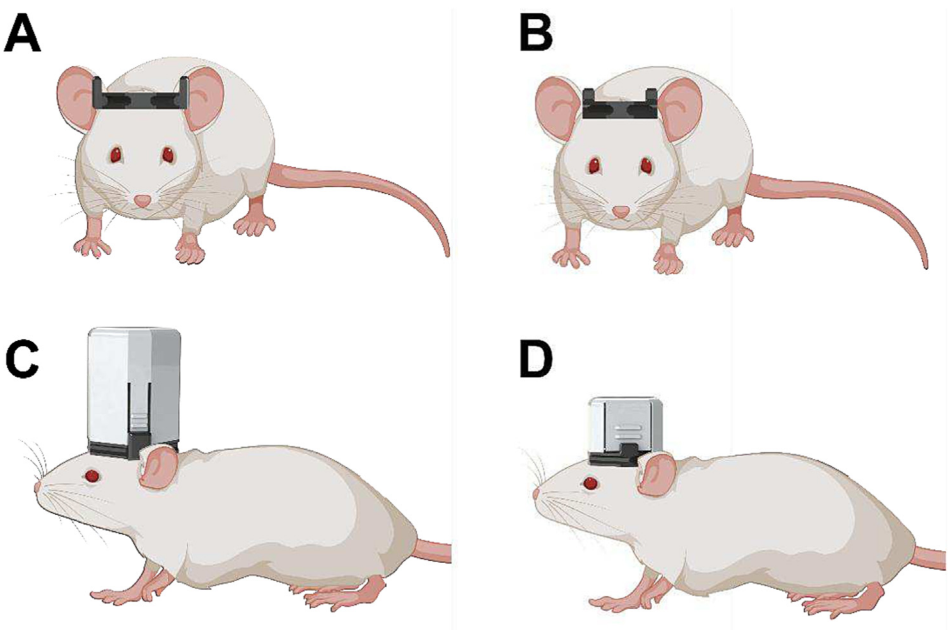

Fig. 1. Mice with headbar and headhat. A) Front view of mouse with headbar V4.0. B) Front view of mouse with headbar V3.1. C) Side view of mouse with headbar V4.0 and headhat V4.0 attached. D) Side view of mouse with headbar V3.1 and headhat V3.1 attached.

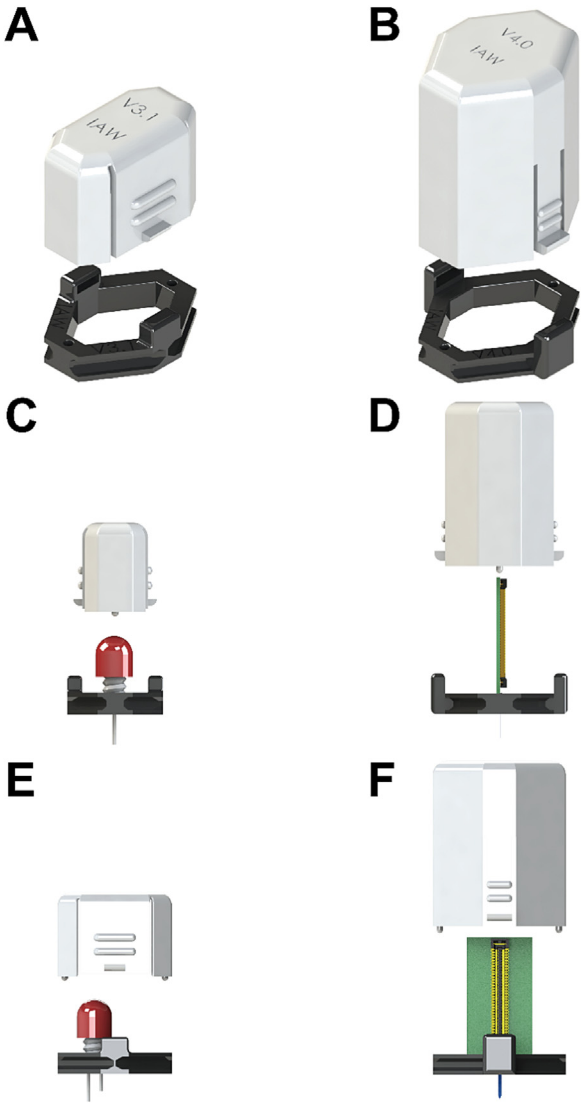

Fig. 2. Comparison of compact and large headbar and headhat systems: A) Headbar V3.1 and headhat V3.1 (the compact design combination) in profile view. B) Headbar V4.0 and headhat V4.0 (the large design combination) in profile view. C) Front view of V3.1 combination shown protecting a cannula guide. D) Front view of V4.0 combination shown protecting an implanted electrode. E) Side view of V3.1 combination. F) Side view of V4.0 combination.

