Methods to Probe Brain Circuitries *in vivo*

Technical Journal Club

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MD PhD student
Adriano Aguzzi Group
23.01.2018

outline

- Scientific question and requirements
- Paper 1: Long-range population dynamics of anatomically defined neocortical networks
- Paper 2: Fully integrated silicon probes for high-density recording of neural activity
- Side by side comparison
- Concluding remarks and outlook

Scientific Question and Requirements

- Brain: different anatomical and functional areas
- Cortical and subcortical
- Extensive, often bidirectional connectivity

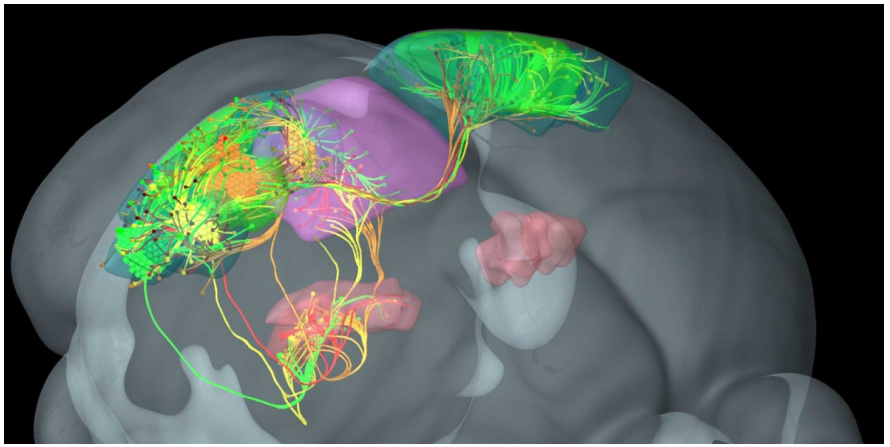
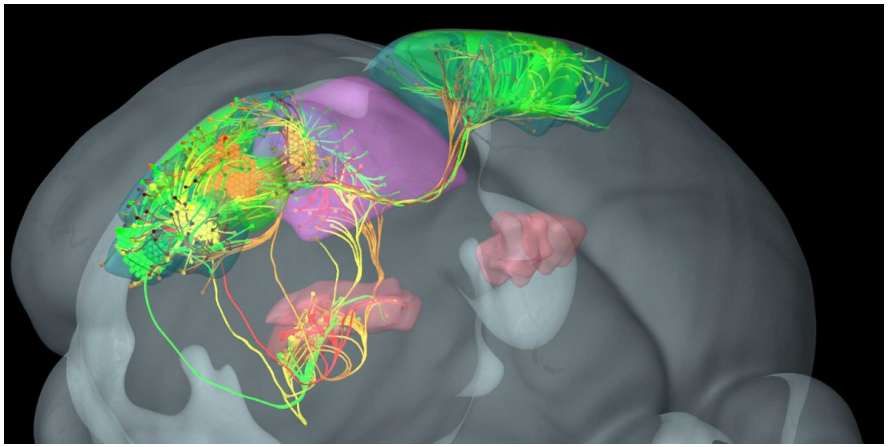


Image: Allen Institute for Brain Science

Scientific Question and Requirements

- Brain: different anatomical and functional areas
- Cortical and subcortical
- Extensive, often bidirectional connectivity

- Detect neuronal activity
- Multiple brain regions
- (Stable over time)



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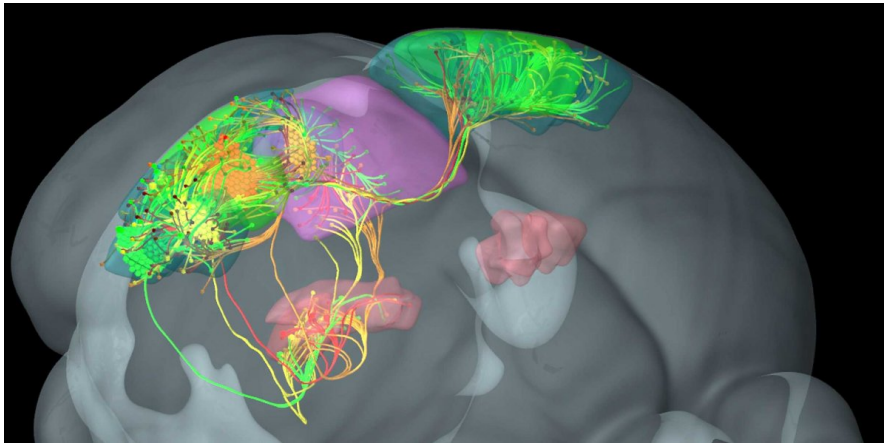
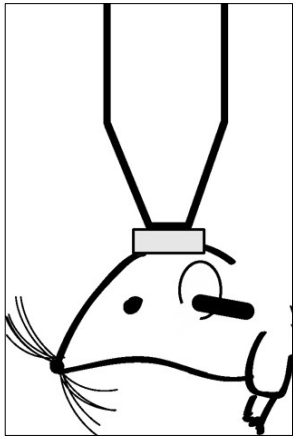
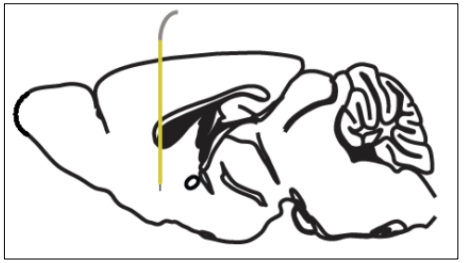


Calcium Imaging



Electrode Recording



Paper 1

NEUROSCIENCE

Long-range population dynamics of anatomically defined neocortical networks

Jerry L Chen, Fabian F Voigt, Mitra Javadzadeh, Roland Krueppel, Fritjof Helmchen
University of Zurich, Switzerland; University of Zurich, ETH Zurich, Switzerland

RESEARCH ARTICLE May 24, 2016

CITED 9 VIEWS 3,347 COMMENTS 1

CITE AS: eLife 2016;5:e14679 DOI: 10.7554/eLife.14679

Long-range population dynamics of anatomically defined neocortical networks: aim

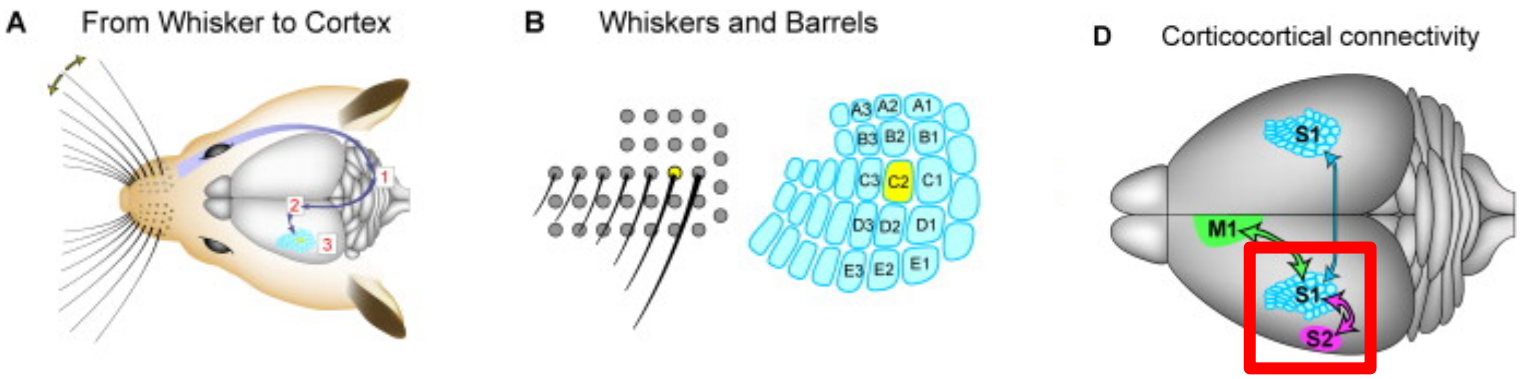
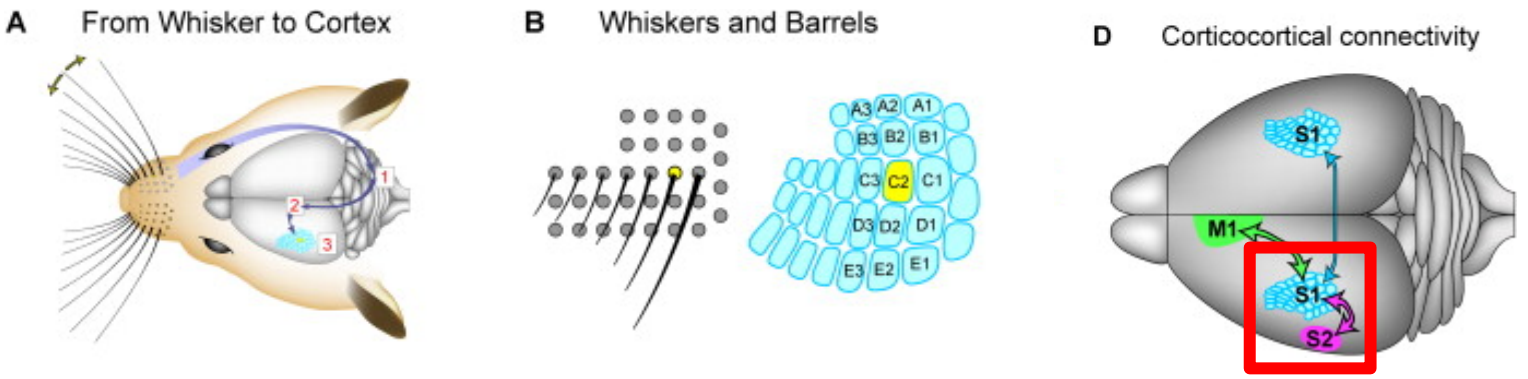


Image adapted from: https://doi.org/10.1016/j.neuron.2007.09.017

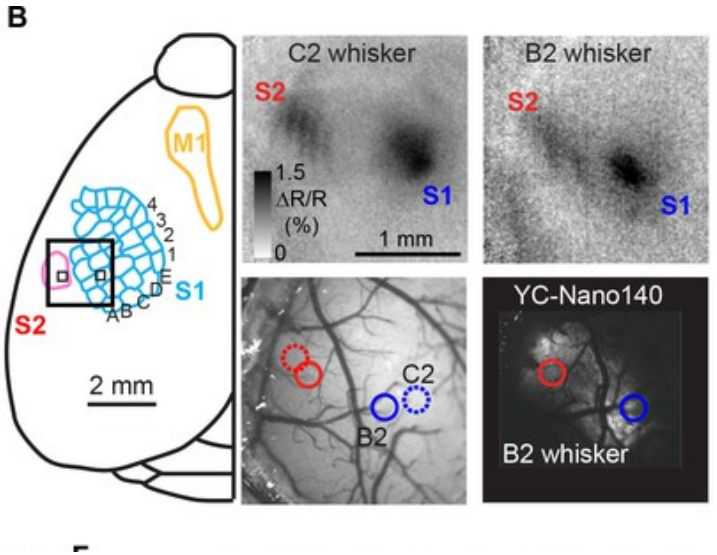
Long-range population dynamics of anatomically defined neocortical networks: aim

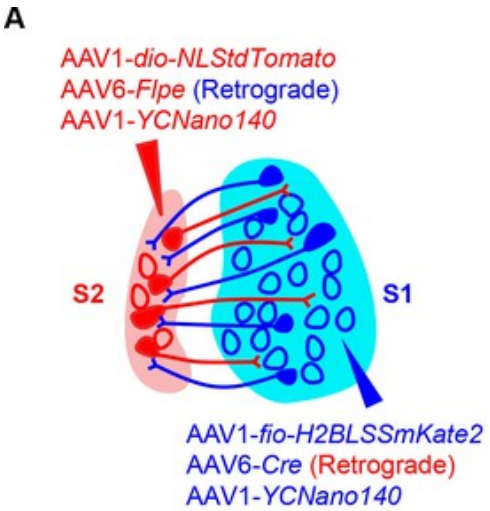


investigate direct interactions between S1 and S2 by simultaneously monitoring activity in feedforward neurons in S1 projecting to S2 (S1S2) and feedback neurons in S2 projecting to S1 (S2S1) in mice during tactile whisker behavior

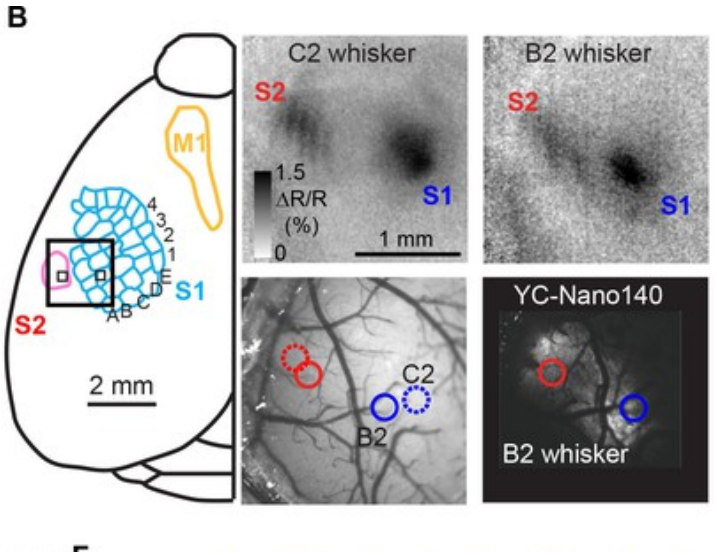
Image adapted from: https://doi.org/10.1016/j.neuron.2007.09.017

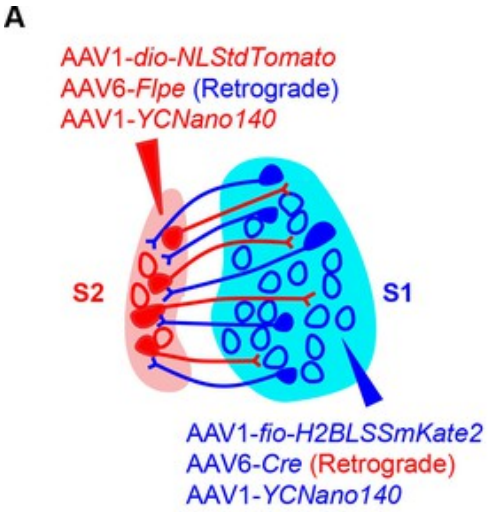
Long-range population dynamics of anatomically defined neocortical networks: experimental strategy

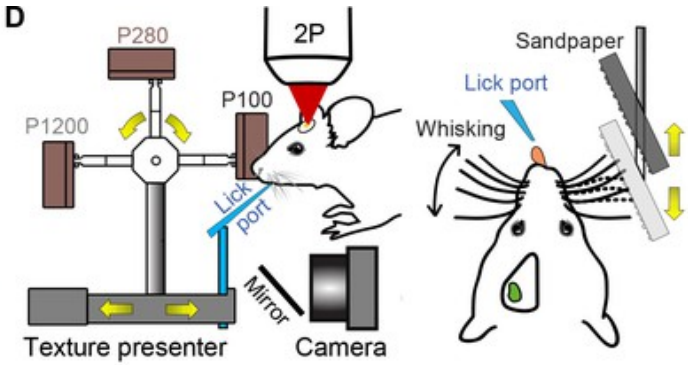


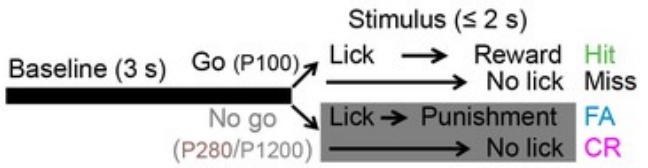


Long-range population dynamics of anatomically defined neocortical networks: experimental strategy

