Large Scale Imaging of the Retina

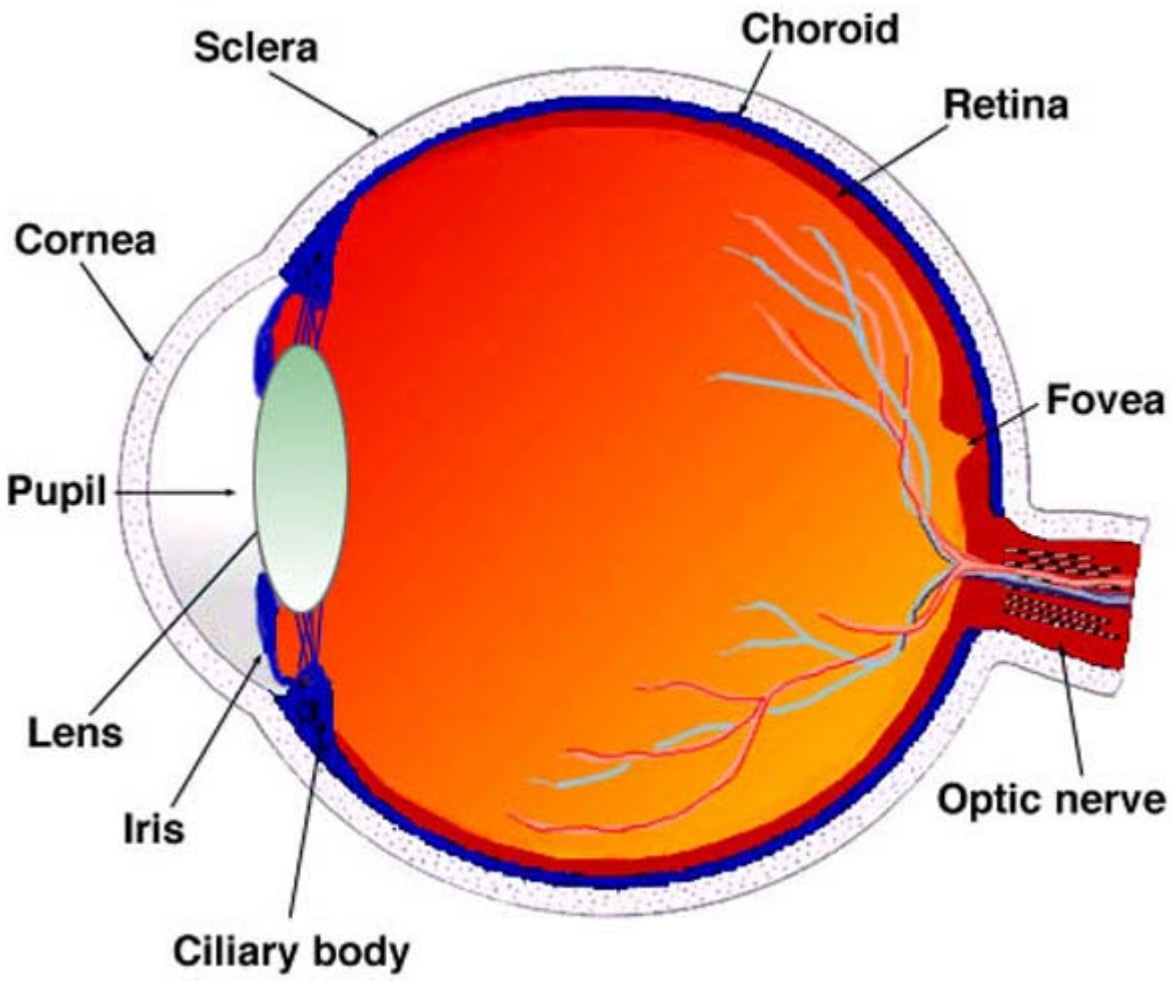
Alan Litke
SCIPP, UC Santa Cruz
SNIC, 6 April 2006

1. The Retina – a Biological Pixel Detector
2. Probing the Retina
   - ⇒understand the language used by the eye to send information about the visual world to the brain
   - ⇒use techniques and expertise from silicon microstrip detector development
3. Some First Results
4. Summary and Additional Applications

Collaborators

- UC Santa Cruz:
  A. Grillo, M. Grivich, S. Kachiguine, D. Petrusca, A. Sher
- AGH U. of Science and Technology, Krakow (I C design):
  W. Dabrowski, P. Hottowy
- U. Glasgow (high density electrode array fabrication):
  D. Gunning, K. Mathieson
- The Salk Institute (neurobiology):
  E. J. Chichilnisky, G. Field, J. Gauthier, J. Shlens

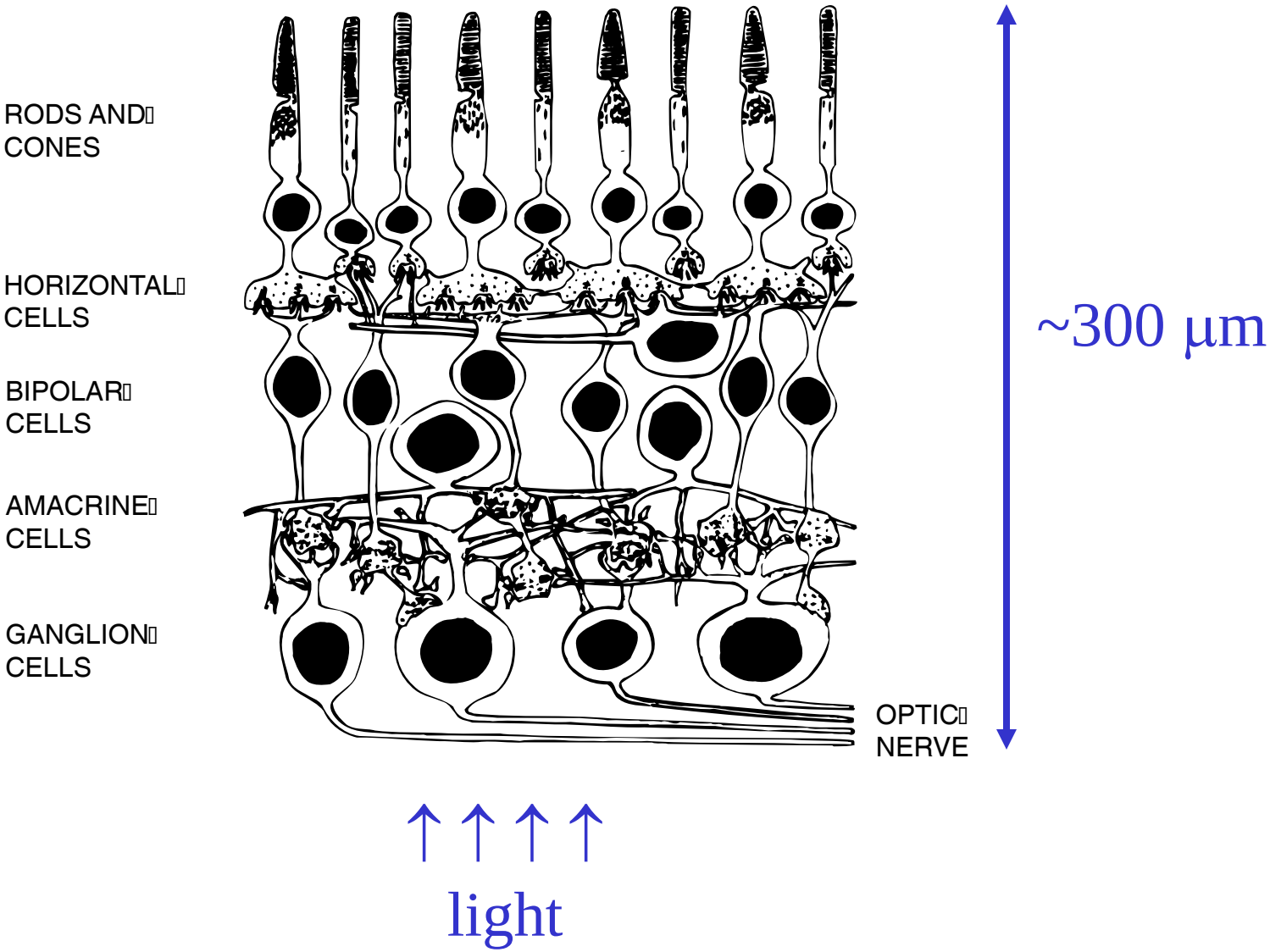
The Eye


Sclera
Choroid
Retina
Cornea
Fovea
Pupil
Lens
Iris
Ciliary body
Optic nerve

The Retina: an Advanced Pixel Detector

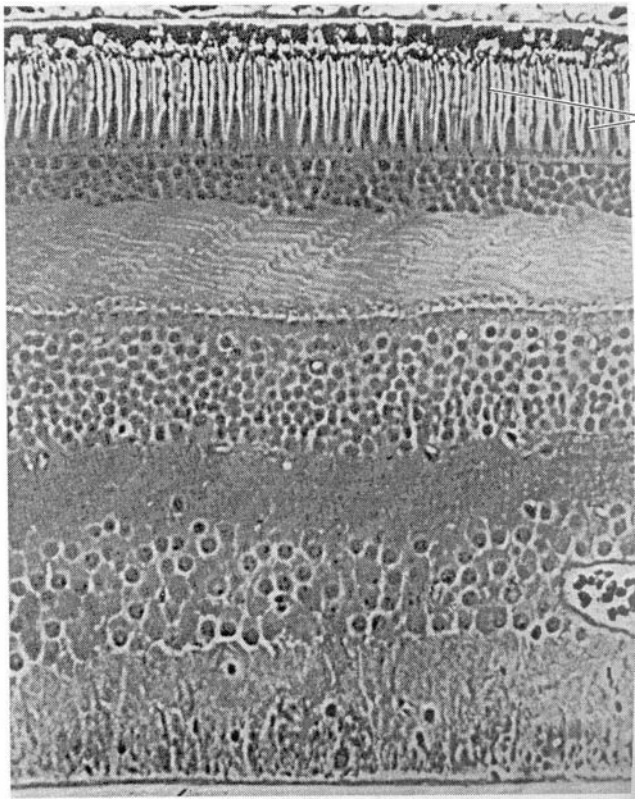
- thickness: ~300 μm
- active area: 10 cm^2
- number of pixels: 10^8
- number of output channels: 10^6
- compression factor (# input channels/# output channels): 100:1
- output signal width: ~1ms
- output format: analog, encoded by the frequency of digital signals
- spatial resolution: down to 2 μm
- 3D (depth perception): stereoscopic vision
- radiation hardness: non-rad-hard
- technology: mature, reliable, and in wide-spread use