Design files summary

Design file name	File type	Open source license	Location of the file
For example: Design file 1	e.g., *CAD files, figures, videos*	*All designs must be submitted under an open hardware license. Enter the corresponding open source license for the file.*	*Either enter the URL for the repository or the sentence: "Available with the article*".
----Design Files--------	----------	---------------------	--
Headfix Mounting Hardware V1.2	CAD	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
Headbar V1.2	CAD	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
Headbar V3.1	CAD	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2

(*continued*)

Design file name	File type	Open source license	Location of the file
HeadHat V3.1	CAD	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
V3.1_dimentions	File	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
Headbar V4.0	CAD	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
HeadHat V4.0	CAD	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
V4.0_dimentions	File	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
Headfix Base Drill Pattern	File	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
-------Use Videos----------	----------	----------------------	---
HomeCageVideo_Headhat_V3.1_wControlAnimals.mov	Video	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2
Headbar_Headhat_V3.1 CERN-OHL-P		_Headfix_Operation.mov https://doi.org/10.17632/tgggwcmhyz.2	Video
Headbar_Headhat_V4.0 CERN-OHL-P		_Headfix_Operation.mov https://doi.org/10.17632/tgggwcmhyz.2	Video
HomeCageVideo_Headhat_V4.0_Animals.mov	Video	CERN-OHL-P	https://doi.org/10.17632/tgggwcmhyz.2

Here we present a suite of open source, customizable head fixation designs with standard variants including head-fix hardware as well as implant protection systems. 3D printable files are included as both.stl (for easy upload and printing with no modifications) as well as.sldprt (for modification in SolidWorks). Please note that ALL headbar designs DO work with the same headfix Mounting Hardware, for ease of integration.

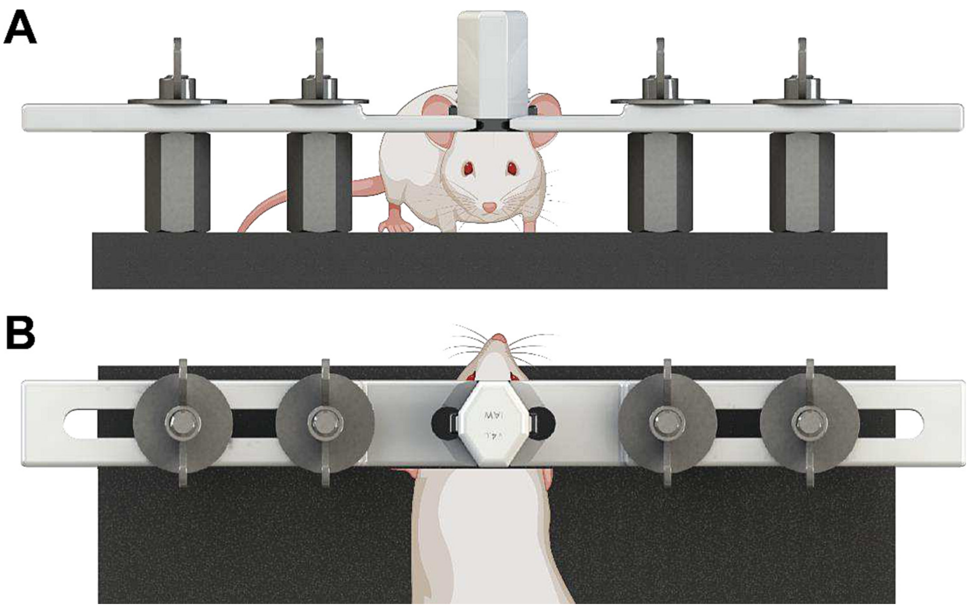

Fig. 3. Mouse with affixed headbar/headhat V4.0 immobilized in headfix base. A) Front profile of head fixed mouse using headfix mounting hardware V1.2. B) Top view of headfixed mouse.