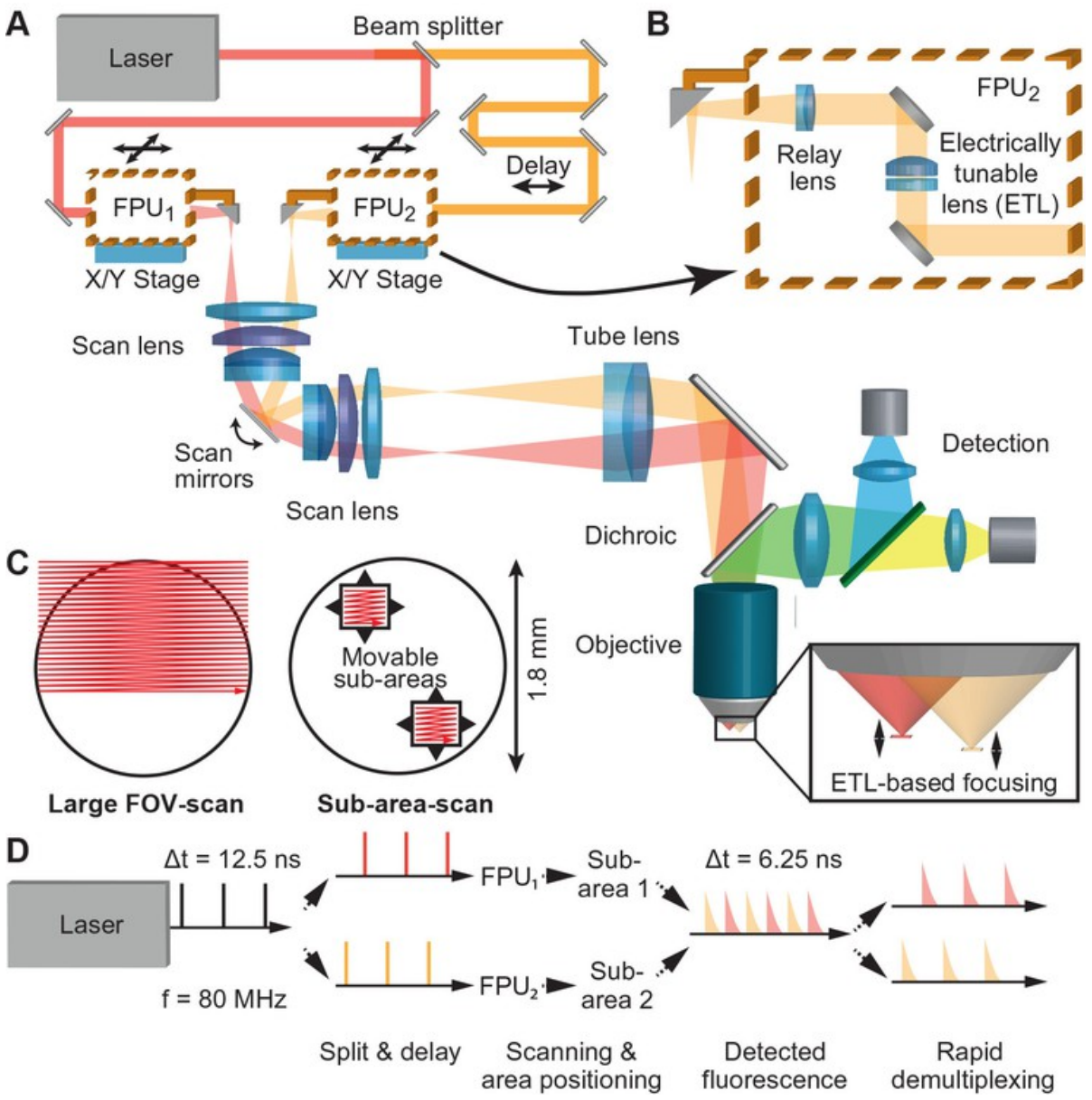








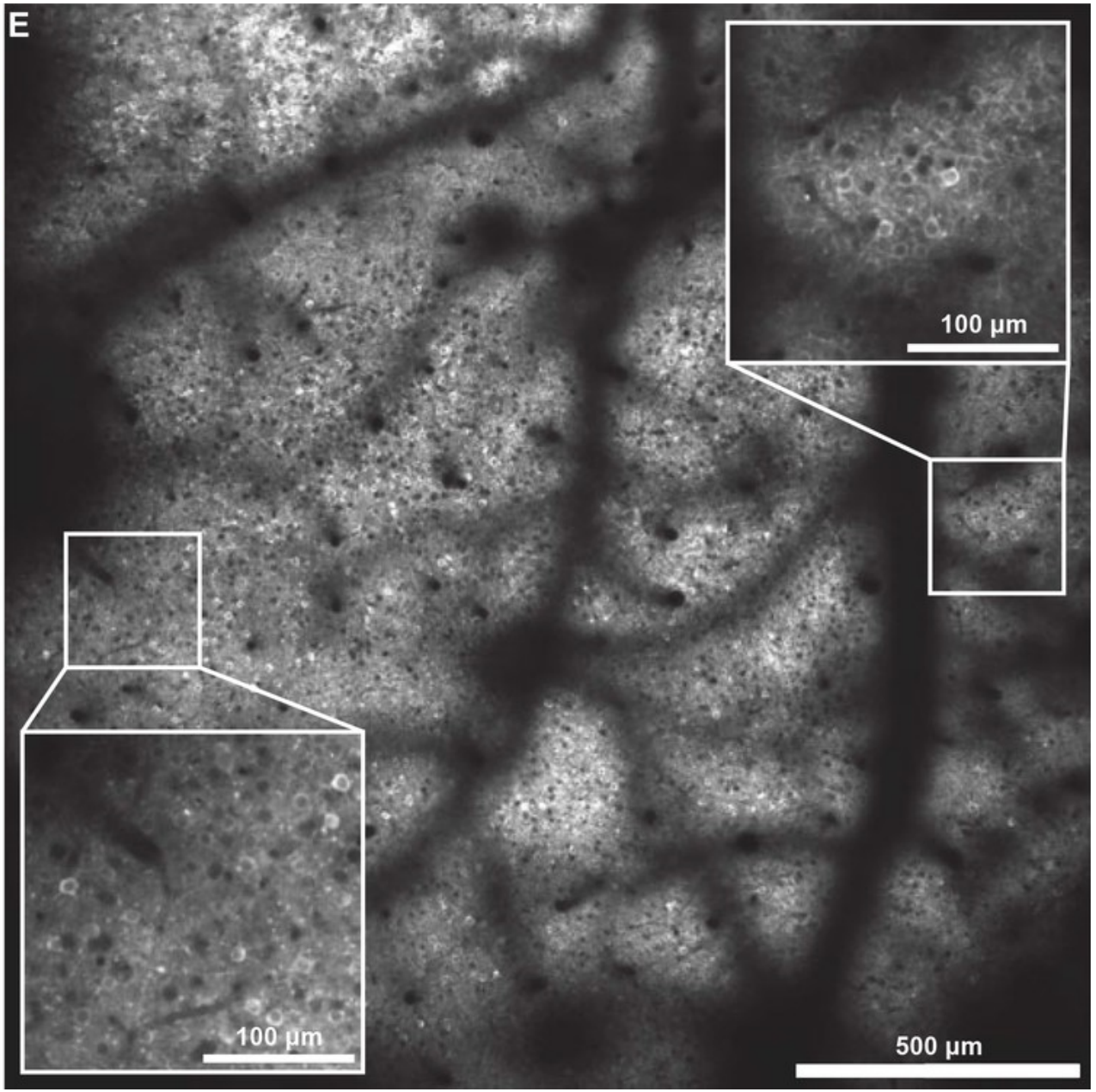
Long-range population dynamics of anatomically defined neocortical networks: microscope



Imaging rate: 7Hz

Combinatorial plane hopping: each FPU is independently refocused on one of three z-planes between trials

Long-range population dynamics of anatomically defined neocortical networks



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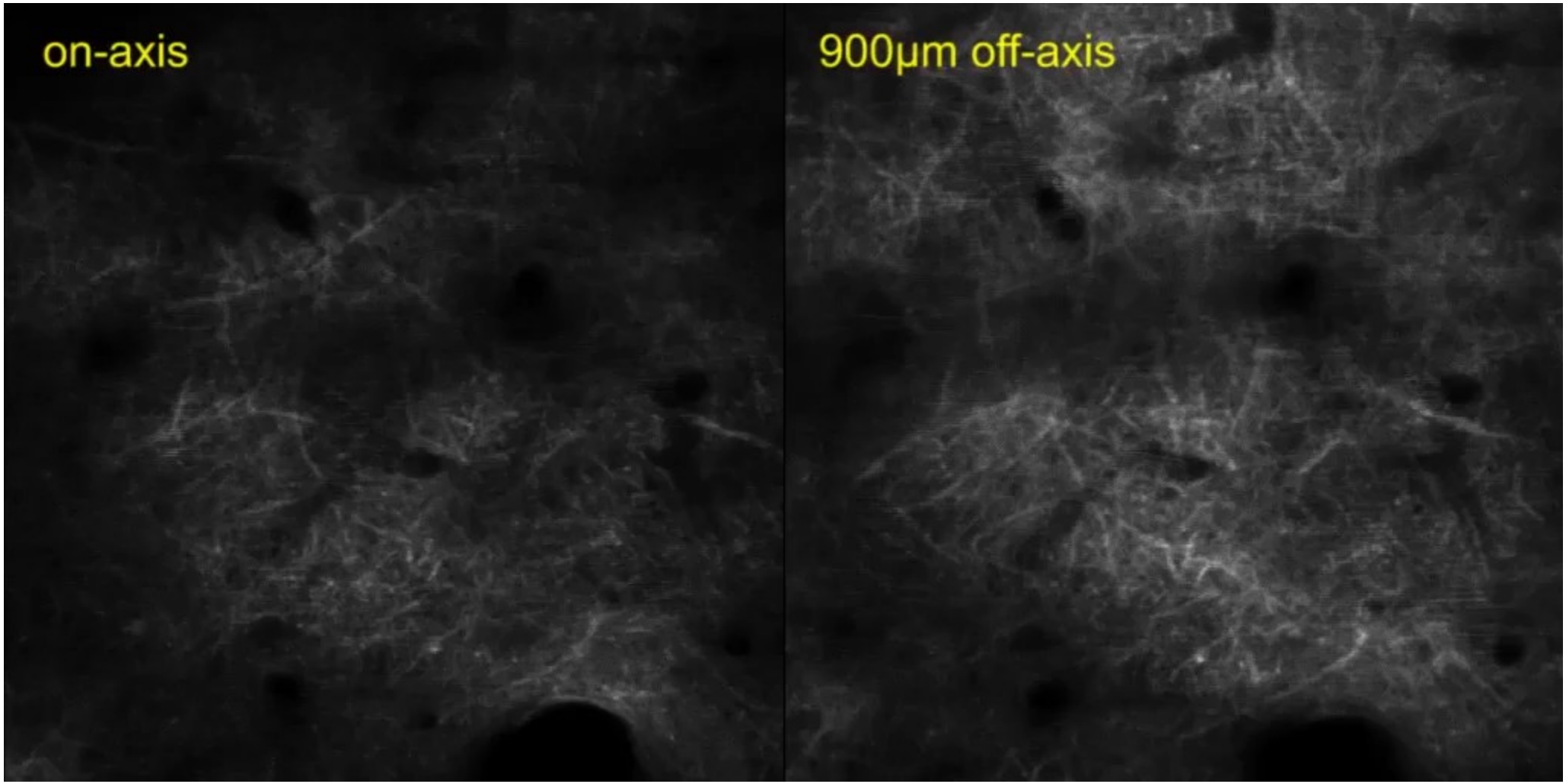

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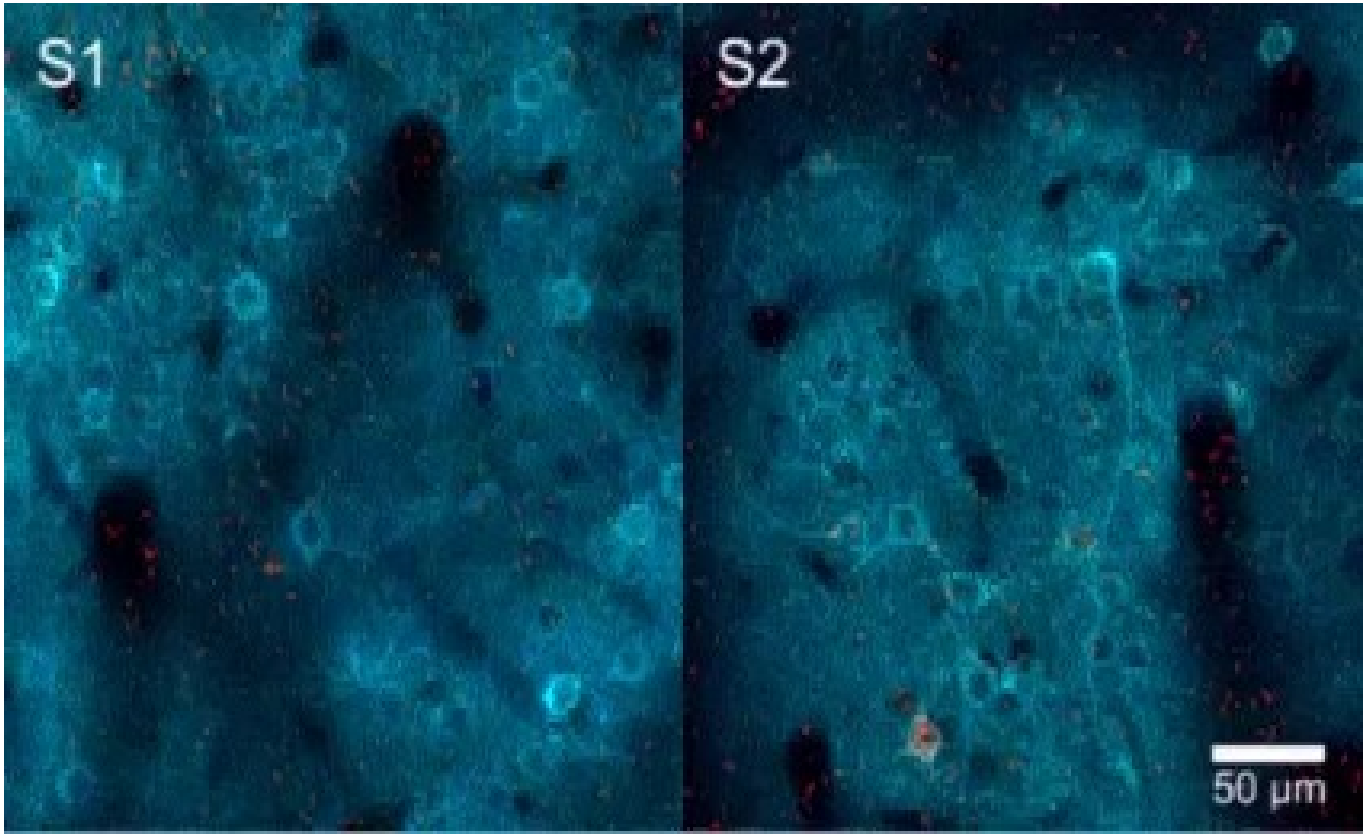


Long-range population dynamics of anatomically defined neocortical networks: analysis

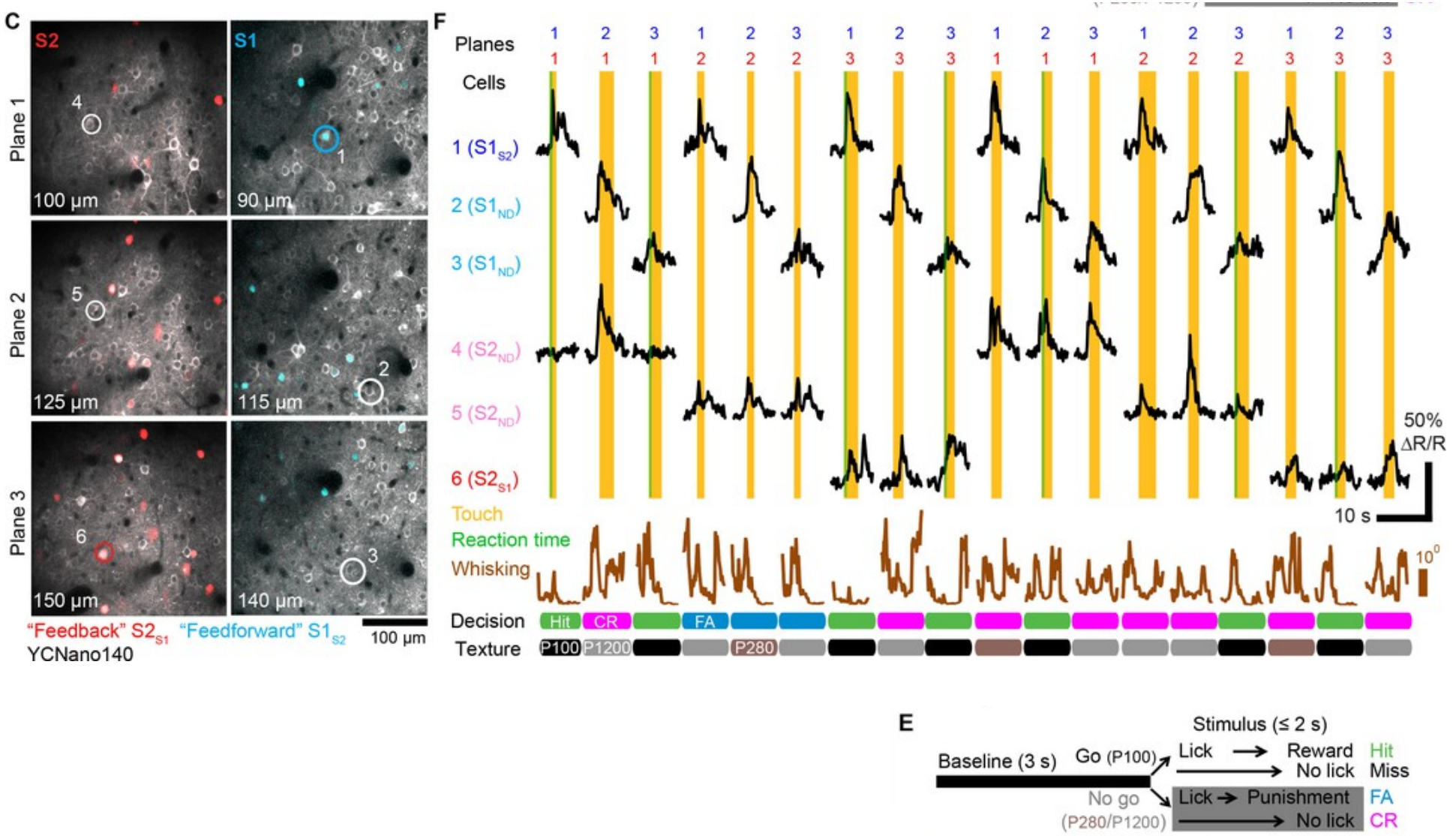
Calcium Imaging:
- correct for crosstalk between channels
- Background subtraction
- motion correction
- choose ROI manually (= neurons)
- extract mean pixel value of each ROI
- calculate relative YFP/CFP ratio change according to:

$$\Delta R/R = (R-R_0)/R_0$$

Whereby R_0 is the bottom 8th percentile of the ratio of a trial

- identify active neurons by two-way ANOVA of the neuronal calcium signal against the neuropil