The Retina


RODS AND
CONES
HORIZONTAL
CELLS
BIPOLAR
CELLS
AMACRINE
CELLS
GANGLION
CELLS
OPTIC
NERVE
~300 μm
light

The Retina

photoreceptors



outer plexiform layer

inner nuclear layer
(horizontal, bipolar,
amacrine cell bodies)

inner plexiform layer

ganglion cell layer

nerve fiber layer

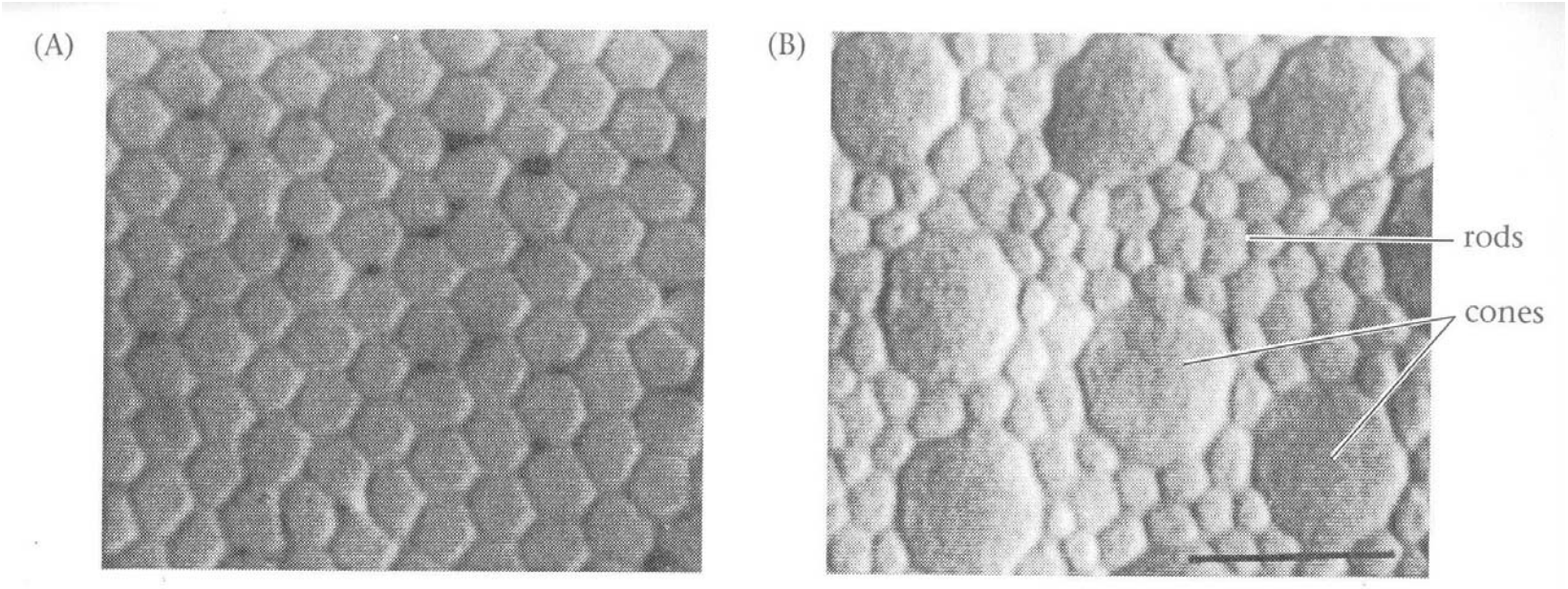
↑ ↑ ↑ ↑

light

The Retina:
pixel detector layout

cones in the fovea

rods and cones in the periphery



center-to-center spacing = 2.5 µm

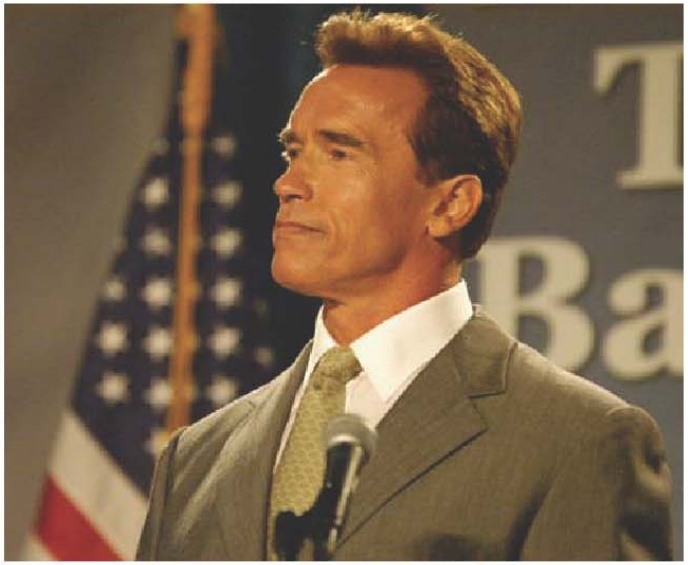
10 µm

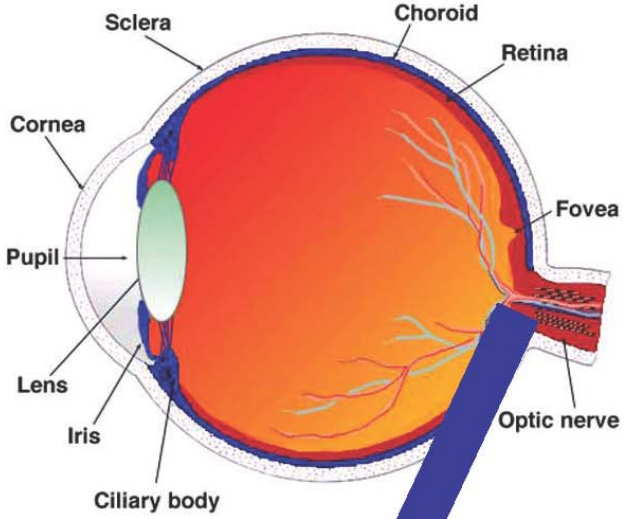
Probing the Retina

•**Goal:** understand how the retina processes and encodes dynamic visual images

•**Method:** record the patterns of electrical activity generated by hundreds of retinal output neurons in response to a movie focused on the input neurons

•**Technology:** based on silicon microstrip detector techniques and expertise developed for high energy physics experiments – an example of the application of expertise in HEP instrumentation to neurobiology




Sclera
Choroid
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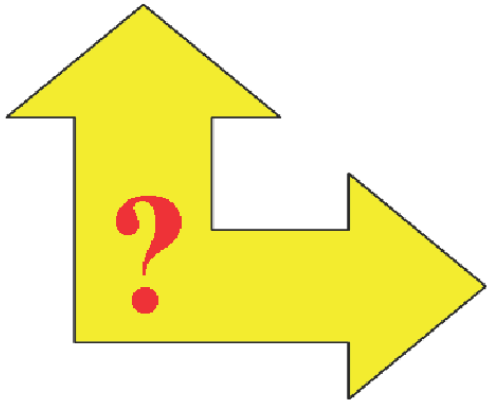
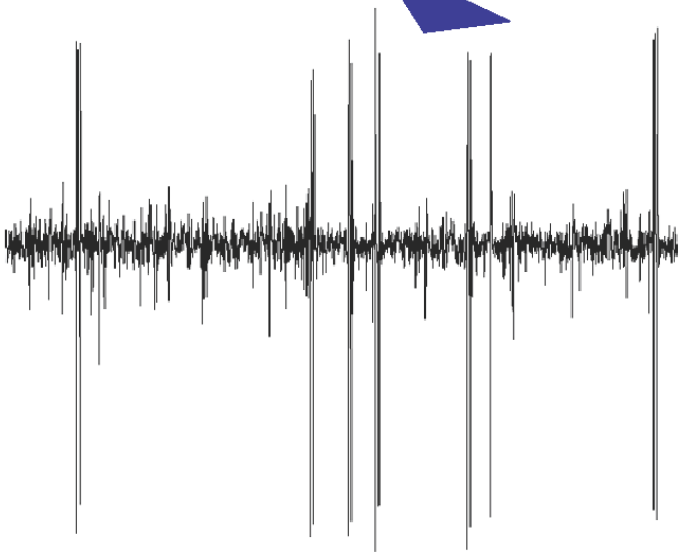
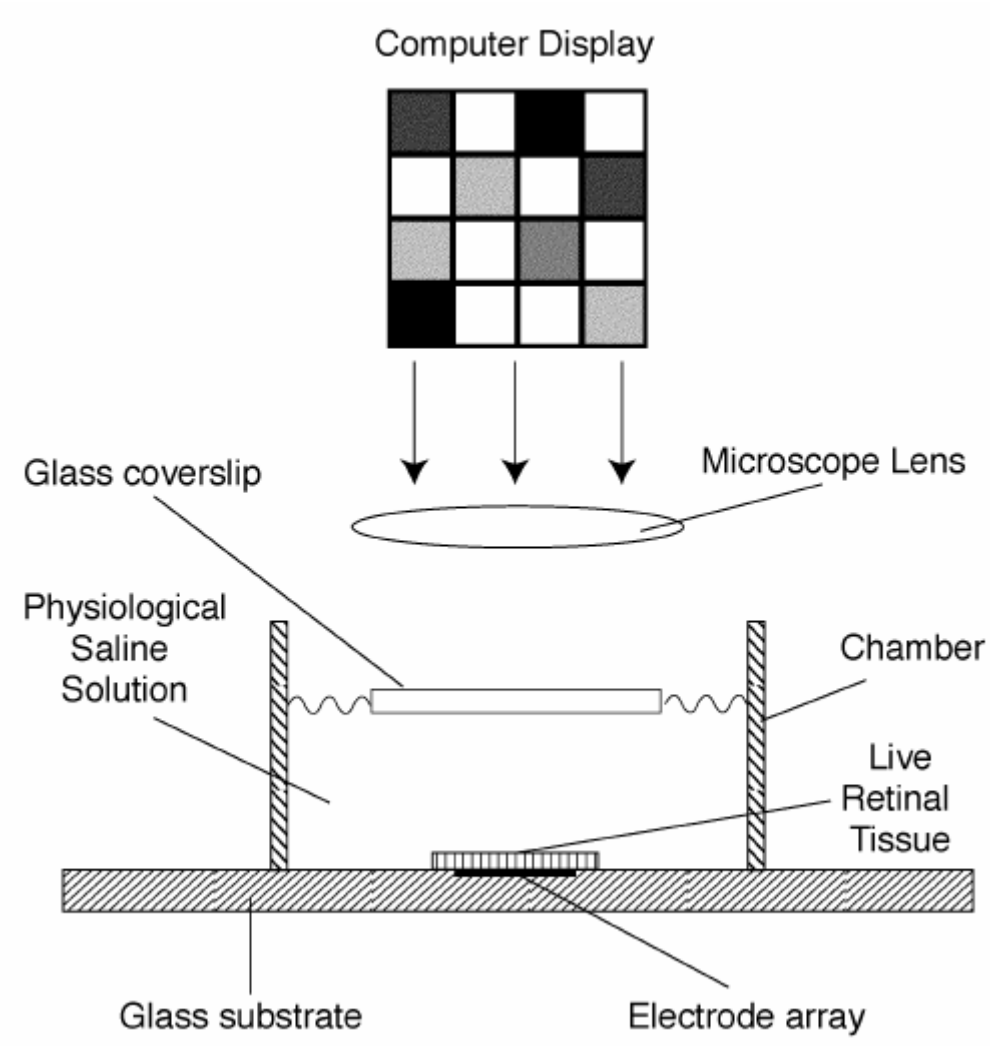
Optic nerve