In this file repository you will find:

1) "Headfix Mounting Hardware V1.2" – these files correspond to the fork-like design in the headfix rig used to immobilize the headbar affixed to the mouse's skull. This design works with ALL headbar variants (e.g. V1.2, V3.1, V4.0).
2) "Headbar V1.2" – these files correspond to the simplified headbar design that does not have attachment points for the headhat. This design offers no implant protection.
3) "Headbar V3.1" – these files correspond to a modified headbar design that does have attachment points for the V3.1 headhat. This headbar/headhat combination is appropriate for small electrode and cannula guide implants.
4) "Headhat V3.1" – these files are for the headhat V3.1 which clips onto the headbar V3.1 for the protection of implants
5) "V3.1_dimensions" – this is a.pdf file detailing dimensions of this headbar V3.1/headhat V3.1 combination.
6) "Headbar V4.0" – these files correspond to a modified headbar design that does have attachment points for the V4.0 headhat. This design has a larger footprint than the other variants. This headbar/headhat combination is appropriate for large electrode or other implants.
7) "HeadHat V4.0" – these files are for the headhat V4.0 which clips onto the headbar V4.0 for the protection of implants
8) "V4.0_dimensions" – this is a.pdf file detailing dimensions of this headbar V4.0/headhat V4.0 combination.
9) "Headfix Base Drill Pattern" – this is a.pdf diagram detailing the four holes that will need to be made in the plastic base during assembly of the headfix rig in order to headfix an animal.
10) "HomeCageVideo_Headhat_V3.1_wControlAnimals.mov" – this is a video taken showing an animal with V31 headbar/headhat exhibiting normal behavior in the home cage environment, with control animals for reference.
11) "Headbar_Headhat_V3.1 _Headfix_Operation.mov" – this is a video taken showing the operation of headfixing an animal with V3.1headbar/headhat and subsequent headhat removal and reattachment.
12) "Headbar_Headhat_V4.0 _Headfix_Operation.mov" – this is a video taken showing the operation of headfixing an animal with V4.1 headbar/headhat and subsequent headhat removal and reattachment.
13) "HomeCageVideo_Headhat_V4.0_wControlAnimals.mov" – this is a video taken showing animals with V4.0 headbar/headhat exhibiting normal behavior in the home cage environment.

Bill of materials summary

Designator	Component	Number	Cost per unit - currency	Total cost - currency	Source of materials	Material type
Headfix Base Sheet	Marine Grade Moisture Resistant HDPE Sheet, 12″ x 12″, 1/2″ Thick	1	$10.04* Two bases can be cut from one 12x12 sheet	$20.08* Due to minimum sheet size	https://www.mcmaster.com/9785T802-9785T212/	plastic
Hex Standoff	Male-Female Threaded Hex Standoff Aluminum, 1/2″ Hex Size, 7/8″ Long, 1/4″-20 Thread Size	4	$3.28	$13.12	https://www.mcmaster.com/93505A952/	Aluminum
Base Screw	18–8 Stainless Steel Hex Drive Flat Head Screw 82°Countersink Angle, 1/4″-20 Thread Size, 1″ Long	4 (out of 50 pack)	$0.2642	$13.21* Due to minimum package size	https://www.mcmaster.com/92210A542/	18–8 stainless steel
Wing Nut	Flanged Wing Nut Zinc Alloy, 1/4″-20 Thread Size, 1–1/8″ Wide	4 (out of 25 pack)	$0.61	$15.09	https://www.mcmaster.com/92239A200/	Zinc Alloy Steel
Headfix Tunnel	Impact-Resistant Polycarobate Round Tube	1	$4.14* For 4″ section	$12.42* Minimum 12″ length	https://www.mcmaster.com/8585K25-8585K251/	Plastic
Loctite	Threadlocker Loctite® 242, 0.34 oz. Bottle	1	$17.62	$17.62	https://www.mcmaster.com/91458A112/	Adhesive
82° fluted countersink bit	High-Speed Steel Countersink for Screws, 82°, 1 Flute, 5/8″ Body Diameter	1	$15.92	$15.92	https://www.mcmaster.com/2846A52/	Steel
Cyano-acrylate Adhesive	Quick-Set Instant-Bond Adhesive	1	$0.50* per surgery	$6.39	https://www.mcmaster.com/7625A16/	Adhesive
Dental Cement	Stoelting Dental Cement Kit	1	$1.00* per surgery	$75.00	https://stoeltingco.com/Neuroscience/Dental-Cement—UNITED-STATES-ONLY~9768	Adhesive
Optibond	Kerr Optibond Solo Plus Unidose Pakets (100 pack)	1	$2.00* per surgery	$199.95	https://ansondental.com/products/optibond-solo-plus-unidose-packets-package-of-100?variant=25743570689&msclkid=f9a9c735ae611e1e68d3a5096ecac7c8&utm_source=bing&utm_medium=cpc&utm_campaign=Shopping%20Standard%20Search&utm_term=4587780992124293&utm_content=shopping%20standard%20ad%20group	Adhesive