Long-range population dynamics of anatomically defined neocortical networks: example data



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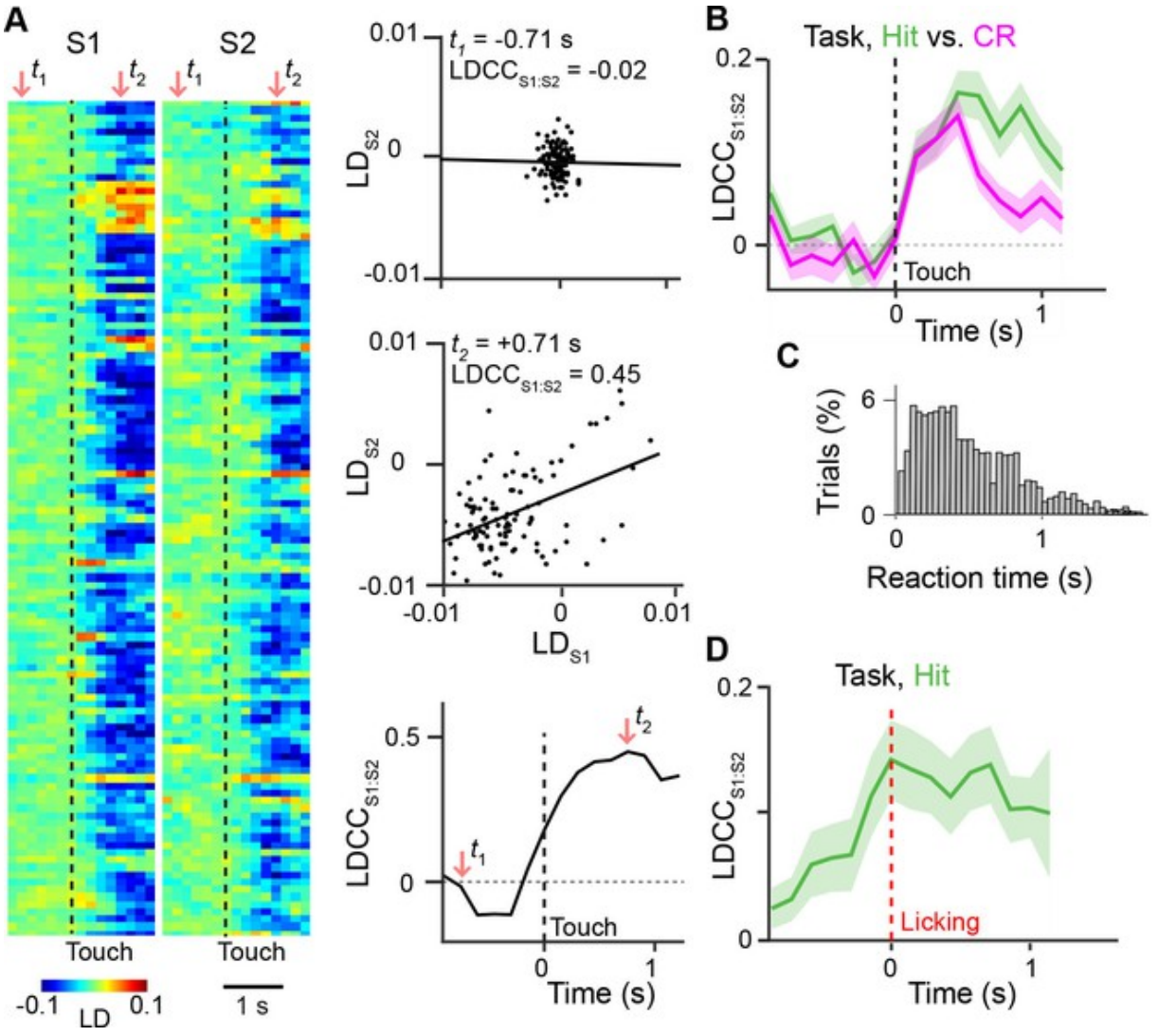
Neuronal population responses

Linear discriminant analysis
- can be used as a dimensionality reduction method

- seeks to find a vector representing maximal separation of two conditions for each timepoint (represented as LD)

- for whole region analysis, LD values from all imaging areas/planes were averaged and then cross-correlated between regions

Long-range population dynamics of anatomically defined neocortical networks



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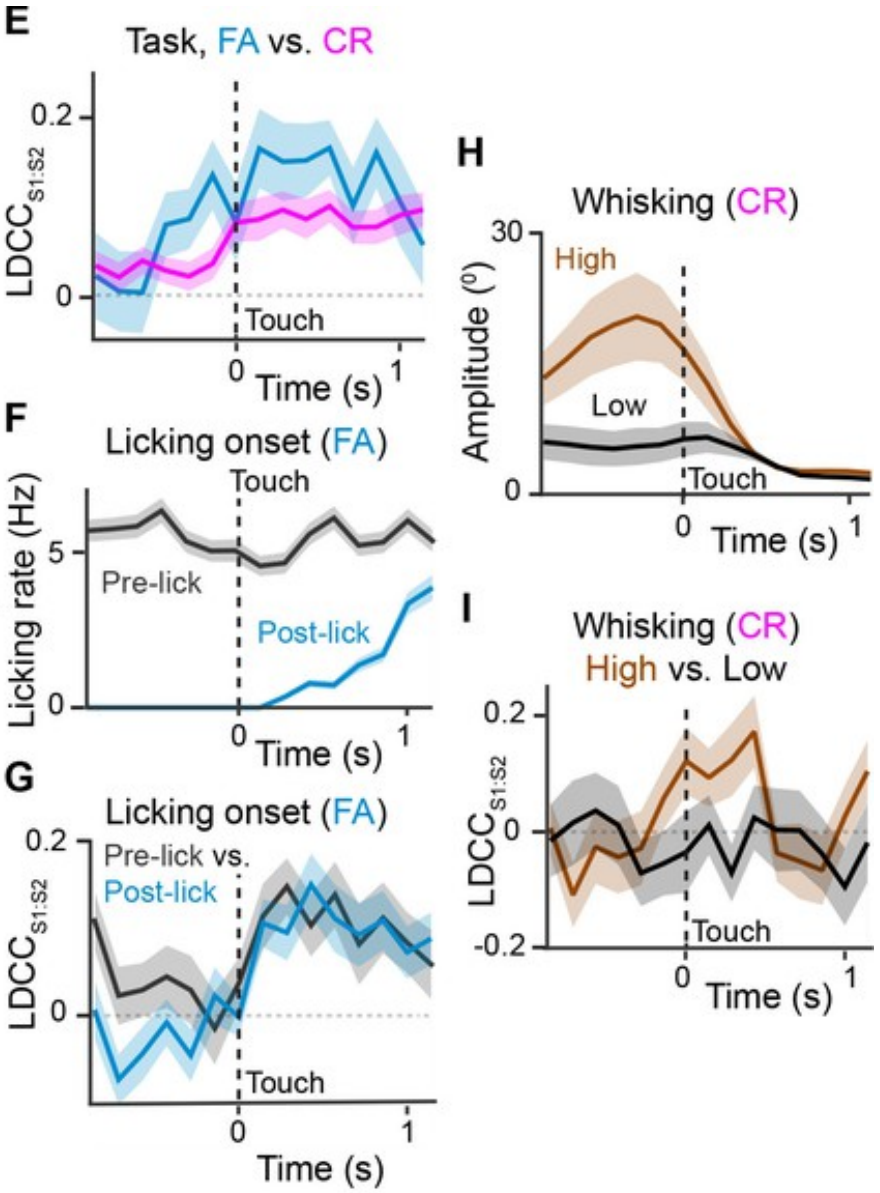
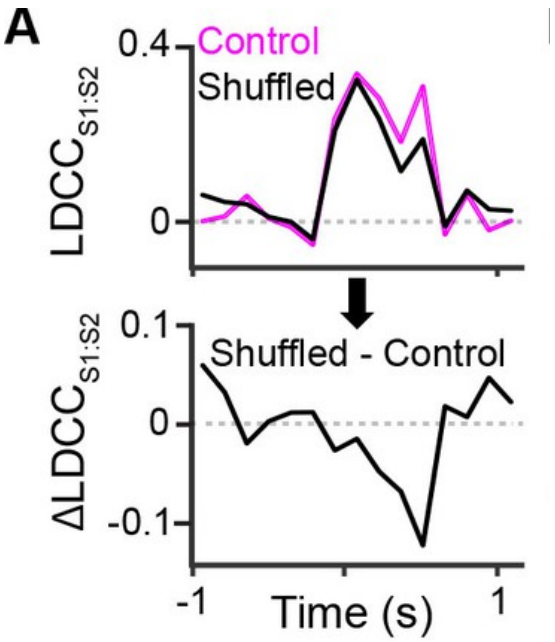


Coordination of population activity across S1 and S2 can be associated with licking and whisking behaviour that is independent of sensory stimulus

Long-range population dynamics of anatomically defined neocortical networks



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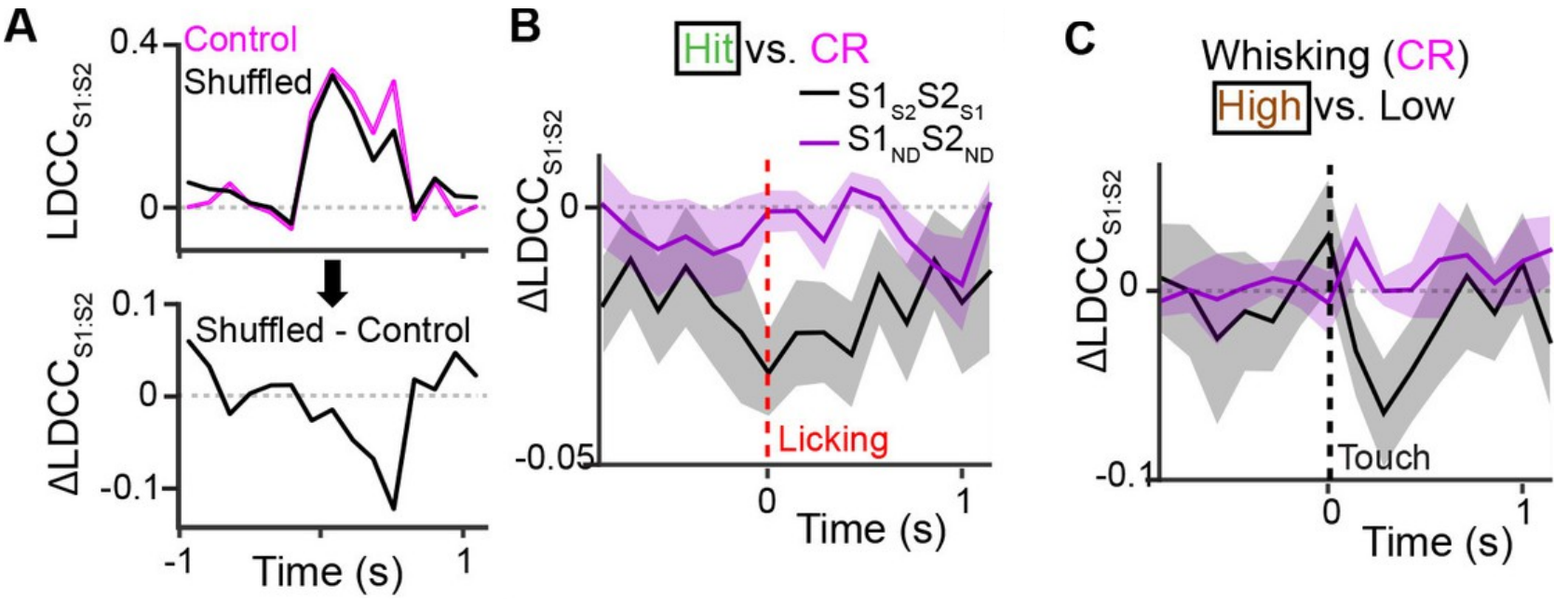

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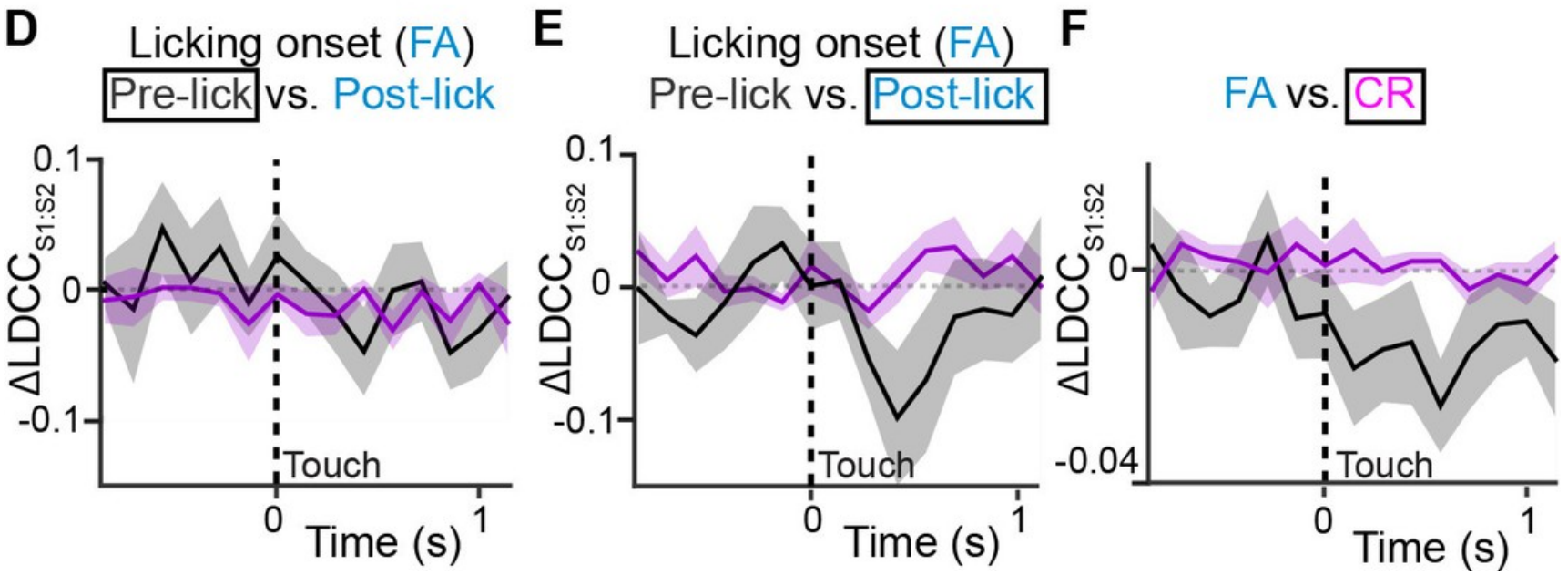
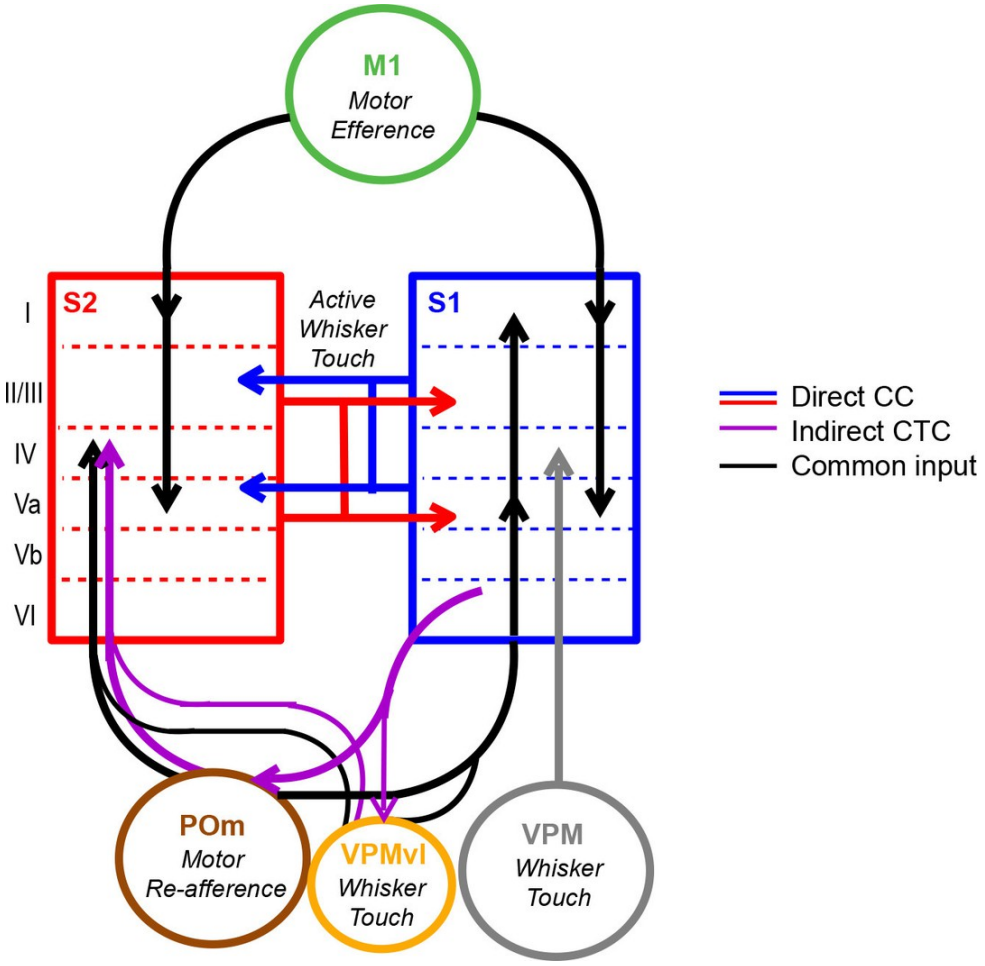