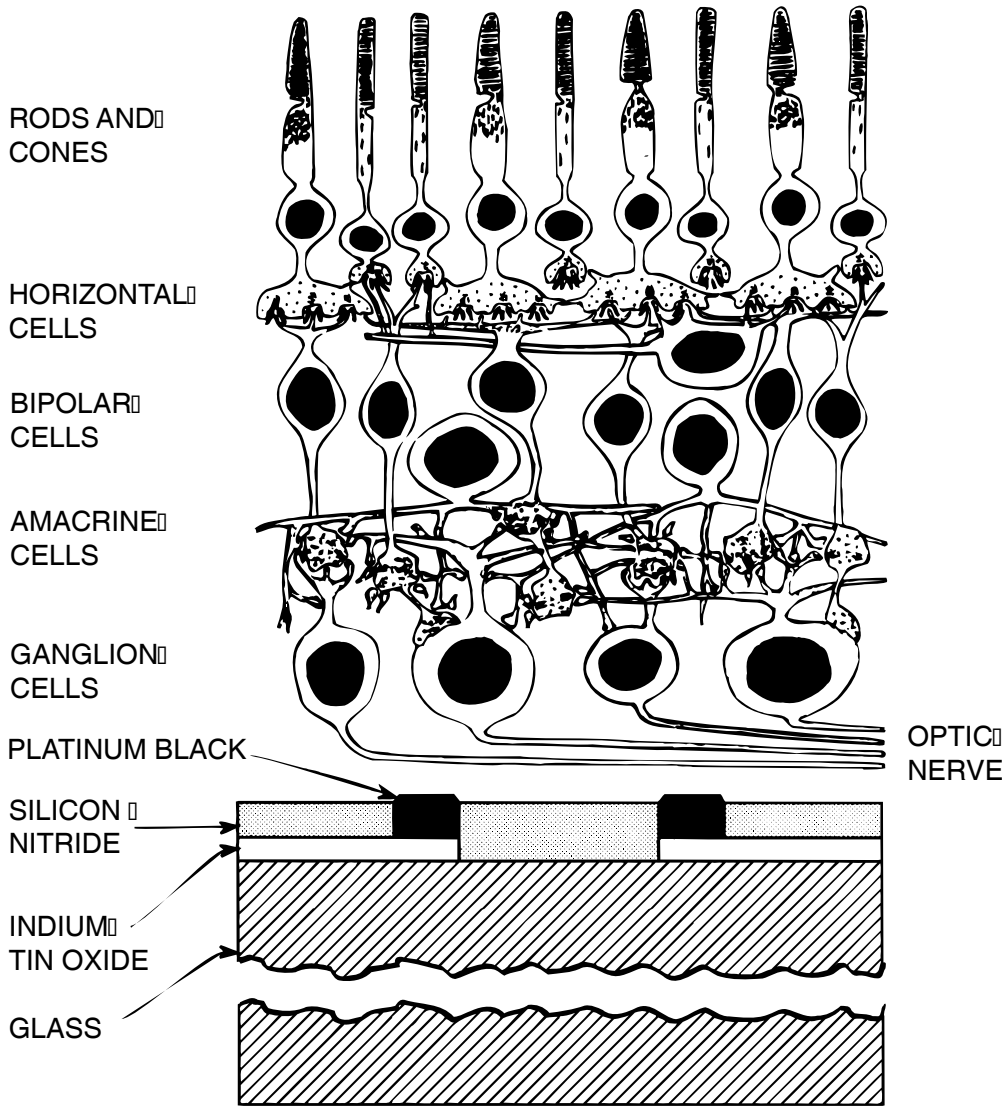

?



Experimental Technique

(based on work by Meister, Pine and Baylor)




RODS AND
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PLATINUM BLACK
OPTIC
NERVE
SILICON
NITRIDE
INDIUM
TIN OXIDE
GLASS

Species?

Guinea Pig, Monkey, Mouse

Scale?

•Record from a population of neurons approaching a scale of interest for neural computation

•order-of-magnitude improvement in state-of-the-art

$\Rightarrow$ Record simultaneously from hundreds to thousands of retinal ganglion cells in a single preparation

System Specifications

Spatial resolution: 30-60 μm electrode spacing

- efficiently detect/image RGCs with small active regions
- electrode density ≥ cell density

Time/ampl. resolution: 50 μs (20 kHz sampling freq.)/12 bits

- characterize spike waveforms well enough to identify the signals from individual neurons (time coincidence)

Sensitive area: 2 - 8 mm^2

- detect # of RGCs comparable to # of inputs used in the visual cortex for neural computation (~hundreds to thousands)
- collect sufficient statistics, even for quite rare RGC classes
- characterize tiling of visual field

Data collection time/experiment: ~24 hours

Implications

⇒ Large number of channels (512- 2048)
⇒ High data collection rate (15-60 MB/s)
⇒ Vast amount of data (1.2 – 4.8 TB per one-day experiment)

Compare to "traditional" neurophysiology:

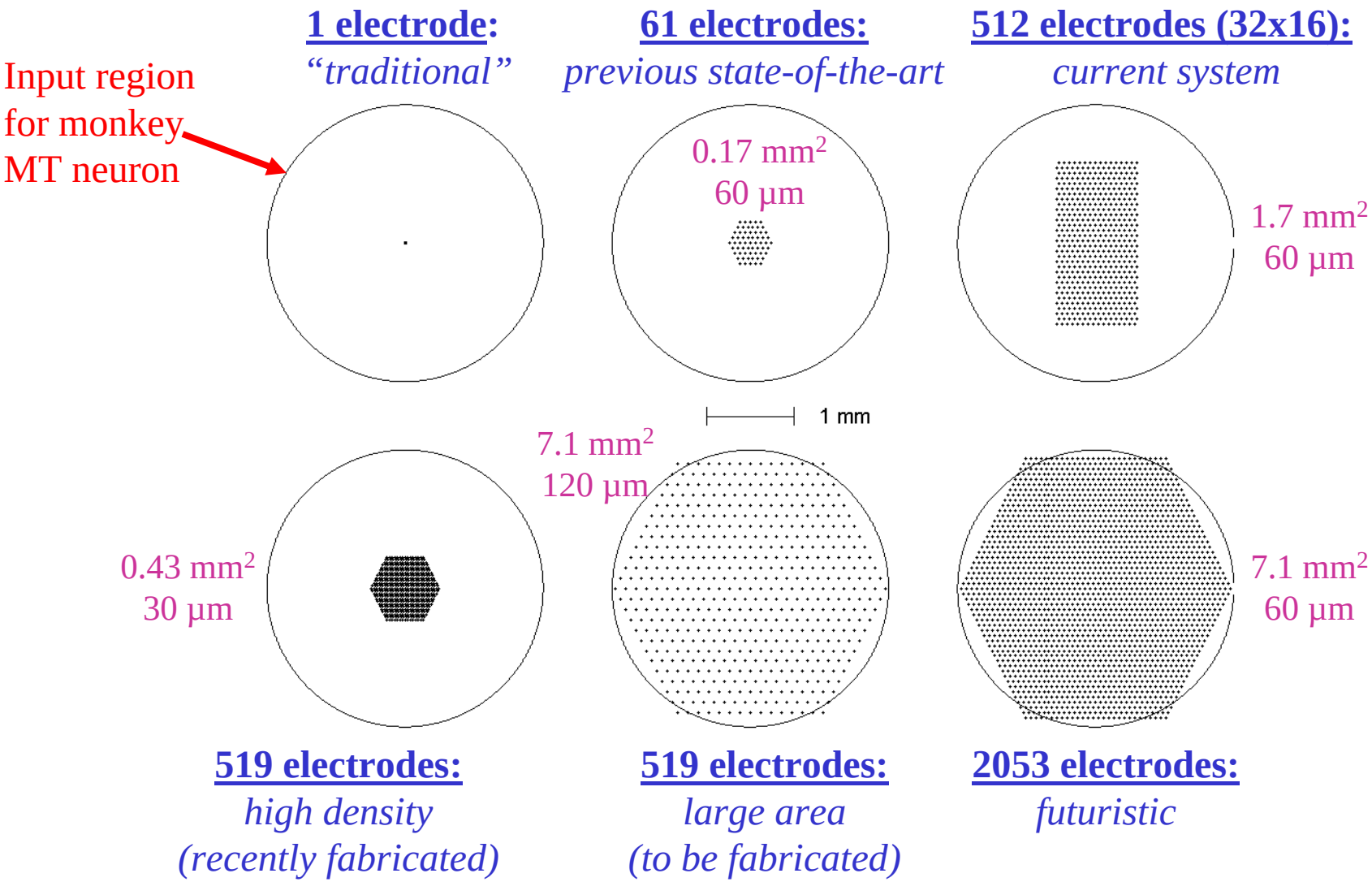
- Number of channels = 1
- Data collection rate = 30 kB/s
- Data/day = 2.5 MB