Given the minimum package quantities and raw material sizes, a single headfix base will cost $73.92 while building two headfix bases will cost $87.04, for a total of $43.52 each. Additional costs of thread locker and a countersink bit add $33.54 to this if you do not already have these materials. The last three items on this list (cyanoacrylate adhesive, dental cement, and Optibond) are the adhesives used for surgeries in our lab but may be substituted with standard adhesives used by other labs.

Build instructions

Headfix Base:

Cut the 12″ × 12″ × 0.5″ stock plastic sheet down to two 6″ × 7″ × 0.5″ blocks to form the plastic headfix Base Sheet. Drill four holes into the 7″ × 6″ × 0.5″ plastic headfix Base Sheet, following the "Headfix Base Drill Pattern" template. Countersink one side of each hole using the 82° fluted countersink bit (Fig. 4A). Add a drop of Loctite to four Base Screws, then slot the one screws into each hole in the headfix Base Sheet (Fig. 4B). On the other side, tighten the Hex Standoff down securely (Fig. 4C). Let the adhesive set overnight. 3D print two "Headfix Mounting Hardware V1.2" plastic pieces for each headfix Base being built. We recommend 3D printing this part using a high resolution, ABS-like material (i.e. Polyjet or SLA). Place the two "Headfix Mounting Hardware V1.2" plastic pieces on top of the posts with the fork ends of both pieces facing inwards, ensuring both threaded posts pass through the slots in the headfix Mounting Hardware (Fig. 4D). On top of the exposed threaded posts, add a Wing Nut, and tighten to secure the distance between the headfix Mounting Hardware Forks (Fig. 4E). Finally, make the headfix tunnel by taking the raw plastic tube and cut the 1.75″ diameter × 12″ long tube in half, lengthwise. Take each hemi-cylindrical section of the tube and cut into 4″ long sections (Fig. 4F). After a mouse has been head fixed in headfix base, the headfix tunnel will be taped over the body of the head fixed mouse (Fig. 4G, H).

Headfix Mouse Attachment:

The headbar and headhat can both be 3D-printed in the plastic of your choosing, though we strongly suggest using an SLA based printing technique as close to ABS plastic as possible. There is no assembly required. After repeated use, cosmetic wear is normal (Fig. 5).

During surgery on a stereotaxic apparatus, while the mouse is under anesthesia (in compliance with institution specific IACUC approvals), cut away all tissue from the skull below the desired attachment location of the headbar. Score the skull with a scalpel for good adhesion. Preform the intended surgery (implant, injection, etc) using the appropriate IACUC approved surgery. Ensure that the headbar will fit over top of any implant. In the case this is not true, be sure to place the headbar before the implant.

Continuing with the surgery, coat the exposed area of the skull with Optibond and allow it to dry—this creates a prepared surface to which later adhesives can form a secure attachment. Sterilize the headbar by quickly submerging in ethanol, then dry. Then coat the bottom of the headbar in cyano-acrylate adhesive and place the headbar on the skull in the desired location—with the clip attach points along the medial–lateral axis. The adhesive should hold within a few minutes.