This data indicates that S1S2 interactions reflect exchange of sensory or decision information rather than motor information

Long-range population dynamics of anatomically defined neocortical networks: Discussion



Summary:
Simultanous calcium imaging in two different brain areas

Changes in correlated activity of projection neurons associated significantly with sensory input

Paper 2

Letter

Fully integrated silicon probes for high-density recording of neural activity

James J. Jun, Nicholas A. Steinmetz [...] Timothy D. Harris

Nature **551**, 232–236 (09 November 2017)
doi:10.1038/nature24636
Download Citation

Received: 27 February 2017
Accepted: 16 October 2017
Published online: 08 November 2017

Paper 2

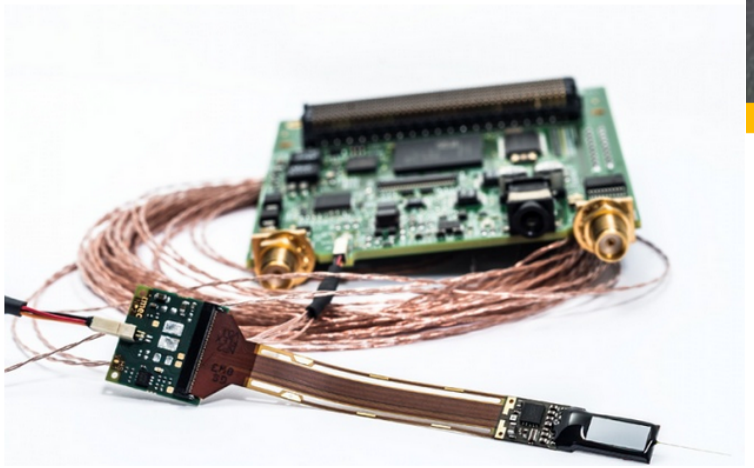
THE ENGINEER NEWS IN-DEPTH OPINION SECTORS CA

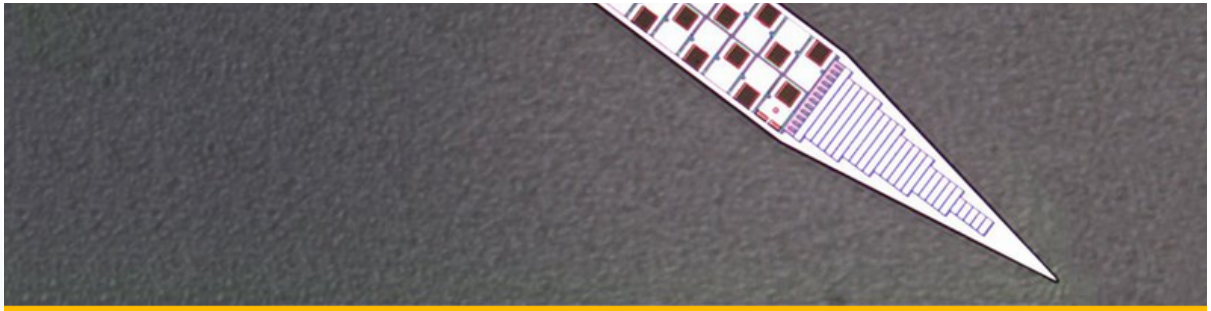
News Materials Medical & healthcare Medical Devices Neuroscience Sensors

Neuropixels probes promise new era of brain research

13th November 2017 11:06 am

Humanity may be on the cusp of an exciting new phase of neuroscience thanks to the development of highly sensitive silicon devices called Neuropixels probes.





Thinner than a human hair. (Timothy Harris Lab, HHMI's Janelia Research Ca

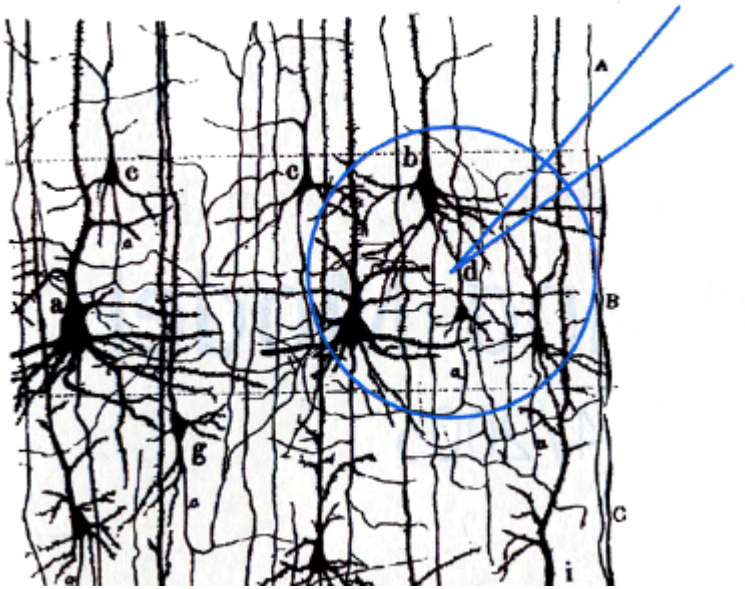
This Incredibly Tiny Probe Can Record Brain Activity Like We've Never Seen Before

It will change what we know about the brain.

DAVID NIELD 10 NOV 2017

Background: Extracellular Recordings

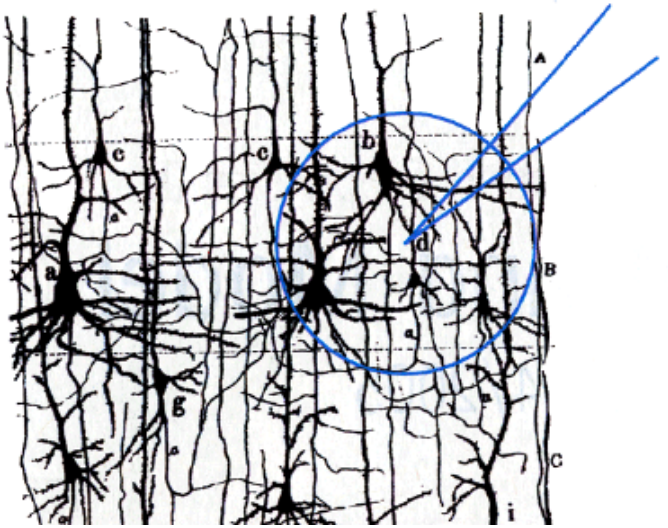
- Voltage changes at electrode site
- Both: Local field potential and Spikes
- Spikes: fast frequency component. Reflects the AP of one or more neurons
- LFP: slow frequency component. Reflects simultaneous activity of dendrites of similar orientation and geometry

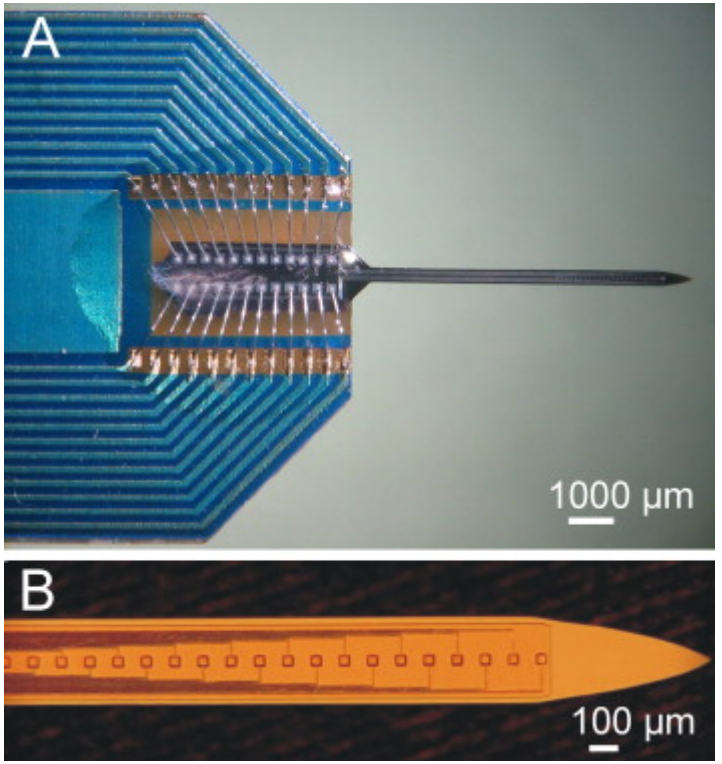


Background: Silicon Probes

Extracellular electrode

Based on silicone as carrier material
Multiple recording sites





Picture from: https://doi.org/10.1016/j.sna.2010.12.019

Neuropixels: goals

To develop a silicon probe with

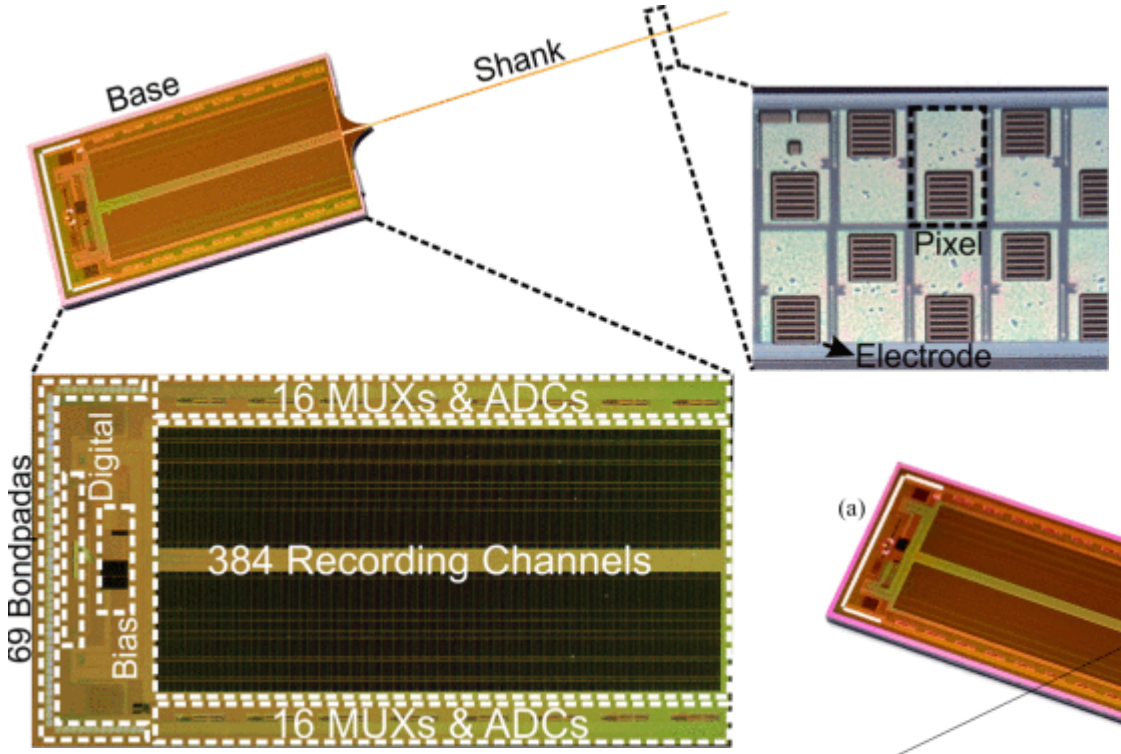
1) dense and extensive recording sites
2) small cross-sectional area
3) low noise
4) resistance to movement artefacts
5) efficient data transmission
6) long-term recording stability
7) low-cost scalable fabrication

1) dense and extensive recording sites

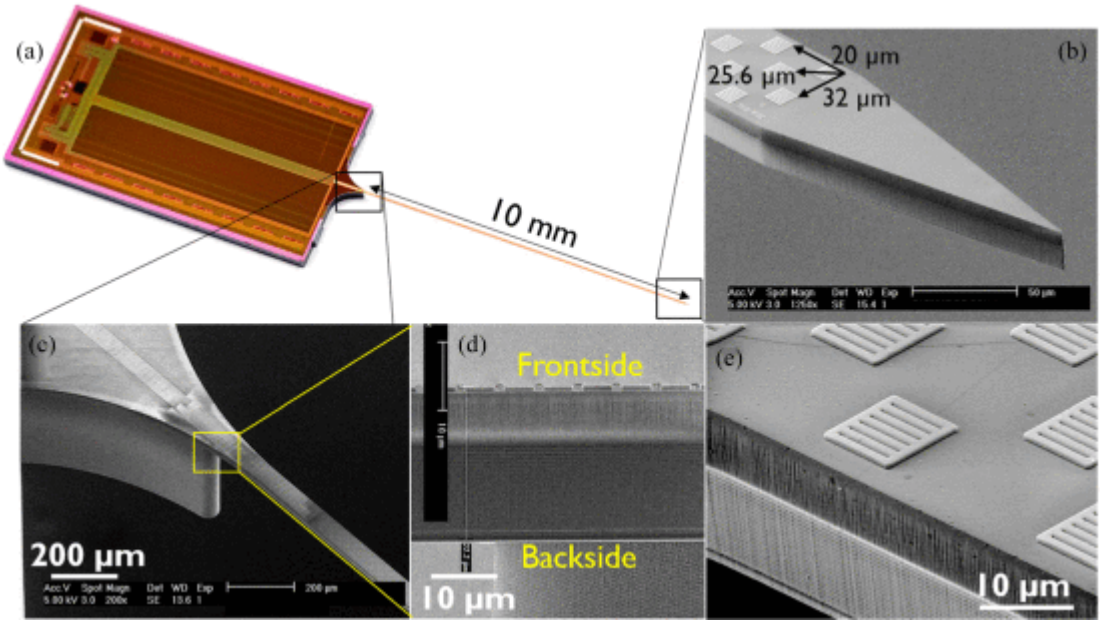
A Neural Probe With Up to 966 Electrodes and Up to 384 Configurable Channels in 0.13 μm SOI CMOS

Carolina Mora Lopez, *Member, IEEE*, Jan Putzeys, Bogdan Cristian Raducanu, *Graduate Student Member, IEEE*, Marco Ballini, *Member, IEEE*, Shiwei Wang, Alexandru Andrei, Veronique Rochus, Roeland Vandebriel, Simone Severi, Chris Van Hoof, Silke Musa, Nick Van Helleputte, Refet Firat Yazicioglu, *Member, IEEE*, and Srinjoy Mitra, *Member, IEEE*