System Ingredients

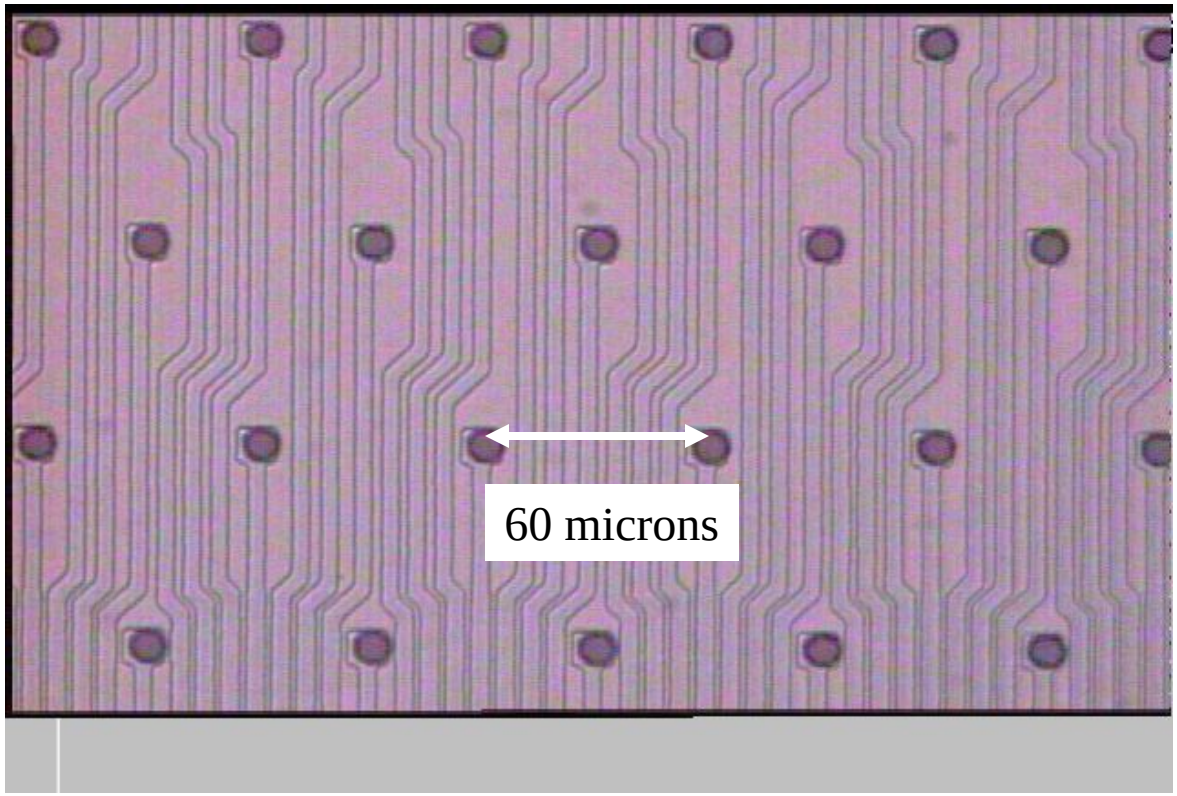
- High density/large area electrode arrays
- Multichannel VLSI analog/digital circuitry for readout
- High speed data acquisition and processing
- Automated (or at least semi-automated) data analysis

Electrode Array Geometries

(Electrode diameters = 5 µm; area and electrode spacing given below.)

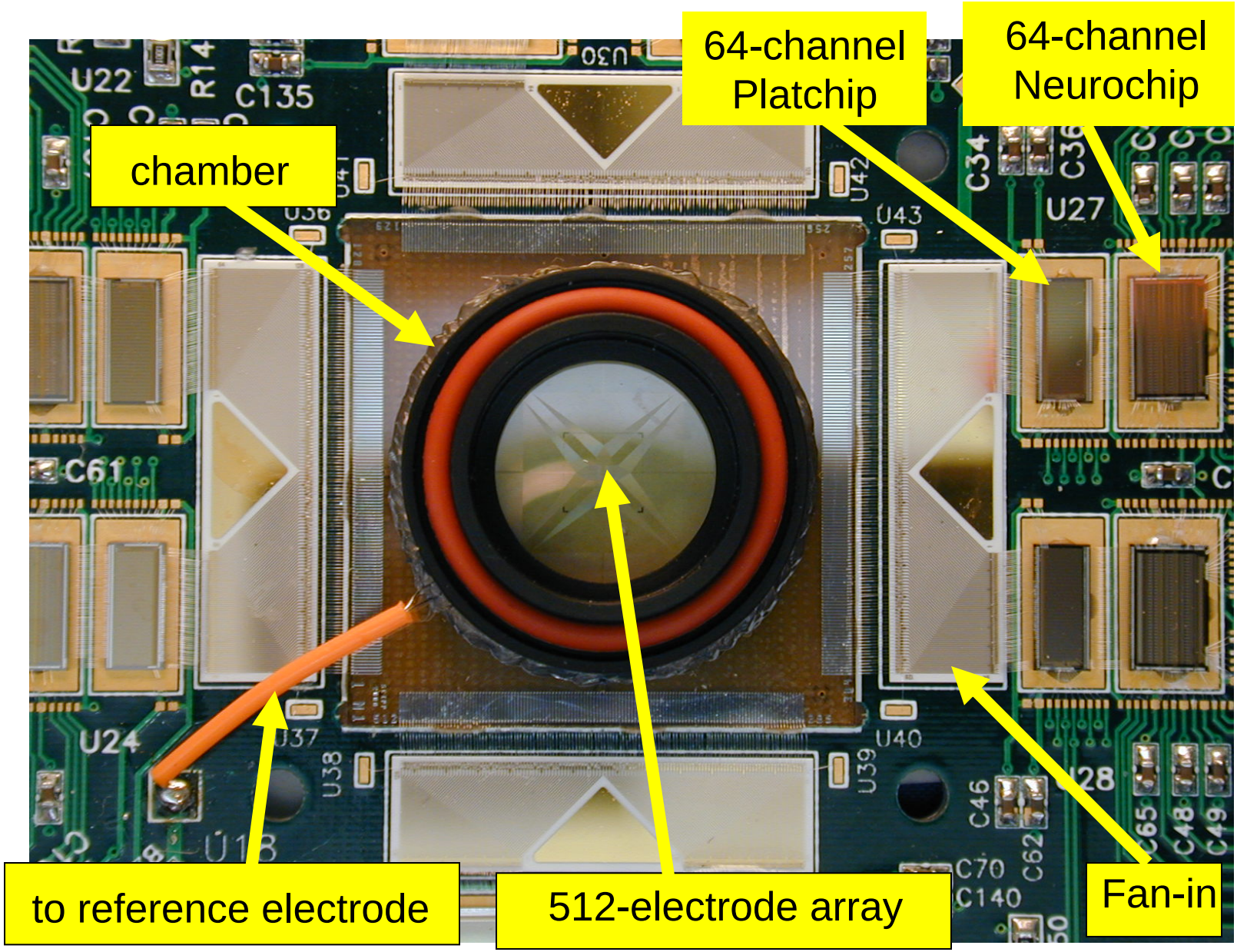


Section of
512-electrode Array (32x16)