Assuming all implants are in place and the surgery is at its end, fill the cavity between the skull and the headbar to the ledge with dental cement and let it cure. Make sure not to overfill this cavity as the headhat attachment points must remain clear. Following the surgery, let the animal recover as written in your protocol.

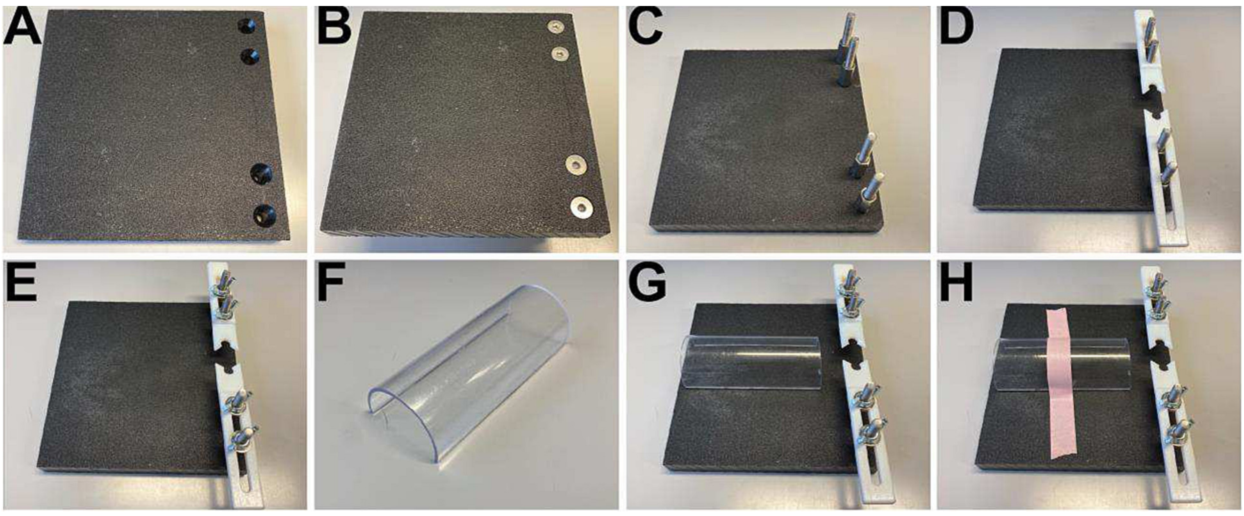

Fig. 4. Headfix Base Assembly.

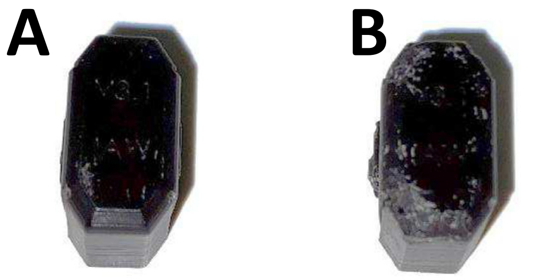

Fig. 5. Headhat V3.1 showing use. A) Headhat of a singly housed animal after use exhibiting minimal damage. B) Headhat after weeks of use on multiple animals in co-housing environment showing superficial damage that did not impact function.

When the animal is recovered and the glue is dry, align the headhat to the headbar, and press down to secure. The clip mechanism should engage and feel secure against the headbar. To release the headhat, press in on the side tabs and the headhat should come right off.

As in all animal handling, take the utmost care to ensure the safety and comfort of the animal and that this operation is in compliance with your IACUC approvals.

Operation instructions

The best practice for securing the animal in the head-fixation apparatus for any length of time is to do a series of habituation exercises. We suggest (at minimum) three habitation exercises before preforming any experiments or tasks. To do this (following all animal handling guidelines at your institution) gently place the animal on the headfix base. Pinching the anterior/posterior faces of the headhat between your forefinger and thumb, gently lead the animal until the octagonal headbar is roughly between the headfix mounting hardware forks of the headfix rig. Loosen the wingnuts on the HEADFIX BASE, slide the forks until they grab both sides of the headhat securely, and tighten the wingnuts. After a mouse has been head fixed in headfix base, tape a headfix Tunnel over the body of the mouse to make it more comfortable and restrict limb movement. For clarity, see "Headbar_Headhat_V4.0_Headfix_Operation" and "Headbar_Headhat_V3.1_Headfix_Operation" in the source file repository.