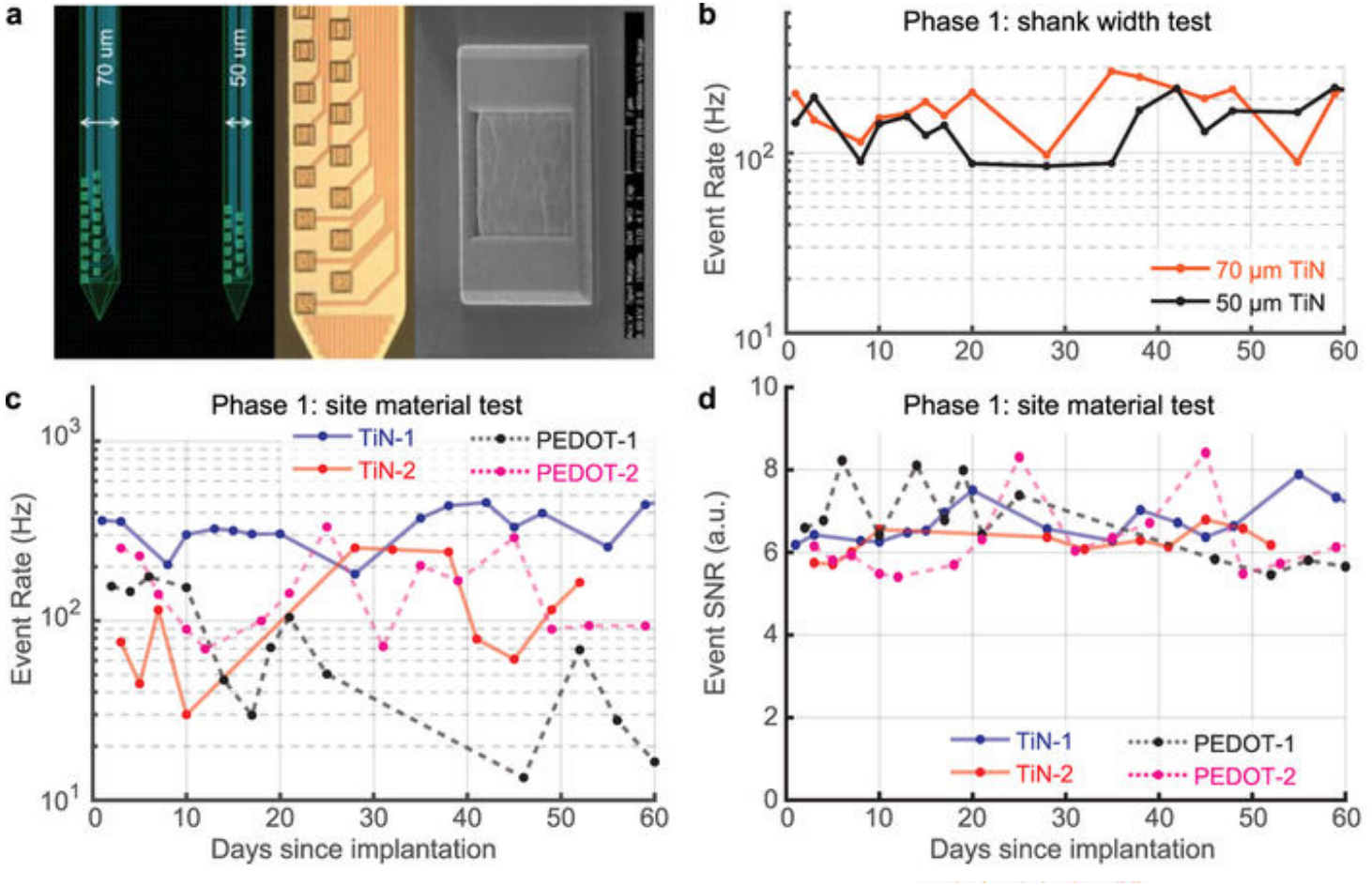
1) dense and extensive recording sites



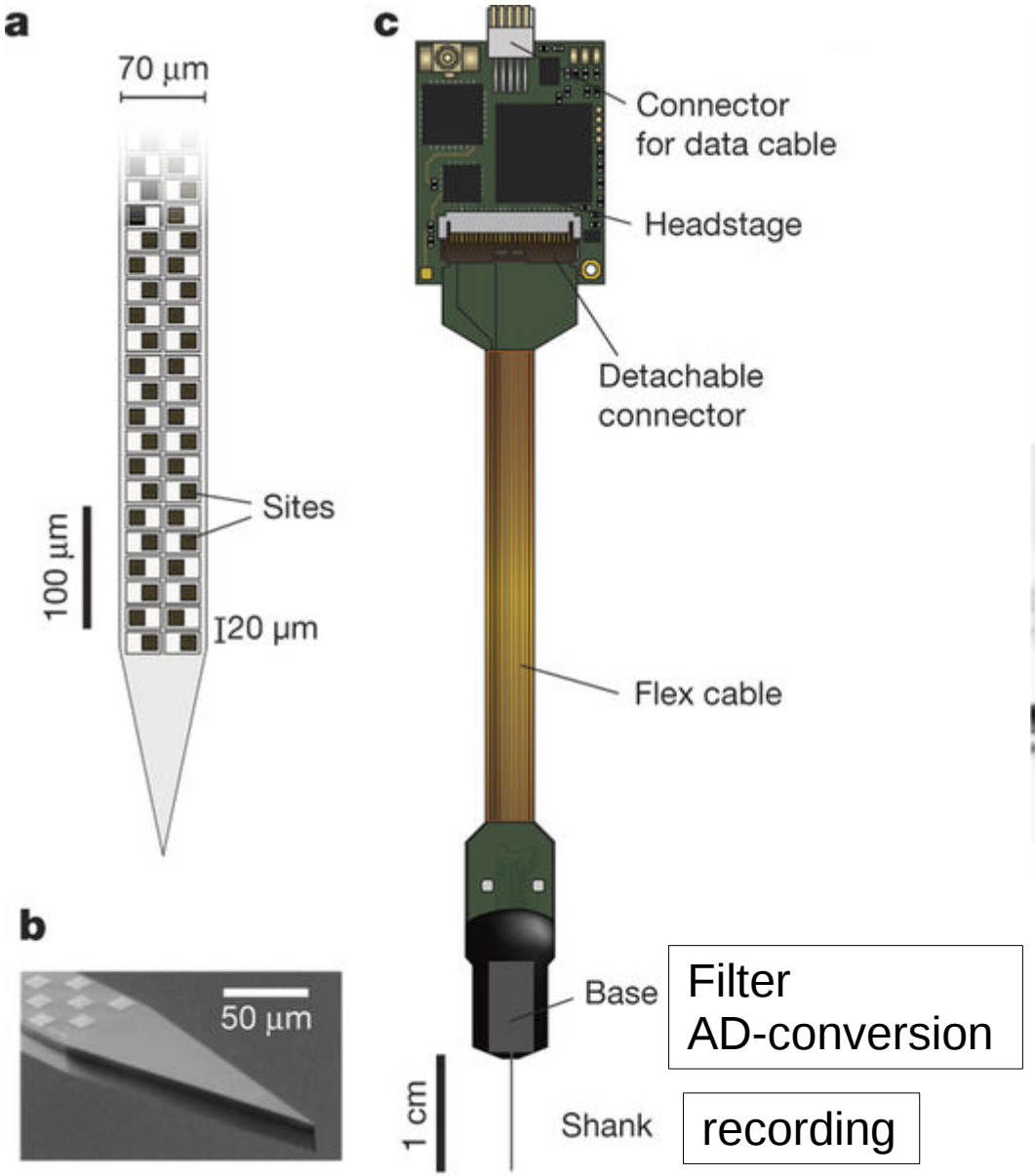
- Titanium nitride electrode
- CMOS-based
- 960 electrodes of which
- 384 can be recorded simultanously



2) small cross-sectional area

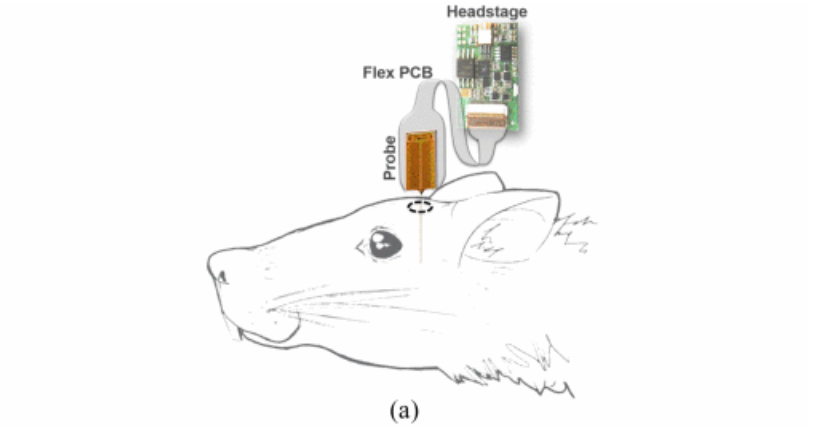


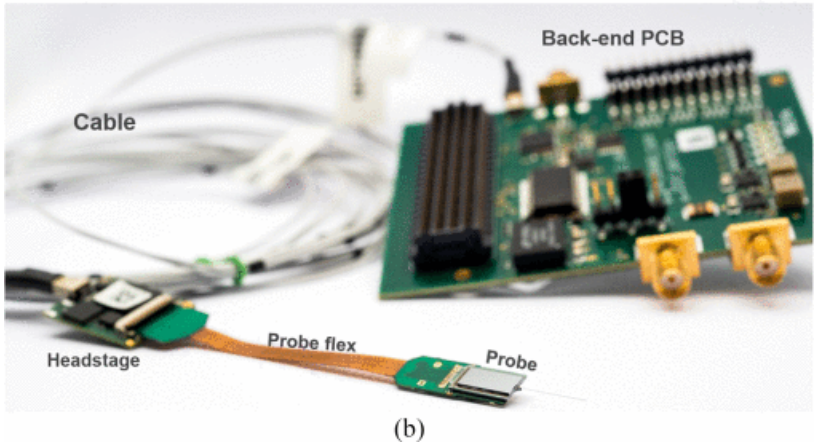
Design



Filter
AD-conversion

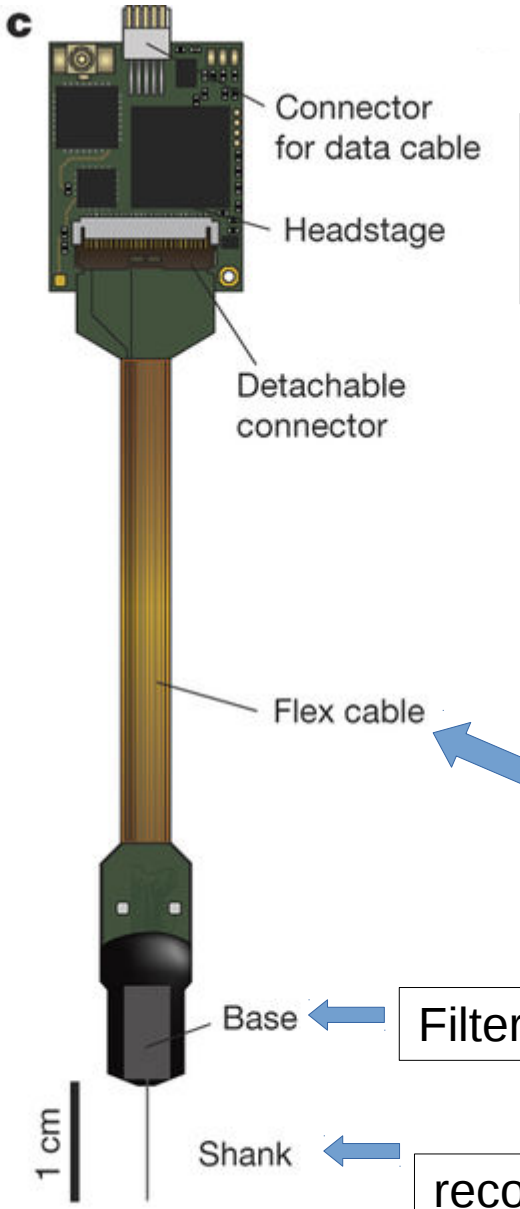
recording





Weight
- shank + base: 250mg

5) efficient data transmission



Power supply
Data transfer
Control of switches

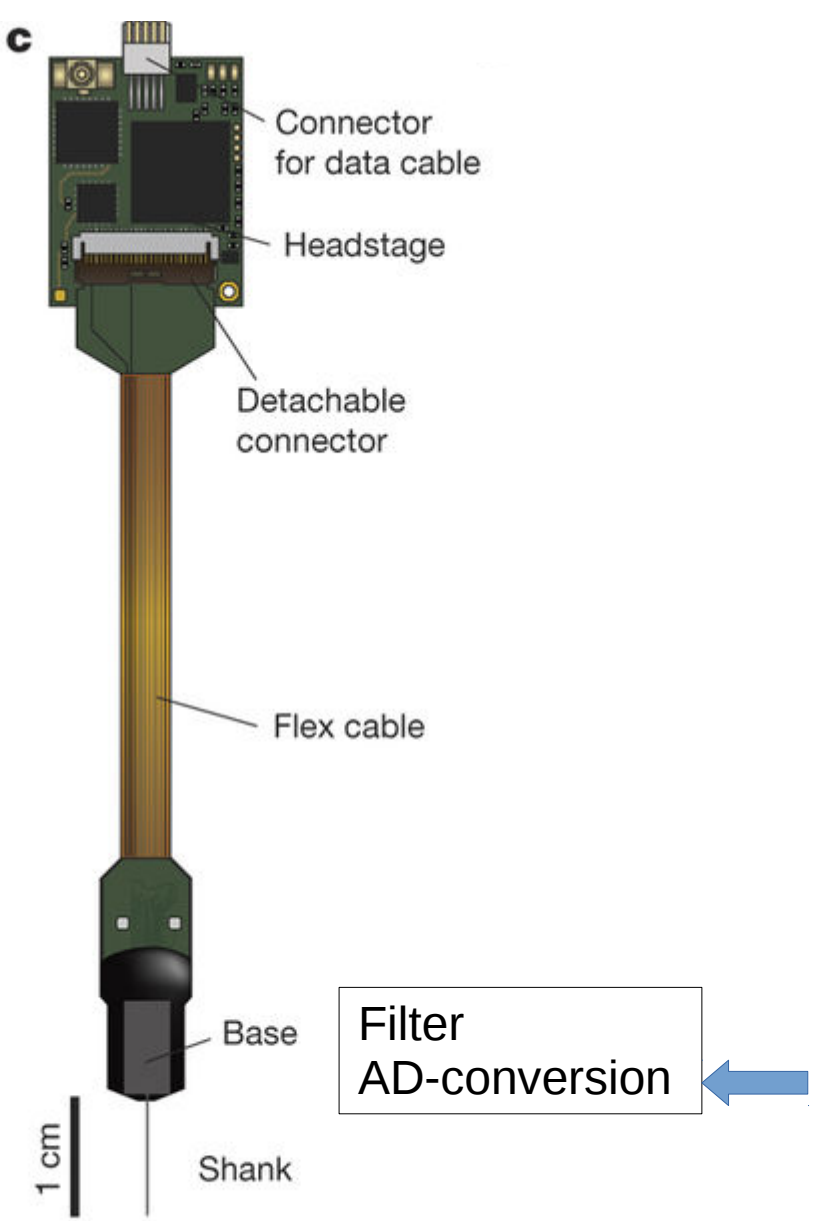
AD-conversion (10bit)
AP: 30kHz
LFP: 2.5kHz