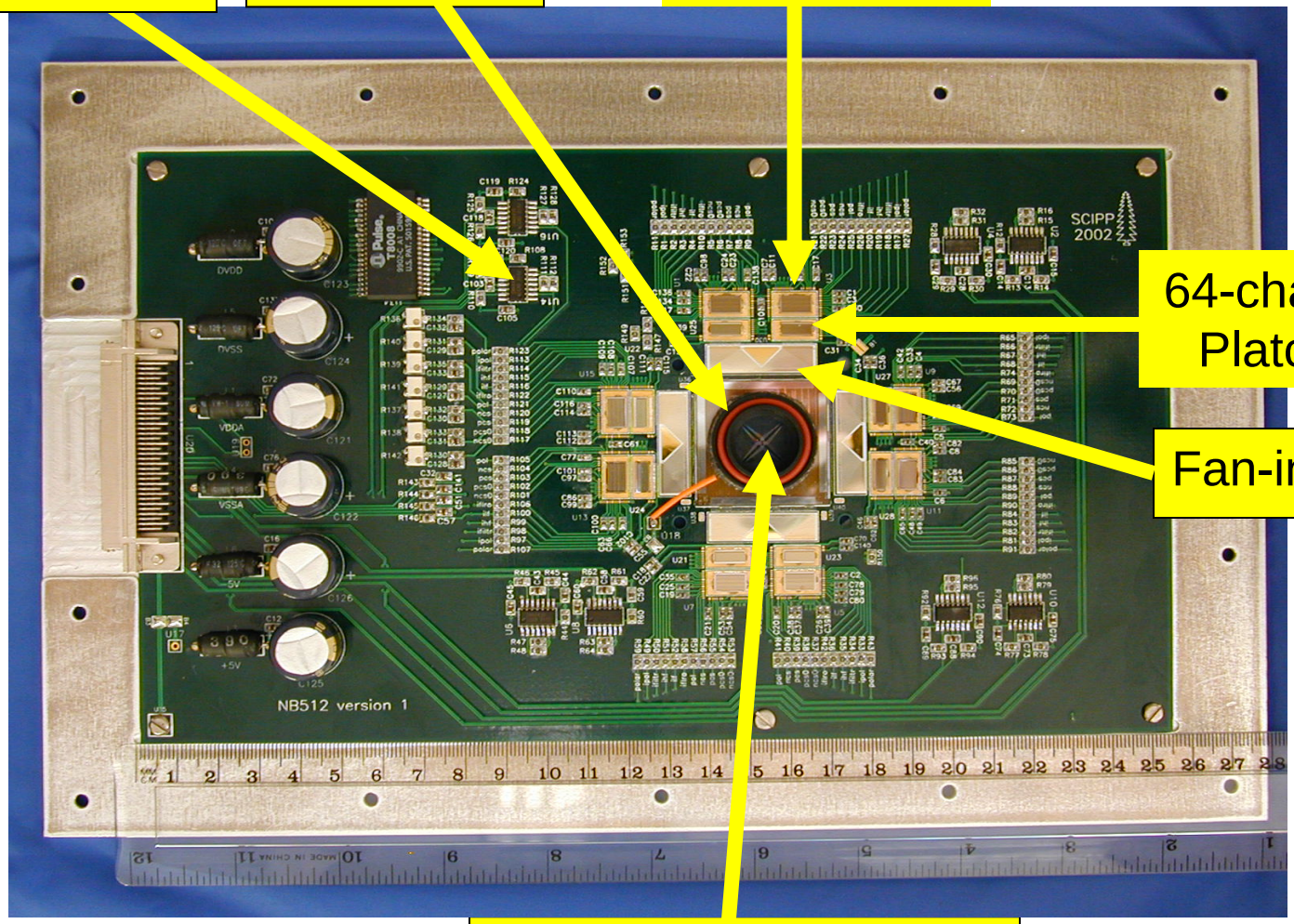


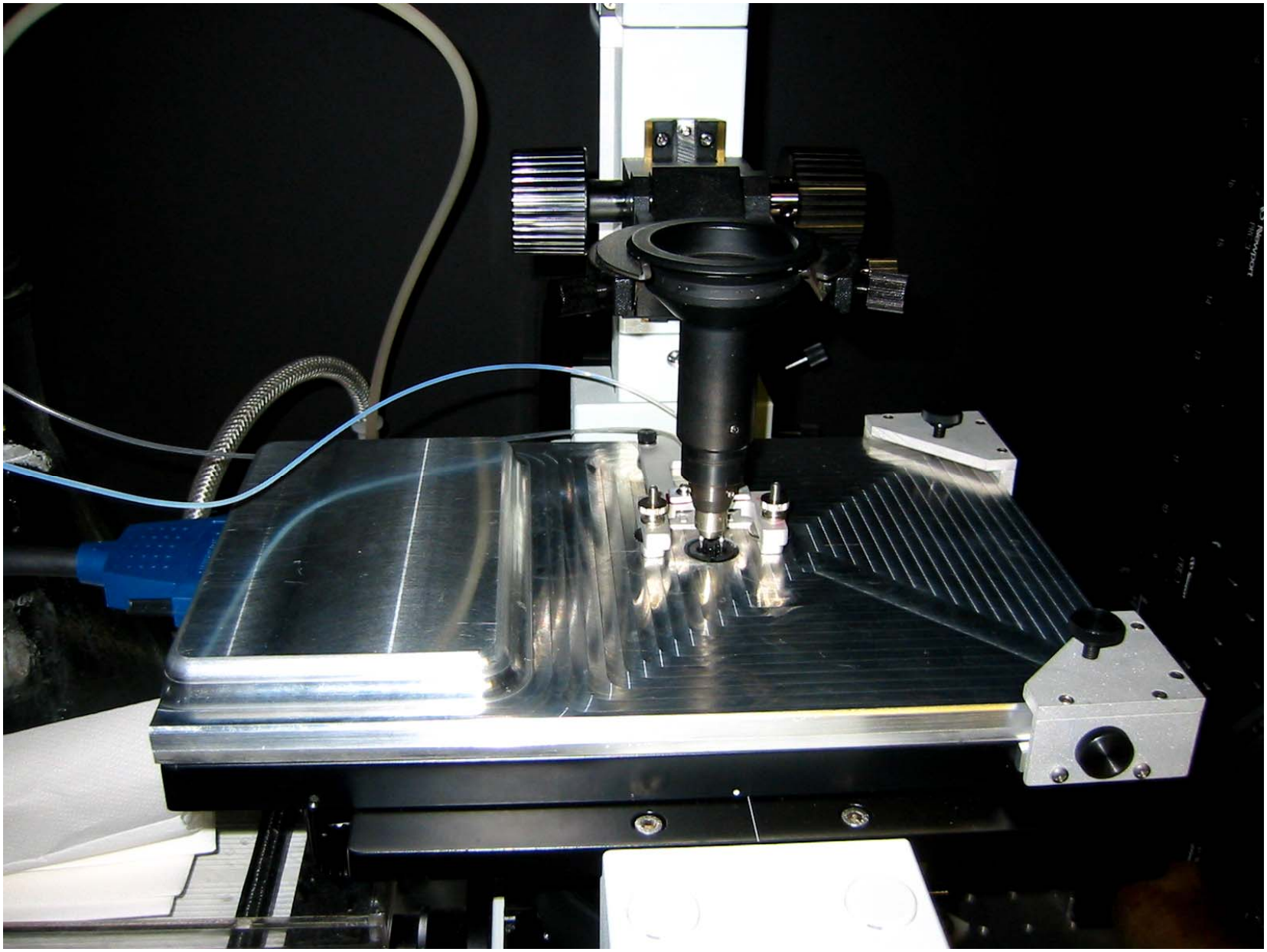
Electrode diameter = 5 μm

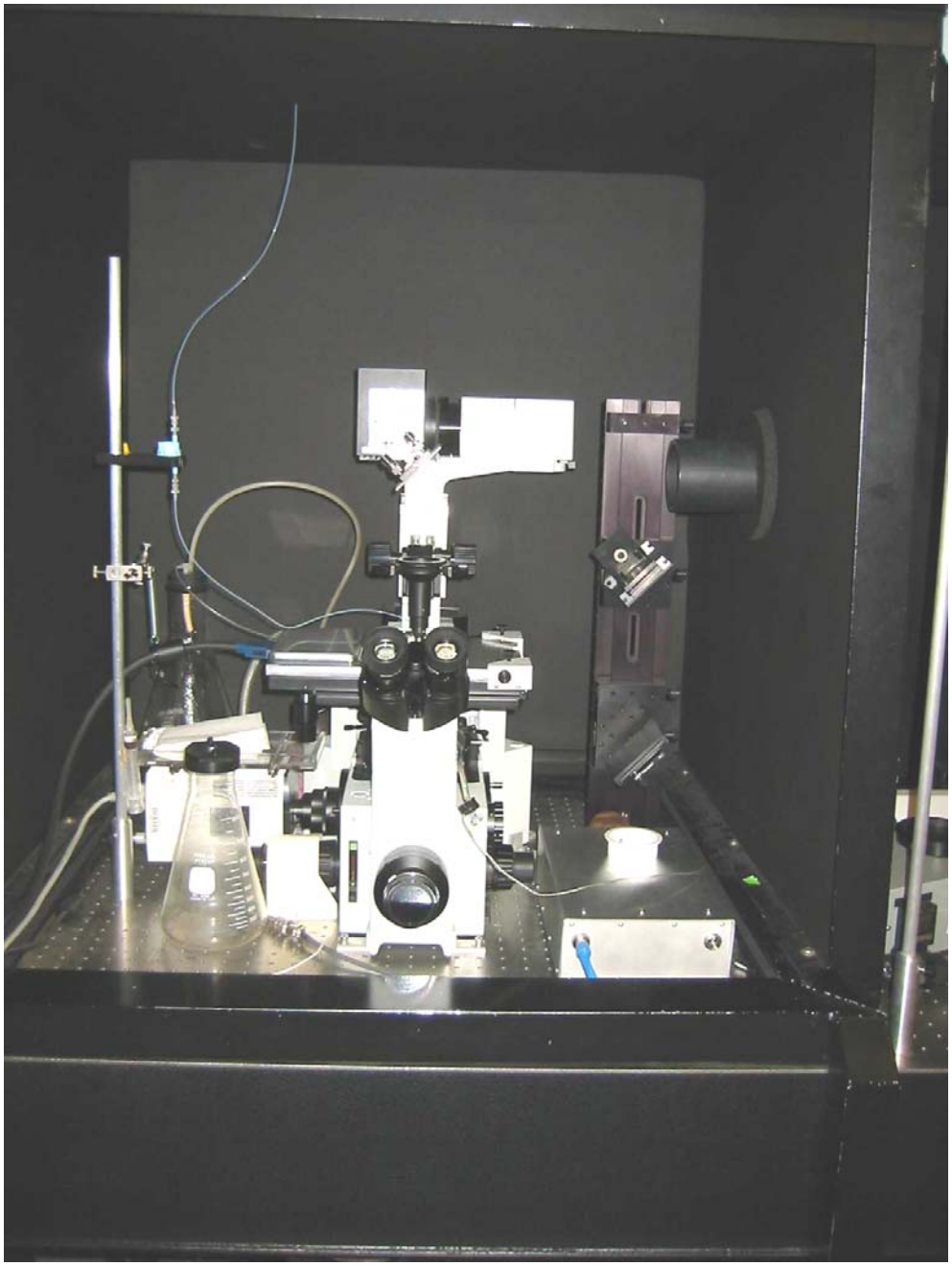
Section of 512-electrode “Neuroboard”