Validation and characterization

We have validated both headbar/headhat designs (V3.1 and V4.0) with mice under a variety of conditions. These designs are now the standard within our lab and have been used in over 200 implant surgeries. Within the lab we have validated that these hardware designs can accommodate cannula guide and electrode implants targeting many brain regions including superficial structures such as the cortex, dorsal striatum, and the hippocampus, and deeper structures like the basolateral amygdala, ventral tegmental area, and nucleus accumbens. Which areas of the brain other labs can target with these designs is highly dependent on their specific implant requirements, but V4.0 headbar/headhat has a very large open area to accommodate implants (see V3.1 and V4.0 dimension files in the design file database). In addition, the headbars can be implanted perpendicular to the anterior-posterior axis of the animal's head to allow access to more lateral structures (e.g., bilateral access).

In observing animals implanted with headbar/headhat V3.1 and V4.0, we did not observe a negative impact on animal health or ability to locomote. We show implanted animals exhibiting normal home cage behavior in videos uploaded to the source file repository entitled "HomeCageVideo_Headhat_V3.1_ wControlAnimals.mov" and "HomeCageVideo_Headhat_V4.0_Animals.mov." Moreover, we have validated that both designs work with industry standard mouse home cages from both Allentown (NextGen Mouse 500 system) and Techniplast (GM500 system). In both environments, the animals are able to access food (both on the floor and in feeding wirebars) as well as water (both from bottle and automatic watering). Most home cage enrichment is compatible with these designs with one notable exception: certain home cage enrichment systems that have small entrances or tunnels can be difficult for animals to access given the added height of the headbar/headhat system.

Affixing V3.1 or V4.0 headbar/headhat to an animal does increase it's weight. Post-op, the average weight increase of animals with V3.1 affixed is about 1.5 g, with about 1 g of that being attributed to the plastic headbar/headhat. In animals implanted with V4.0, the post-op weight goes up about 3.5 g, with just under 3 g of that being attributed to the plastic headbar/headhat. While this weight sounds large for a mouse, this is below 4 g weight limits established by other mouse head mounted implants [12]. As previously mentioned, we have not noticed a negative effect on the animal's health or ability to move—further demonstrated by videos in the source file repository entitled "HomeCageVideo_Headhat_ V3.1_wControlAni mals.mov" and "HomeCageVideo_Headhat_V4.0_Animals.mov." Nevertheless, there is additional torque on the animals head when lowering or raising its head—we have calculated the torque on the head (relative to bregma) to be 9.605×10^{-7} Nm for animals implanted with headbar/headhat V4.0 and 1.87×10^{-8} Nm for animals implanted with headbar/headhat V3.1, in both cases assuming angular acceleration of 0.25 rad/s. We have measured the temperature of the animal's head under the headhat and did not find the temperature to be above body temp—on average in animals with headbar/headhat V3.1 the difference in their head temperature versus their body temperature was 0.03 °C ± 0.31 °C, while in headbar/headhat V4.0 the difference in their head temperature versus their body temperature was −0.1 °C ± 0.63 °C.

In the weeks following the surgery, there is hair and tissue regrowth around the headbar but this has not caused any issues for the headhat attachment or for headfixing the animal. Both headbar/headhat configurations are effective in protecting implants in both single and co-housing environments. There have been instances of cage mates biting and chewing on each other's headbar/headhats, but in most cases the damage was superficial. In the few cases were the damage led to breakage of the headhats, we were able to replace the headhat with a new one. We have also noticed that approximately 10–15 % of the headhats come off after a few days. This is mainly observed when the clipping mechanism is not fully engaged, and replacing the headhat with a new one can resolve the issue. Therefore, we highly recommend that the animals are monitored routinely post-surgery.

Using V3.1 and V4.0 headbar/headhat combinations across 200 animals, we have found our system to be stable, robust, and easy to use. When preforming recording neural electrophysiological signals of headfixed animals, we observe very minimal movement artifacts. We have not attempted high resolution imaging through an implanted grin lens (or similar). Additionally, as a plastic head fixation system, we expect compatibility with ultrasound, CT (computed tomography), MR (magnetic resonance), and PET (positron emission tomography) scanners—though to date we have not tested our designs with these imaging tools.