Filter

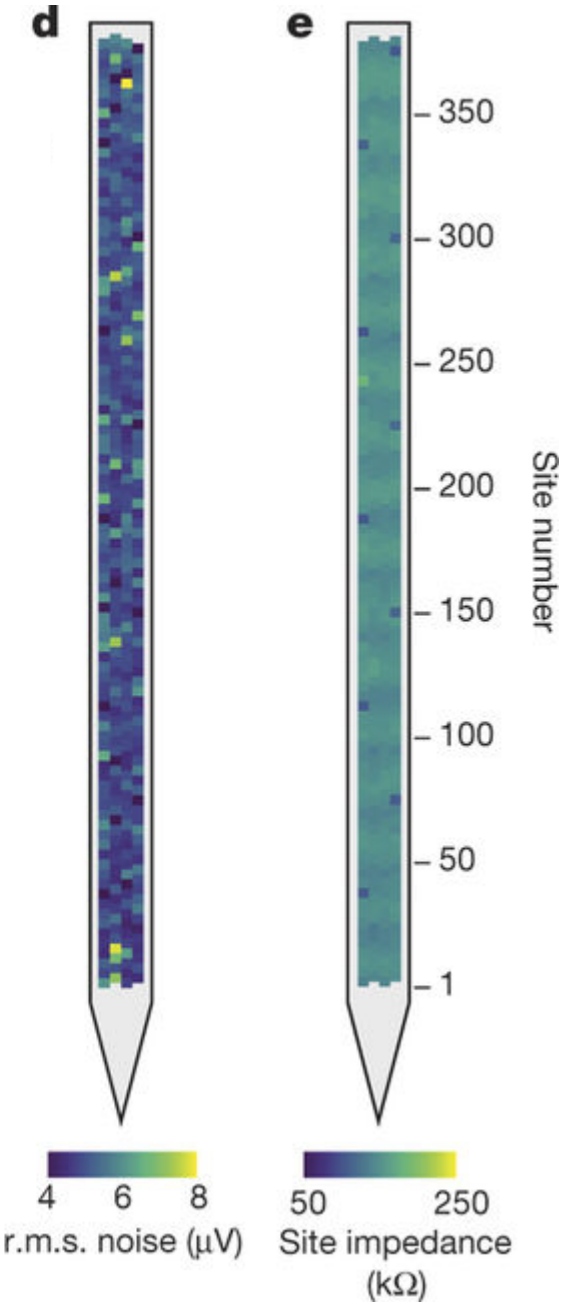
2 channels:
AP: high frequency (0.3 - 10kHz)
LFP: low frequency (0.5-1000Hz)

recording

3) low noise



Filter
AD-conversion



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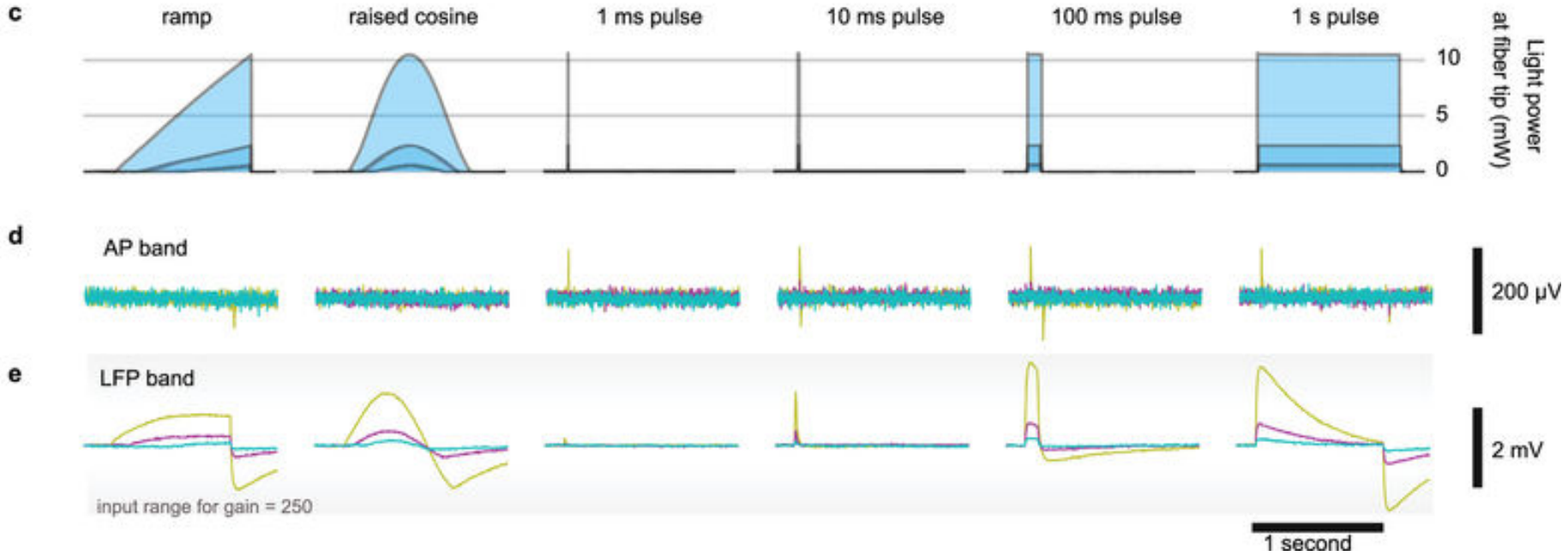
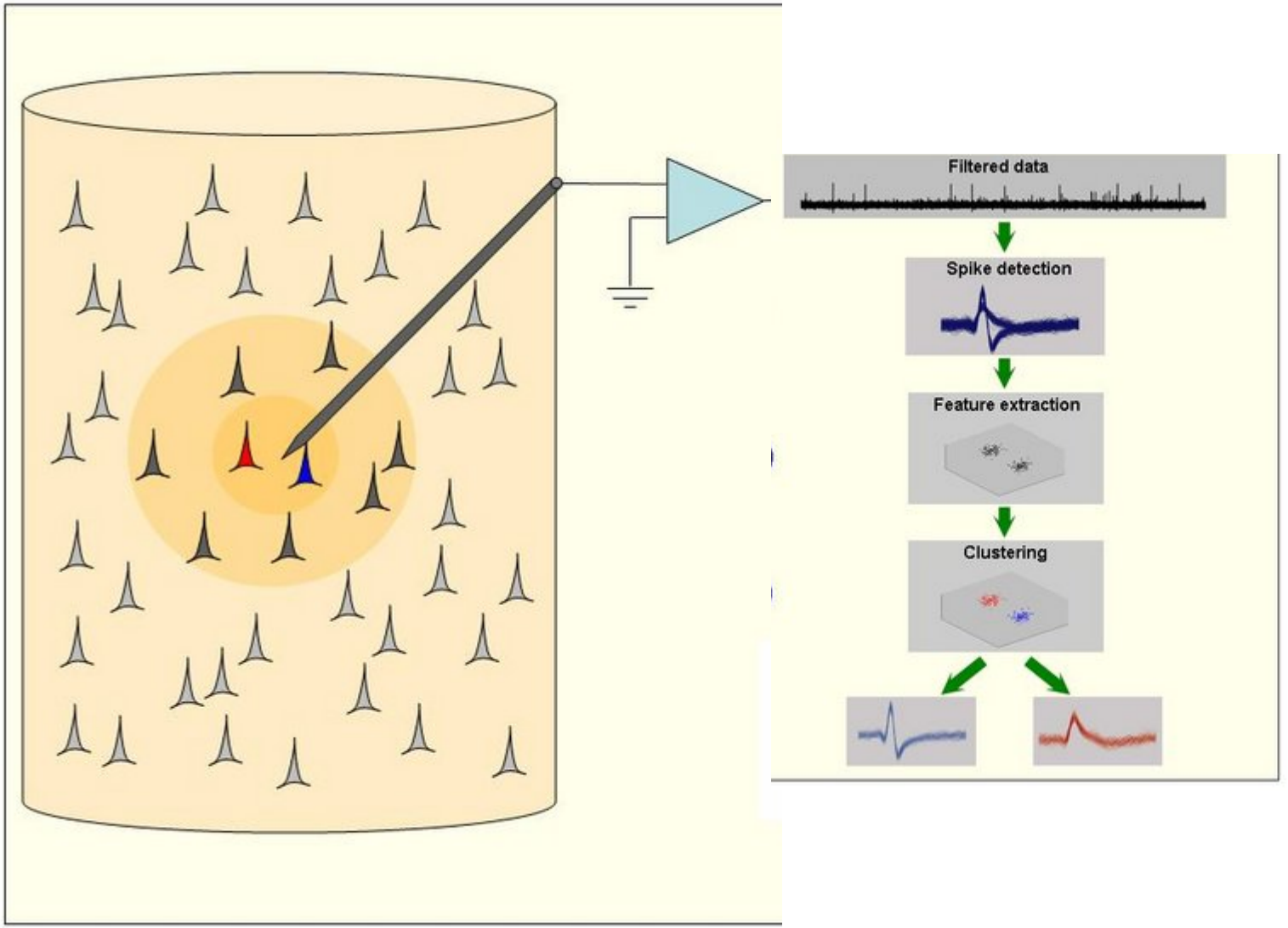


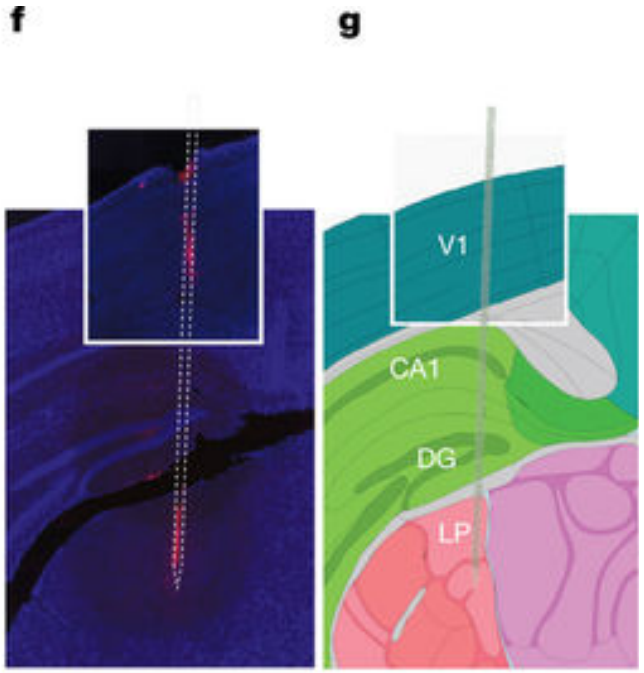
light levels:
Low: cyan
Middle: magenta
High: yellow

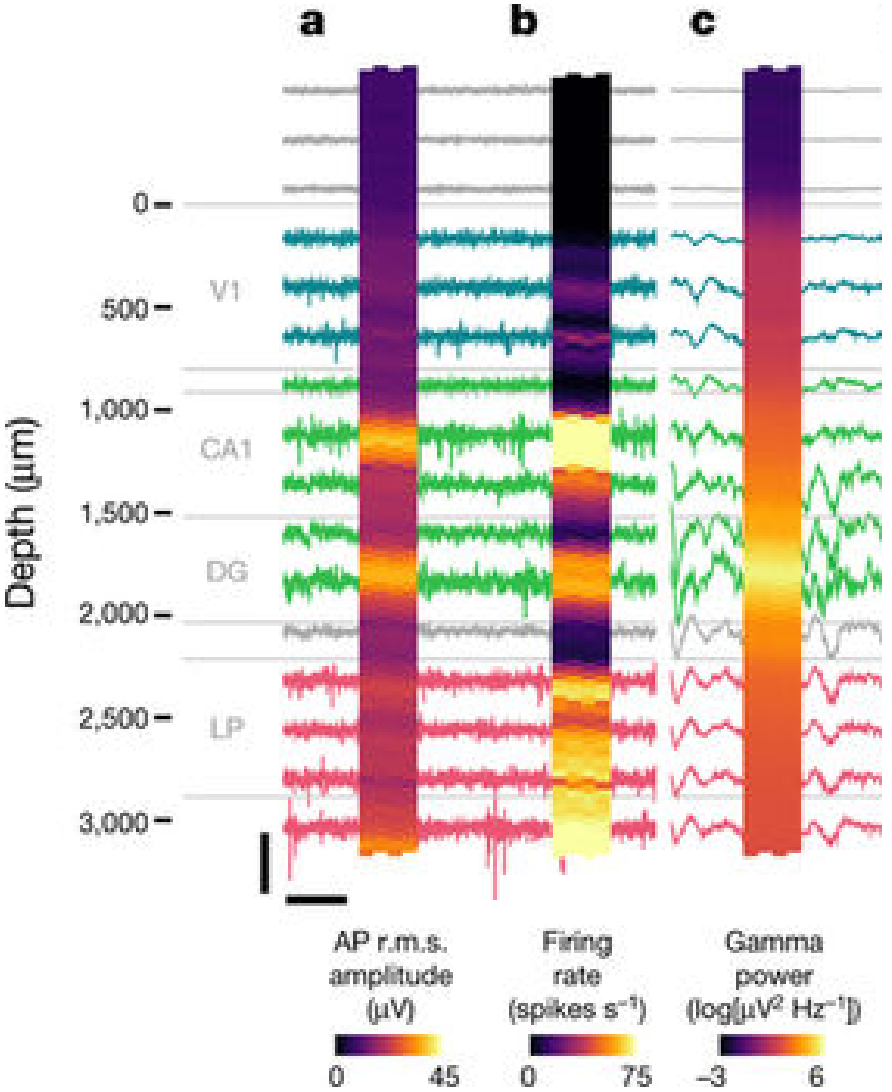
Background: Analysis of extracellular Recordings



Adapted from: http://www.scholarpedia.org/article/File:QQ_Fig1.jpg and
from lecture on extracellular recordings in BIO434 course 2013 by Asli Ayaz

Recording from large neuronal populations with a single probe in an awake head-fixed mouse

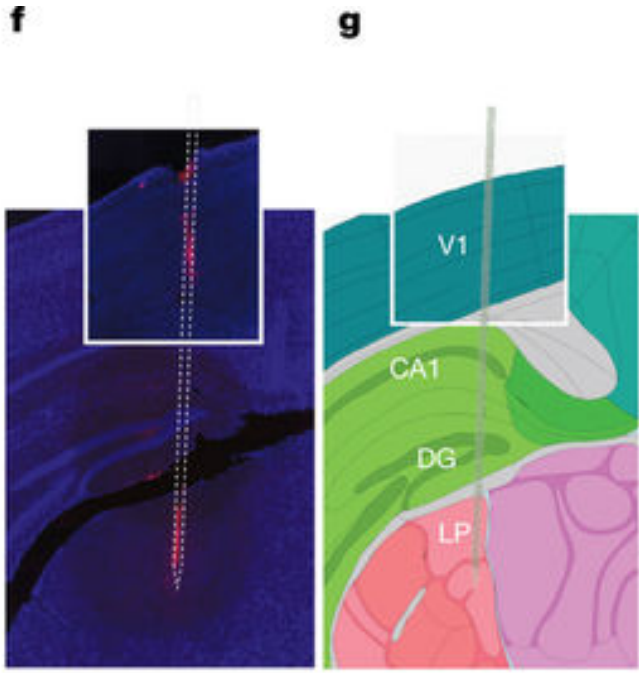




n = 103 neurons in Thalamus,
41 neurons in Hippocampus,
62 neurons in cortex

Figure 1

Recording from large neuronal populations with a single probe in an awake head-fixed mouse