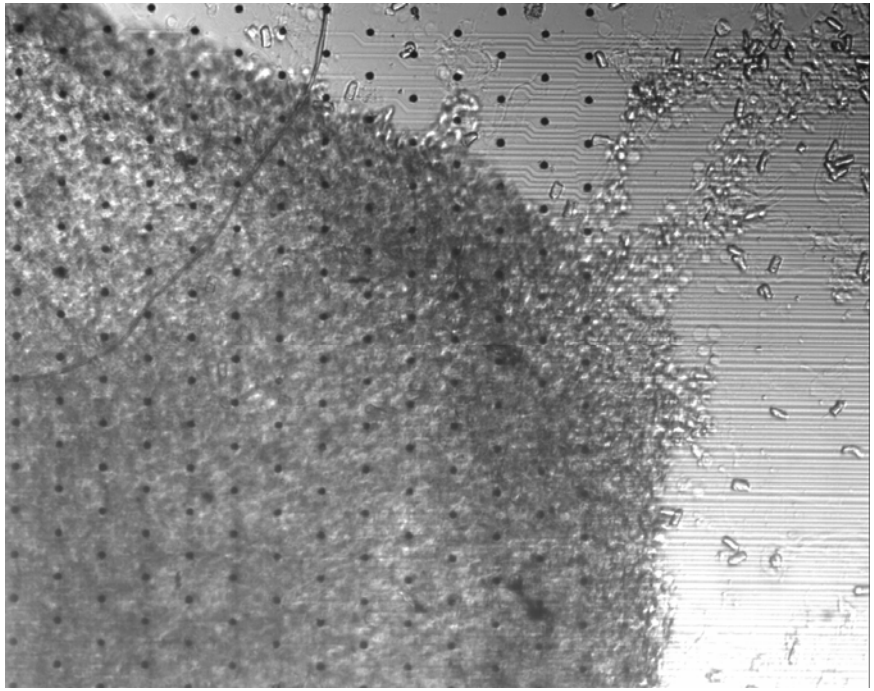


512-electrode “Neuroboard”

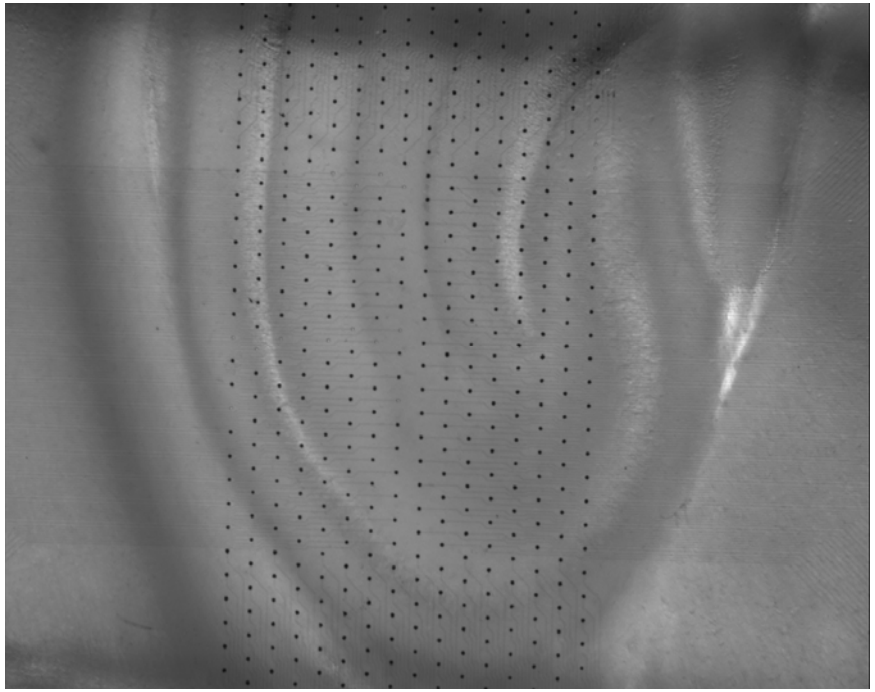






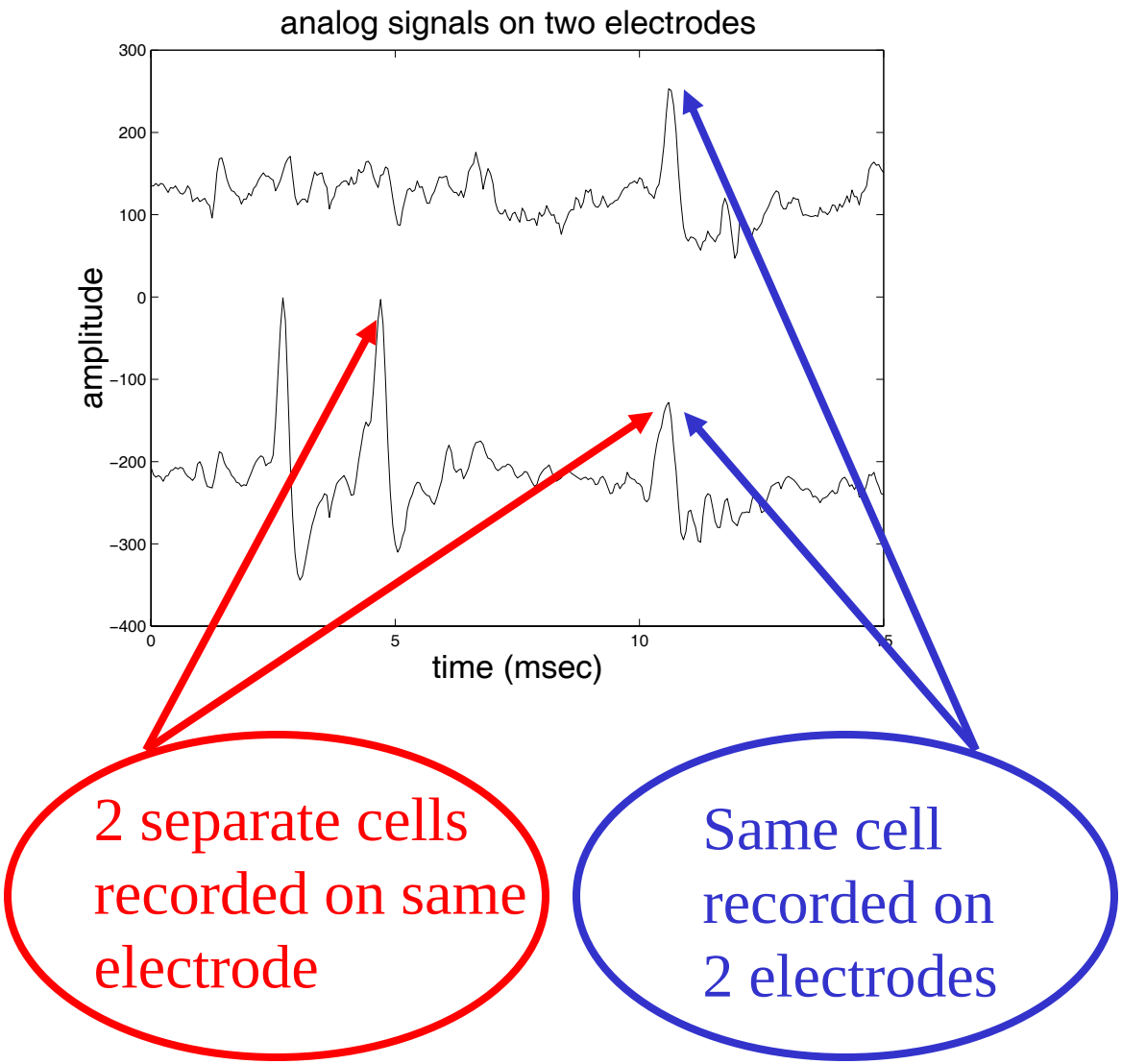


Salamander retina on 512-electrode array



Slice of hippocampal tissue on 512-electrode array

Spikes on electrodes ⇒ spikes from identified neurons

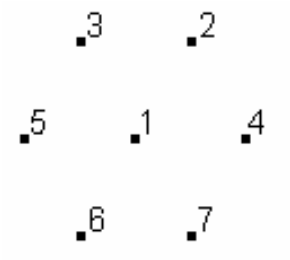


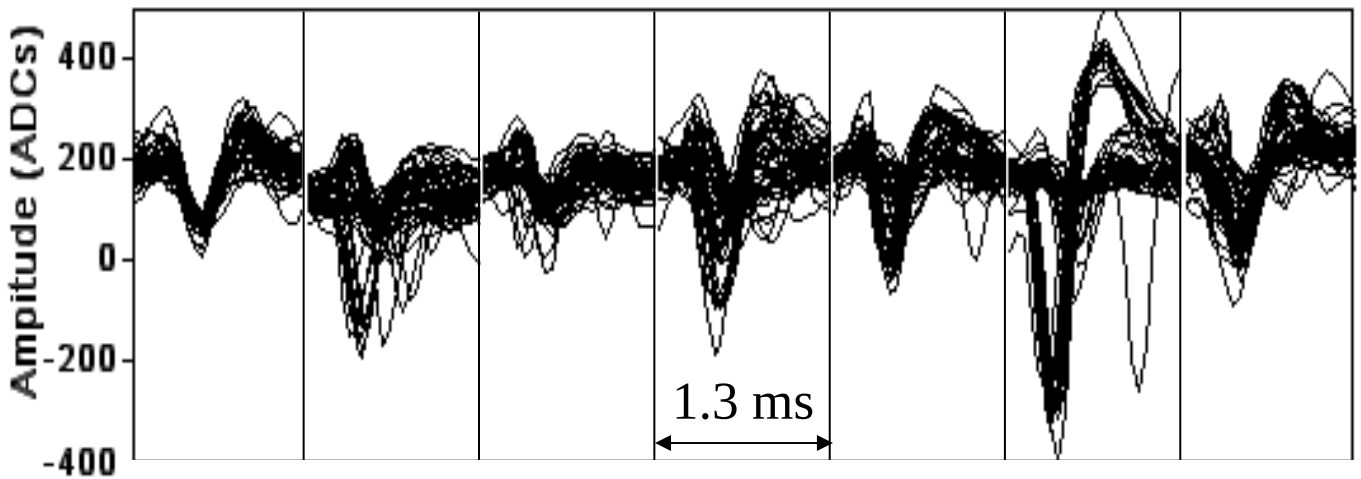
Neuron Identification

(signals on electrodes ⇒ spikes from identified neurons)