Demonstrate the operation of the hardware and characterize its performance for a specific scientific application

- Neuroscience surgeries involving mice
- Easy Integration of Mouse Head Fix hardware into exiting surgeries
- Simple Headfix mechanism
- Easy to build and assemble
- Forgiving and flexible components

Ethics statements

All experiments involving animals were approved by the Duke Institutional Animal Care and Use Committee (approval number A113-20–05), an AAALAC accredited program registered with both the USDA Public Health Service and the NIH Office of Animal Welfare Assurance. Male and female animals were used in this setup. The sex had no impact on headfix hardware performance.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

Thank you to all members of the Tadross lab for their helpful feedback on head fixation designs and incorporating them into lab projects.

Funding: This work was supported by both the National Institutes of Health [Grant No 1DP2MH119425-01] and the Aligning Science Across Parkinson's (ASAP) Collaborative Research Network [grant number 020607].

References

[1] Z.V. Guo et al, Procedures for Behavioral Experiments in Head-Fixed Mice, PLoS One 9 (2) (2014) e101397.
[2] E.J.O. Hamel, B.F. Grewe, J.G. Parker, M.J. Schnitzer, Cellular level brain imaging in behaving mammals: An engineering approach, Neuron 86 (1) (2015) 140–159.
[3] N.J. Sofroniew, J.D. Cohen, A.K. Lee, K. Svoboda, Natural Whisker-Guided Behavior by Head-Fixed Mice in Tactile Virtual Reality, J. Neurosci. 34 (29) (2014) 9537–9550.
[4] D.H.O. Connor, N.G. Clack, D. Huber, T. Komiyama, E.W. Myers, K. Svoboda, Vibrissa-Based Object Localization in Head-Fixed Mice, The 30 (5) (2010) 1947–1967.
[5] W. Schultz, P. Apicella, E. Scarnati, T. Ljungberg, Neuronal activity in monkey ventral striatum related to the expectation of reward, J. Neurosci. 12 (12) (1992) 4595–4610.
[6] I. Ferezou, F. Haiss, L.J. Gentet, R. Aronoff, B. Weber, C.C.H. Petersen, Spatiotemporal Dynamics of Cortical Sensorimotor Integration in Behaving Mice, Neuron 56 (5) (2007) 907–923.
[7] M. Kislin et al, Flat-floored Air-lifted Platform: A New Method for Combining Behavior with Microscopy or Electrophysiology on Awake Freely Moving Rodents, J. Vis. Exp. 88 (2014) 1–11.

[8] M.J. Wagner, J. Savall, T.H. Kim, M.J. Schnitzer, L. Luo, Skilled reaching tasks for head- fi xed mice using a robotic manipulandum, Nat. Protoc. 15 (3) (2020) 1237–1254.
[9] R.N. Hughes, K.I. Bakhurin, J.W. Barter, J. Zhang, H.H. Yin, A Head-Fixation System for Continuous Monitoring of Force Generated During Behavior, Front. Integr. Neurosci. 14 (March) (2020) 1–11.
[10] P.A. Groblewski et al, A standardized head-fixation system for performing large-scale, in vivo physiological recordings in mice, J. Neurosci. Methods 346 (2020) 108922.
[11] L. Yang, K. Lee, J. Villagracia, S.C. Masmanidis, Open source silicon microprobes for high throughput neural recording, J. Neural Eng. 17 (1) (2020) pp.
[12] J. Voigts, J. Siegle, D.L. Pritchett, C.I. Moore, The flexDrive: An ultra-light implant for optical control and highly parallel chronic recording of neuronal ensembles in freely moving mice, Front. Syst. Neurosci. 7 (2013) 1–9.

Dr.Isaac Weaver, PhD recently completed his PhD in Biomedical Engineering at Duke University working on conductive polymer interfaces between neurons and micro-electrodes. He has a background in microelectronic device fabrication with an emphasis on biosensors and neural interfaces. He holds degrees in Biomedical Engineering, Physics, and Biological Physics.