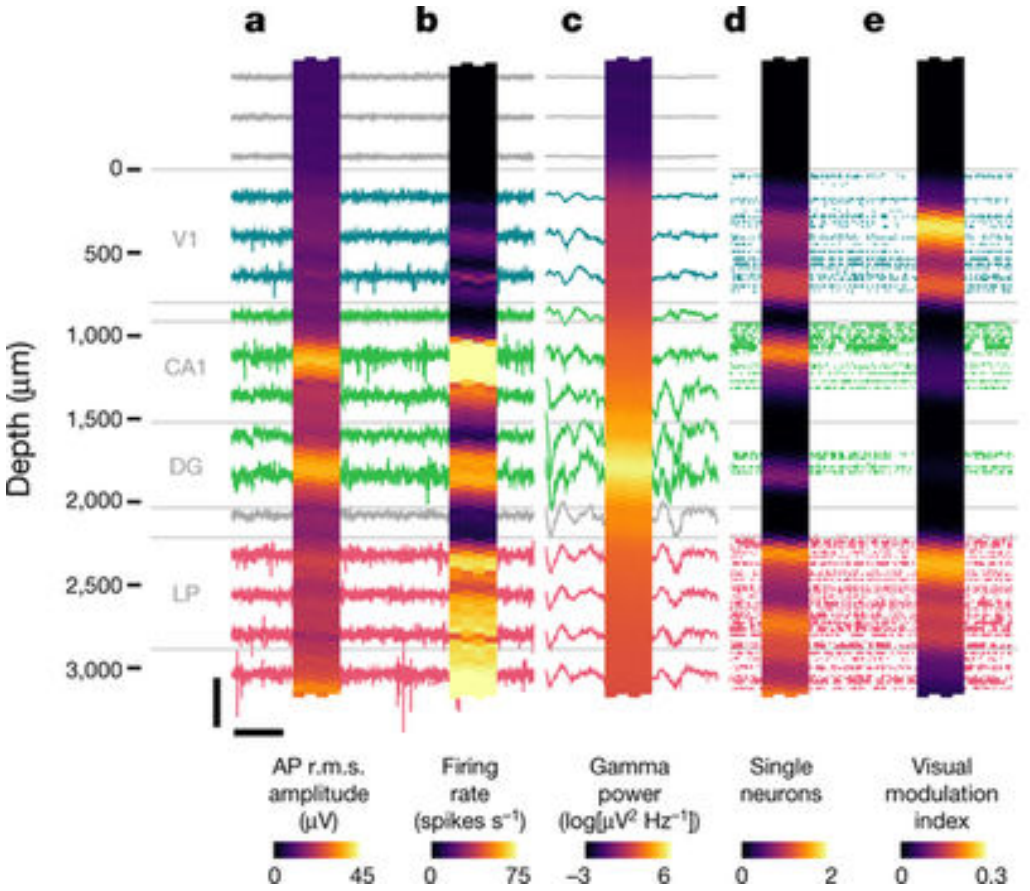


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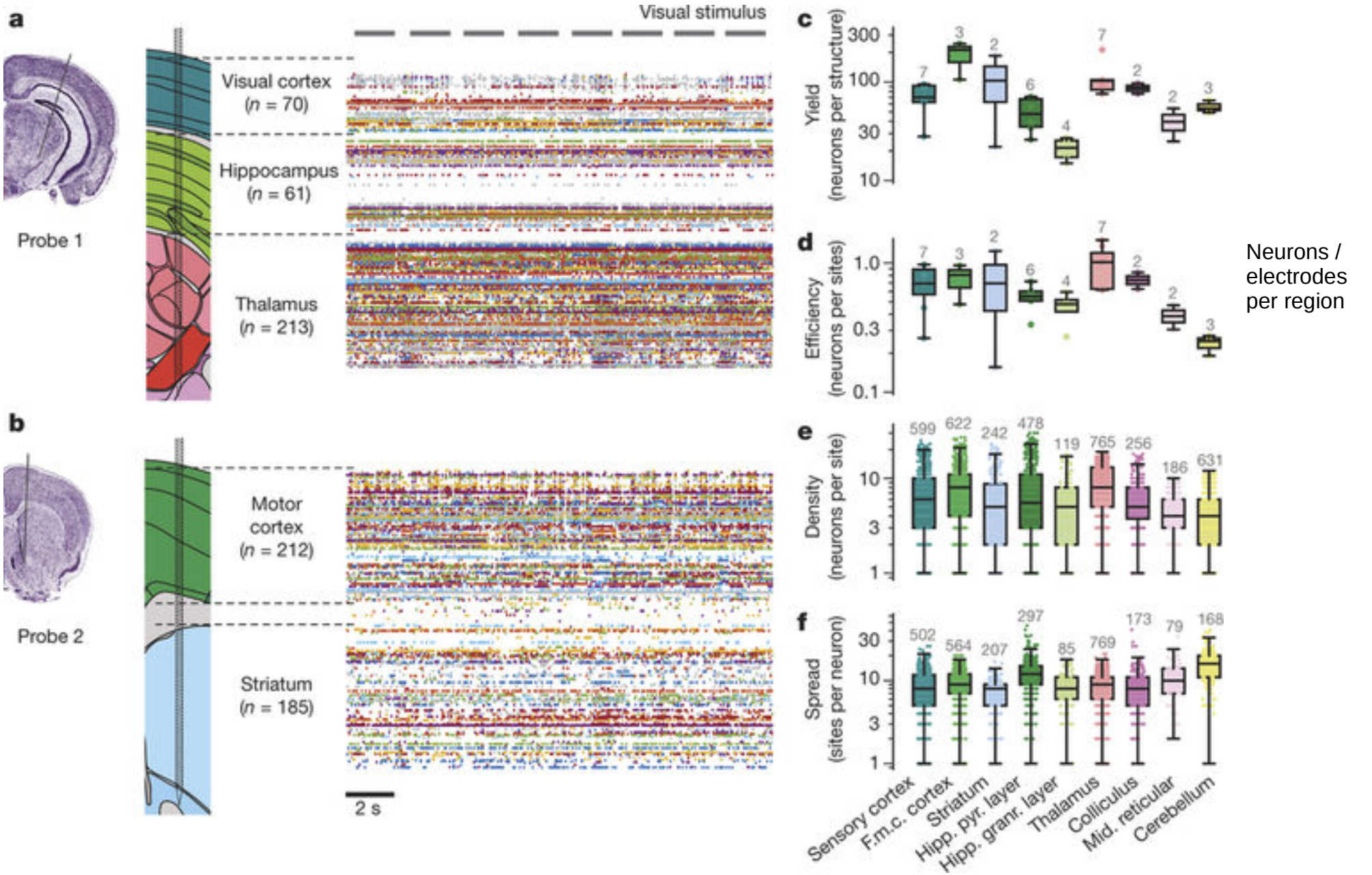
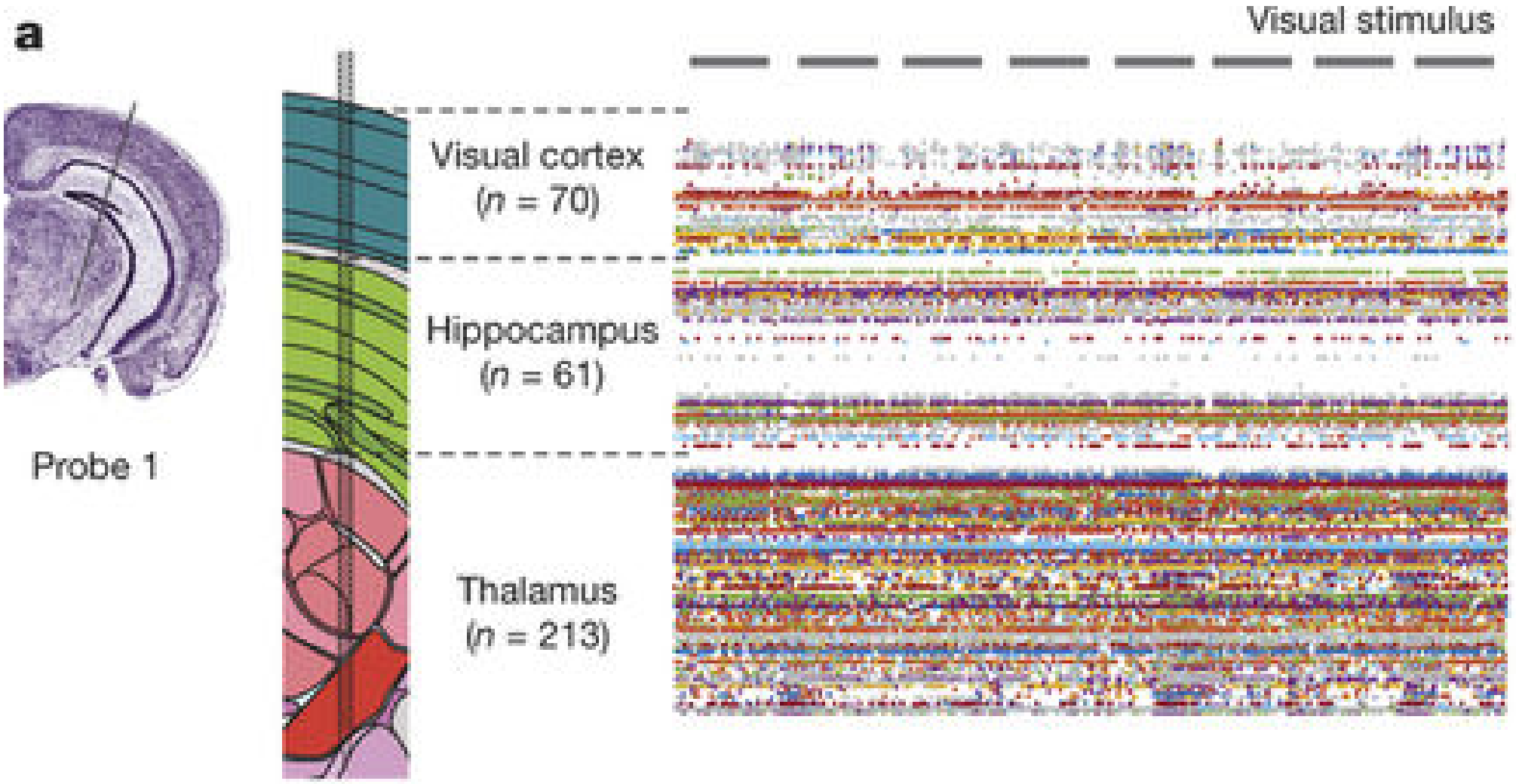
Figure 1

Recording from multiple brain structures in awake head-fixed mice.

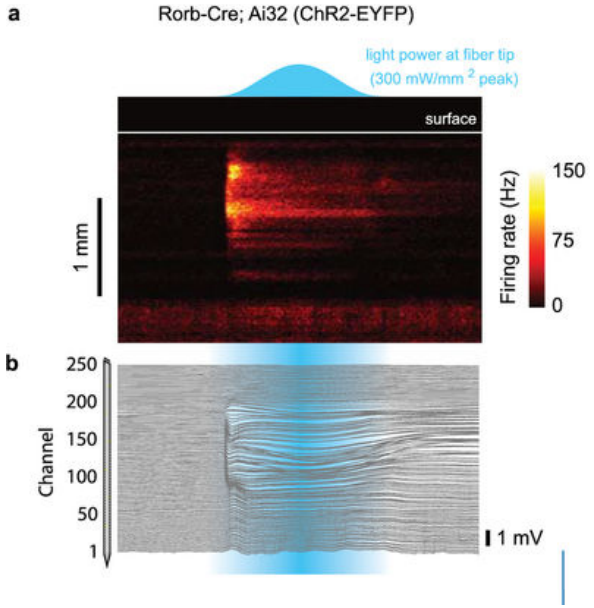


Neurons / electrodes per region

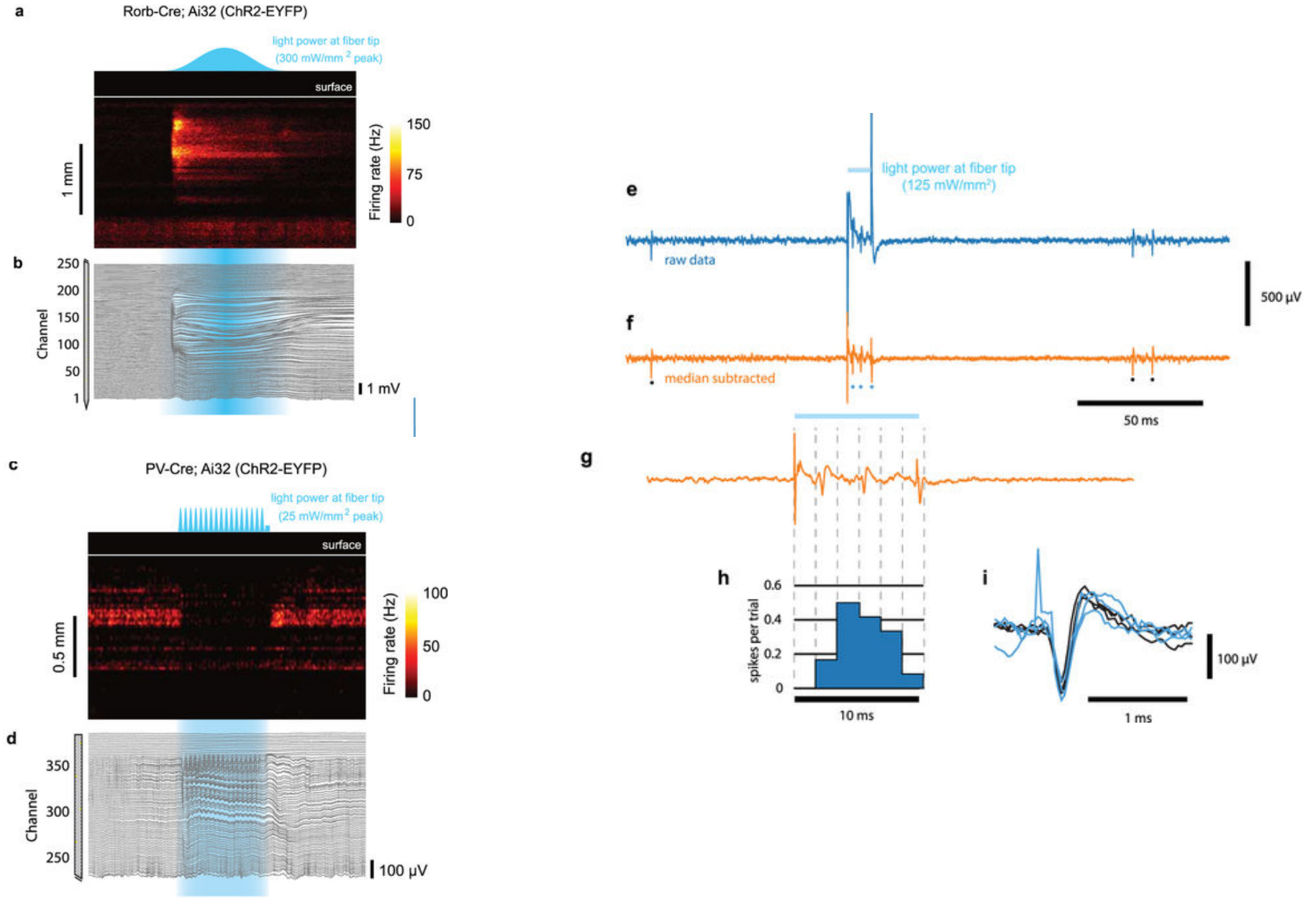
Recording from multiple brain structures in awake head-fixed mice.



Recording during optogenetic stimulation of excitatory and inhibitory cell populations

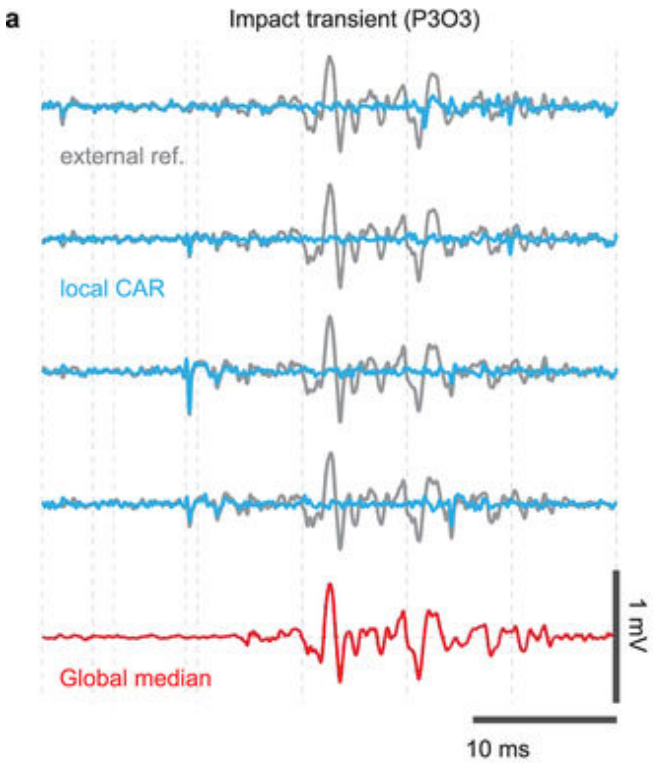


Recording during optogenetic stimulation of excitatory and inhibitory cell populations