7x26=182 measurements

Multiple electrodes





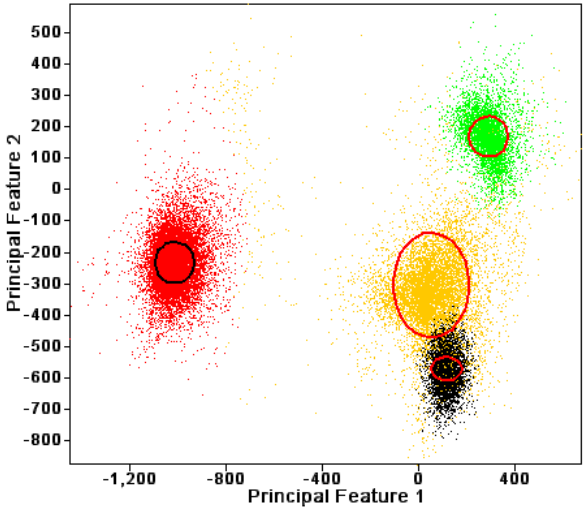
Electrode # 1 2 3 4 5 6 7

- Principal Components Analysis

Find ~5 most significant variables that are linear combinations of the 182 measurements

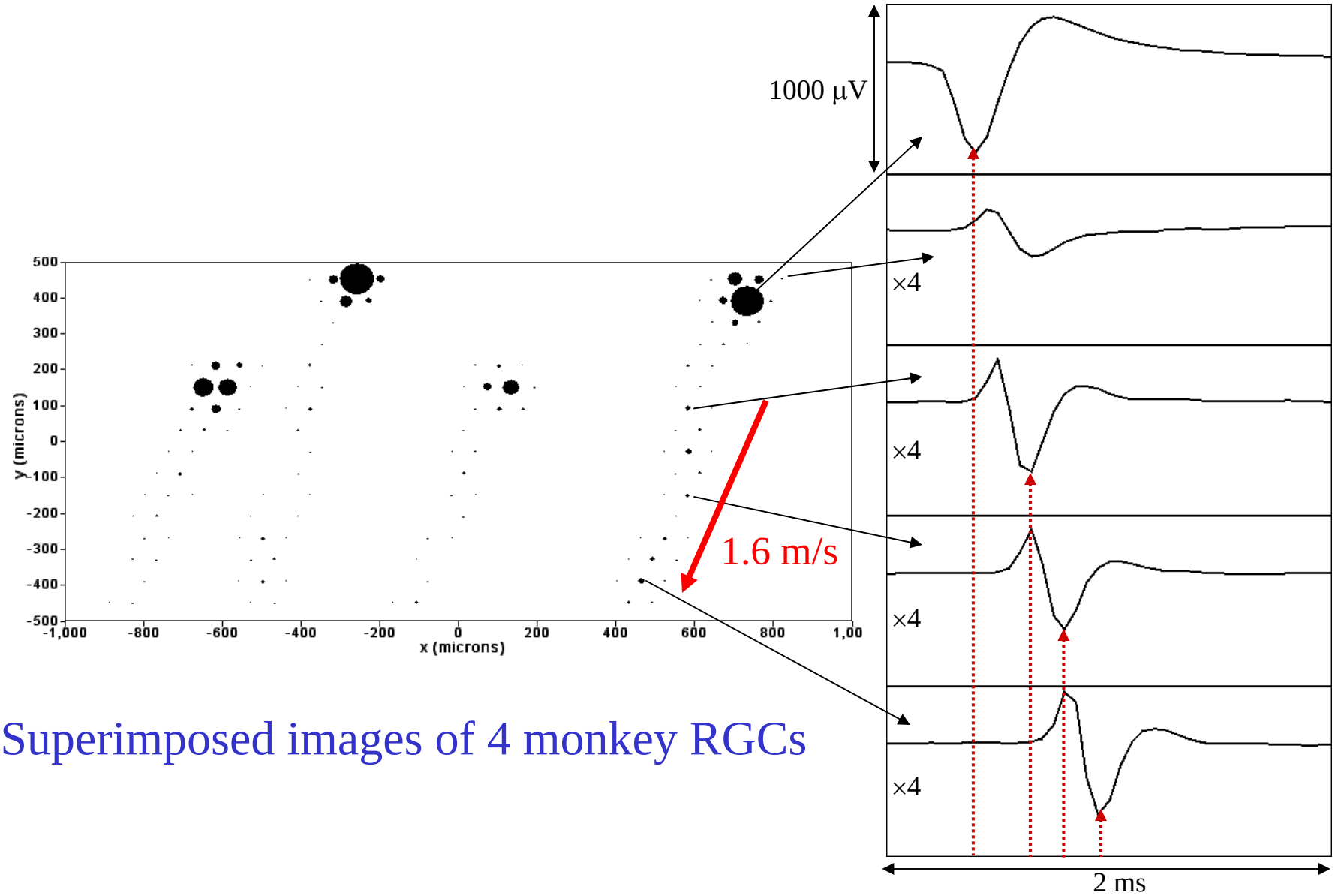
- Multidimensional Clustering

  ⇒ Identified Neurons



Software by D. Petrusca, SCIPP

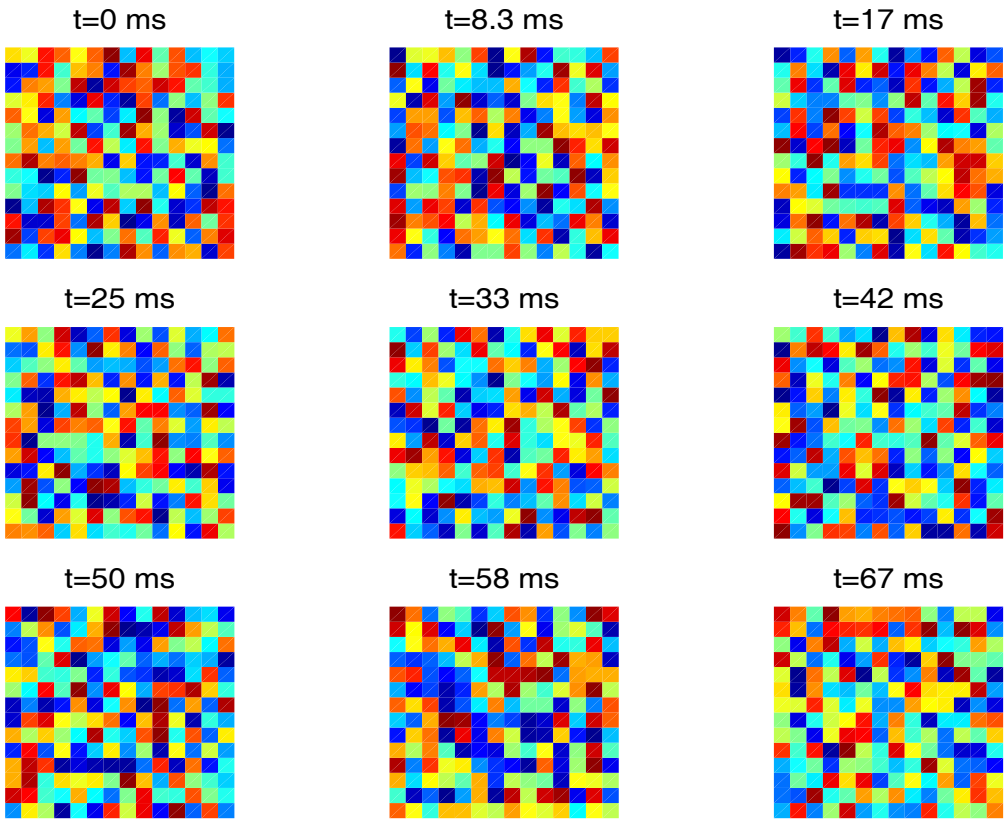
Electrophysiological Imaging



Superimposed images of 4 monkey RGCs

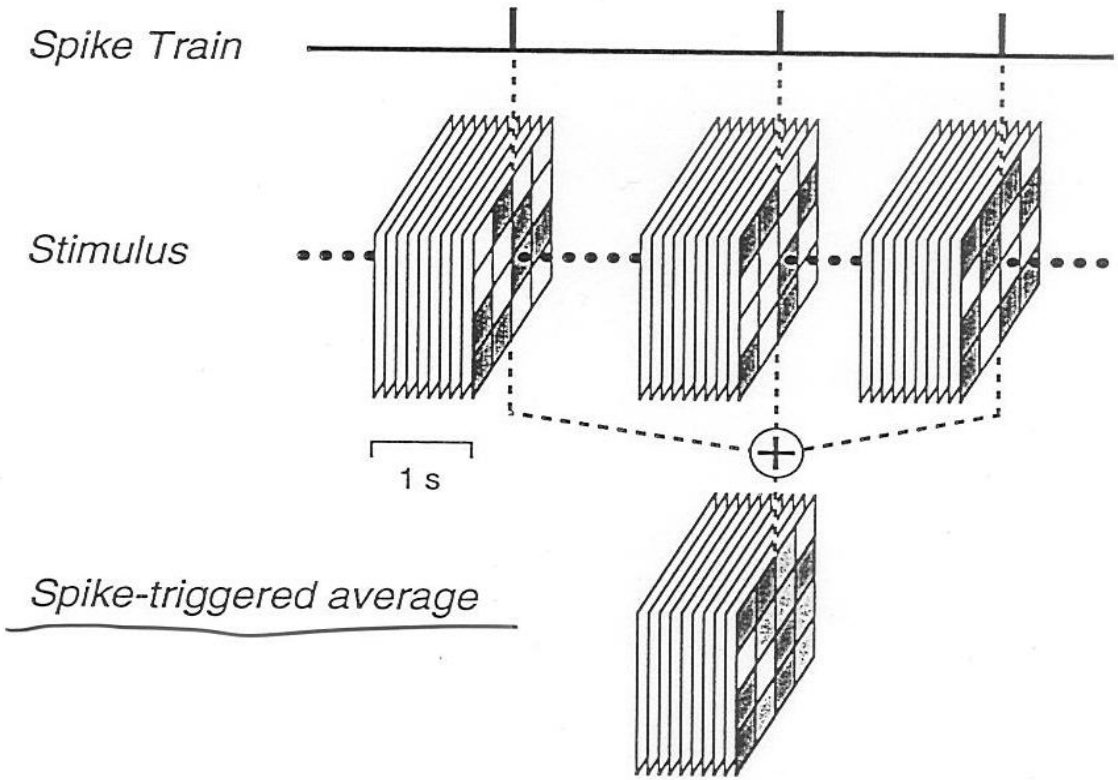
measure the response properties of identified neurons