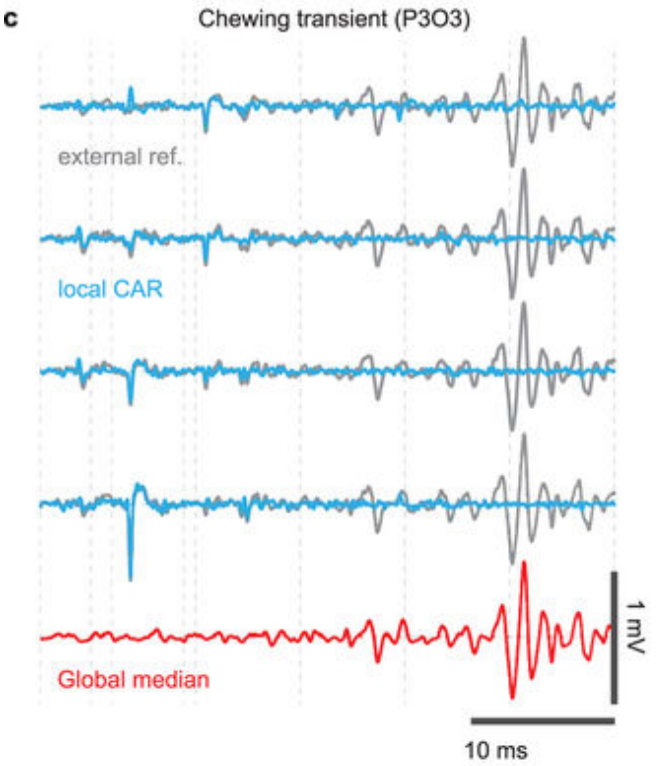


4) resistance to movement artefacts

Hit the implant with cable

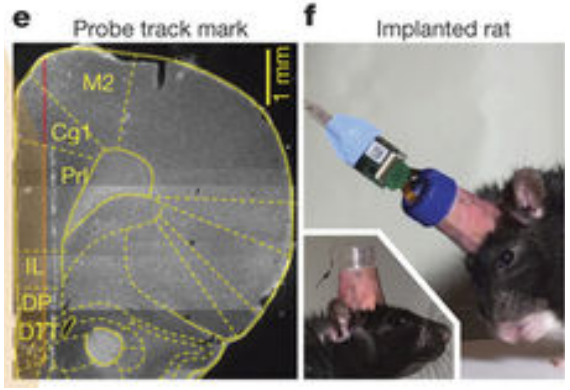


Animal is eating



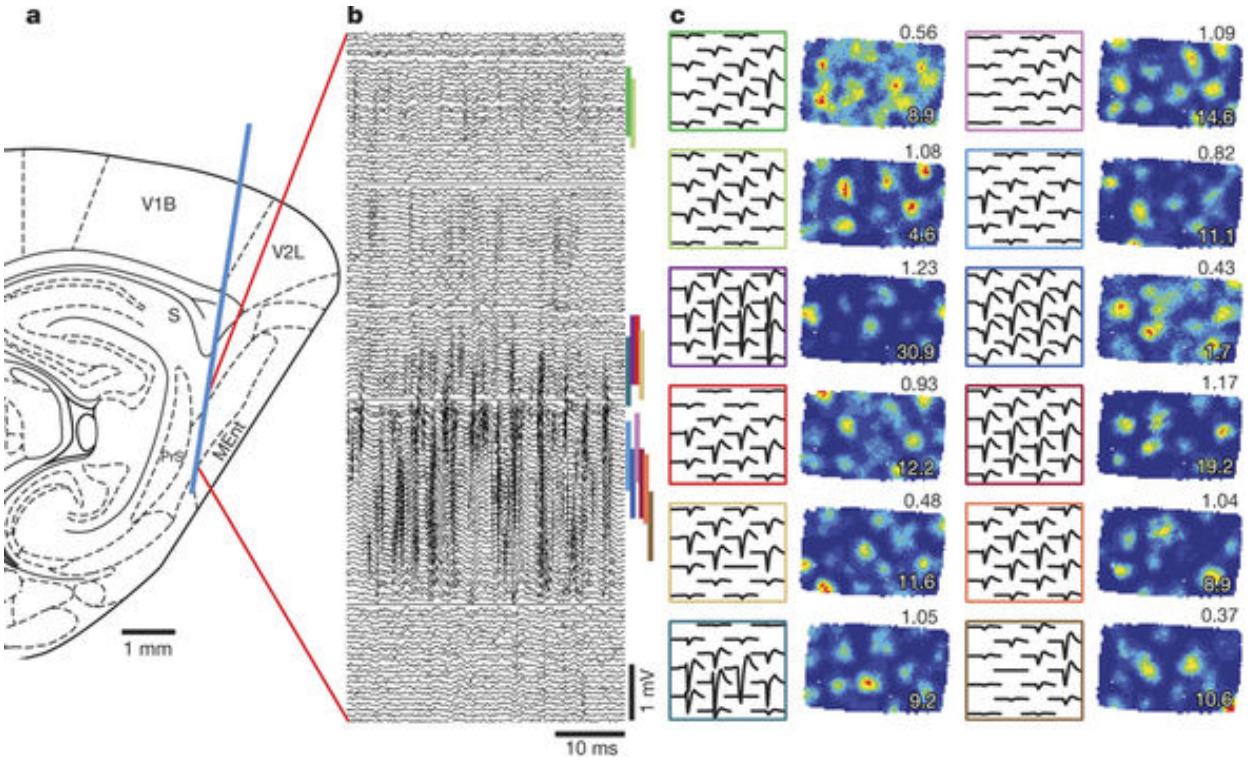
CAR: local common average referencing – substraction of local background

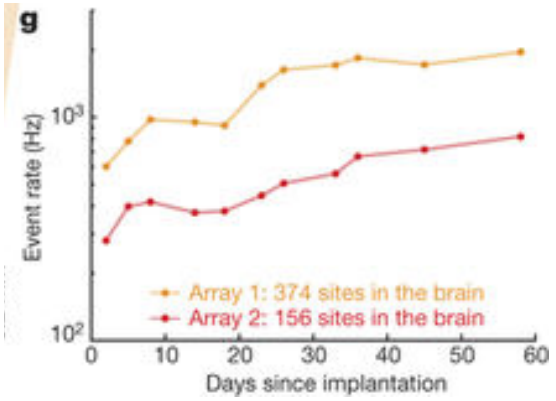
Recordings from entorhinal and medial prefrontal cortices using chronic implants in unrestrained rats



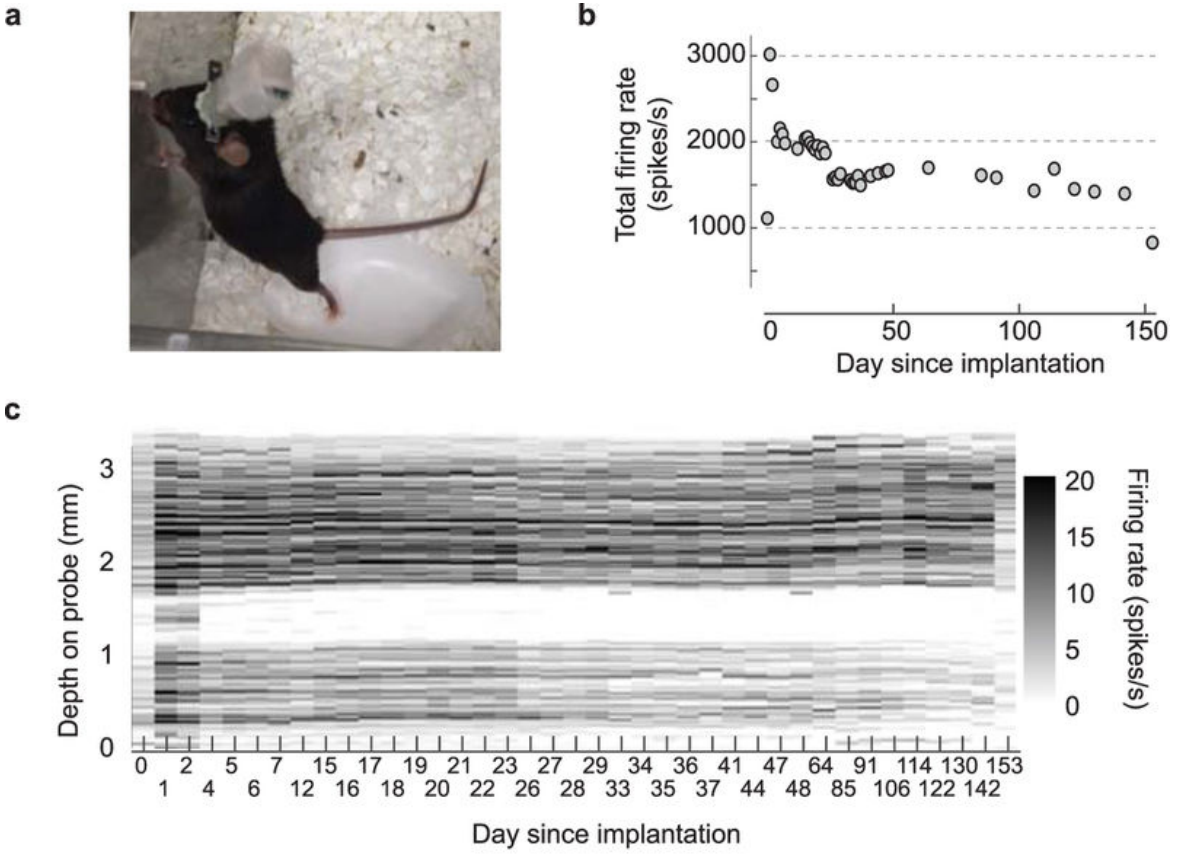
n = 127 cells

grid cell recording

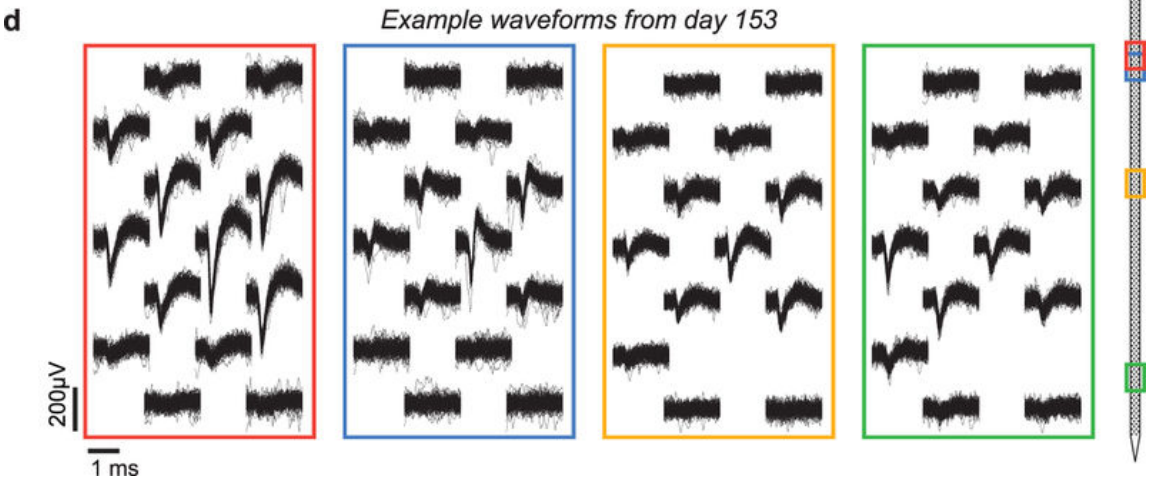




6) long-term recording stability



Weight of the whole headpost: 3g



Neuropixels: Summary

To develop a silicon probe with

1) dense and extensive recording sites
2) small cross-sectional area
3) low noise
4) resistance to movement artefacts
5) efficient data transmission
6) long-term recording stability
7) low-cost scalable fabrication

No demonstration of new insights (yet)

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No demonstration of new insights (yet)

Commercially available sometimes in 2018. We‘ll see….

Side by side comparison

	Calcium Imaging	**Silicon Probe**
Anatomical information	good	Limited (only post-hoc)
Cell type specificity	possible	limited
Temporal resolution	Slow (Hz)	Fast (kHz)
Access to deeper brain regions	limited	easy
coverage	limited	large
Head-fixation	Usually needed	Not necessarily needed
price	Very expensive	expensive

Summary and Outlook

Both methods provide functional data on neuronal activity

Method should be chosen based on experimental requirements regarding anatomical / temporal resolution, cell type specificity etc

If possible, interventions to disrupt the proposed circuitry function should be applied

Conclusion and Outlook

Both methods provide functional data on neuronal activity

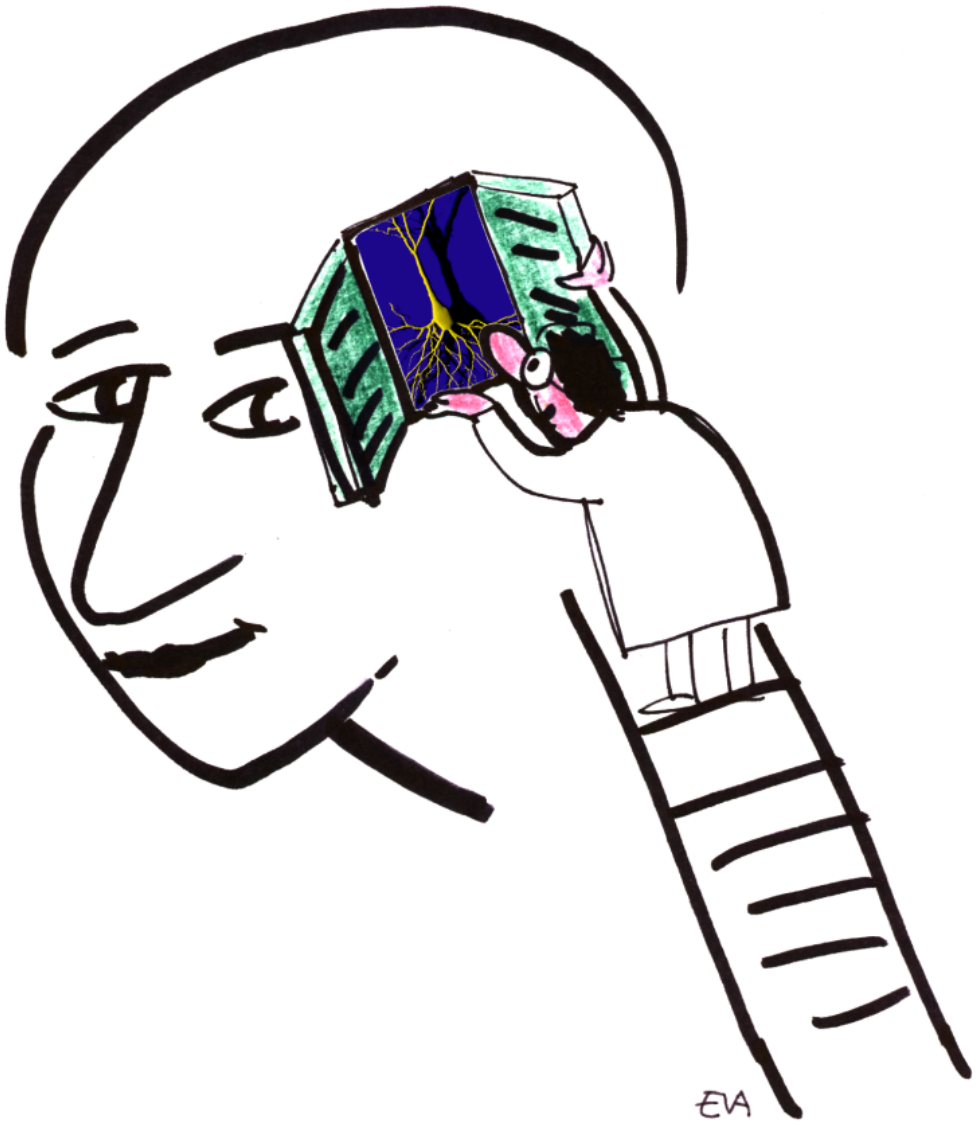
Method should be chosen based on experimental requirements regarding anatomical / temporal resolution, cell type specificity etc

If possible, interventions to disrupt the proposed circuitry function should be applied

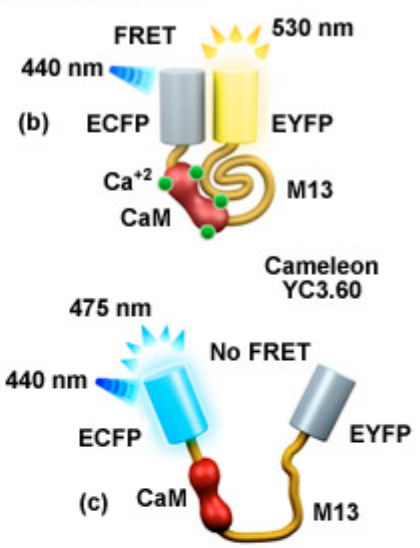
In the future, the spatial resolution of the silicon probes might be further improved, as well as the temporal resolution of calcium imaging

More sophisticated data analysis methods will be available

Thank you

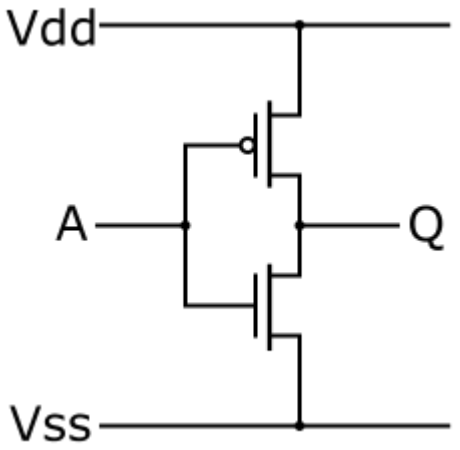


Appendix: Ycnano – yellow cameleon ratiometric calcium indicator



Appendix: CMOS

Complementary metal–oxide–semiconductor,



Appendix: all probe options

	Option 1	Option 2	Option 3	Option 4
Site Count	384	384	960	966
Channel count	384	384	384	276
Electrode type	Passive	Active	Passive	Active
Shank power (mW)	0	1.31	0	1.31
Base power (mW)	17.5			
Electrode area (μm^2)	144			
Crosstalk (at 1kHz)	< 5%			
Gain	selectable from 50 - 2500			
AP band high-pass corner (kHz)	selectable from 0.3 - 1.0			
AP band low-pass corner (kHz)	10			
LFP band high-pass corner (Hz)	0.5			
LFP band low-pass corner (Hz)	1000			
AP band sampling rate (kHz)	30			
LFP band sampling rate (kHz)	2.5			
AP band noise (μV r.m.s.)	5.7 ± 0.8	6.6 ± 0.8	5.5 ± 0.7	6.6 ± 2.5
LFP band noise (μV r.m.s.)	9.6 ± 5.8	13.0 ± 2.8	8.0 ± 2.5	10.2 ± 1.9