⇒ white noise analysis: use time sequence of random checkerboard images



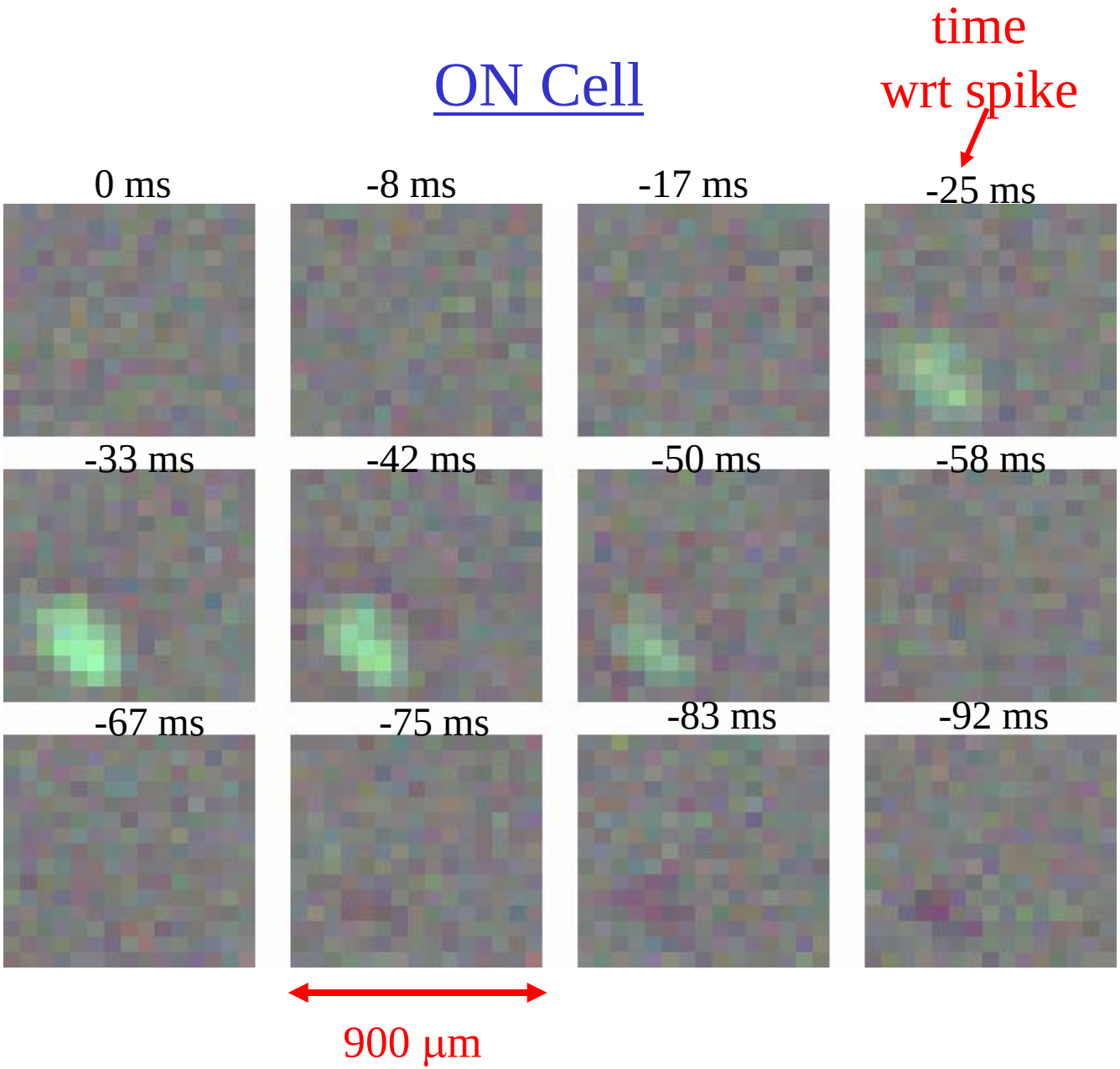
⇒ measure the "spike-triggered average" (sta) response for each neuron

Spike-triggered Average

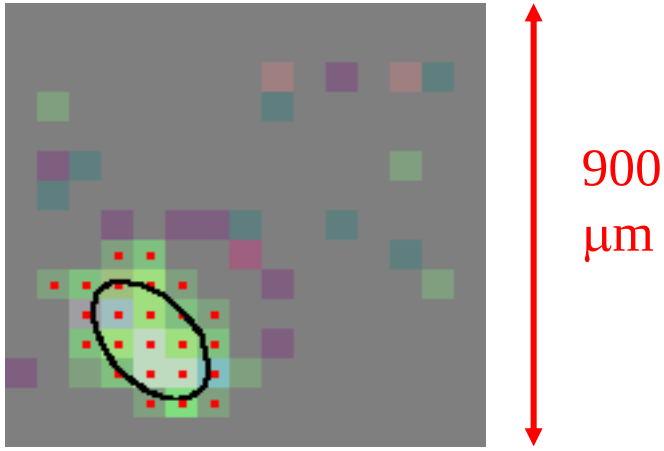


Monkey Retinal Ganglion Cell

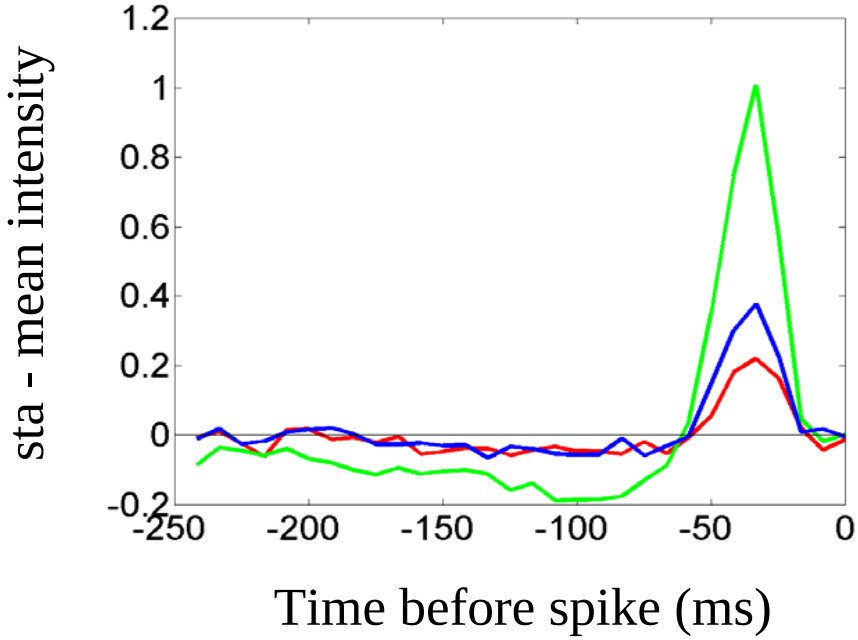
ON Cell

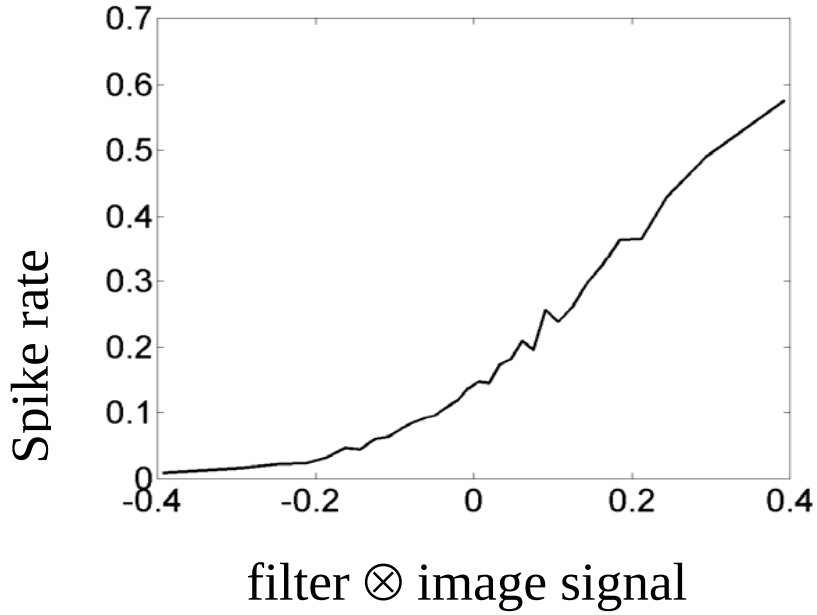


Spike-triggered
average image
at time of maximum
absolute intensity



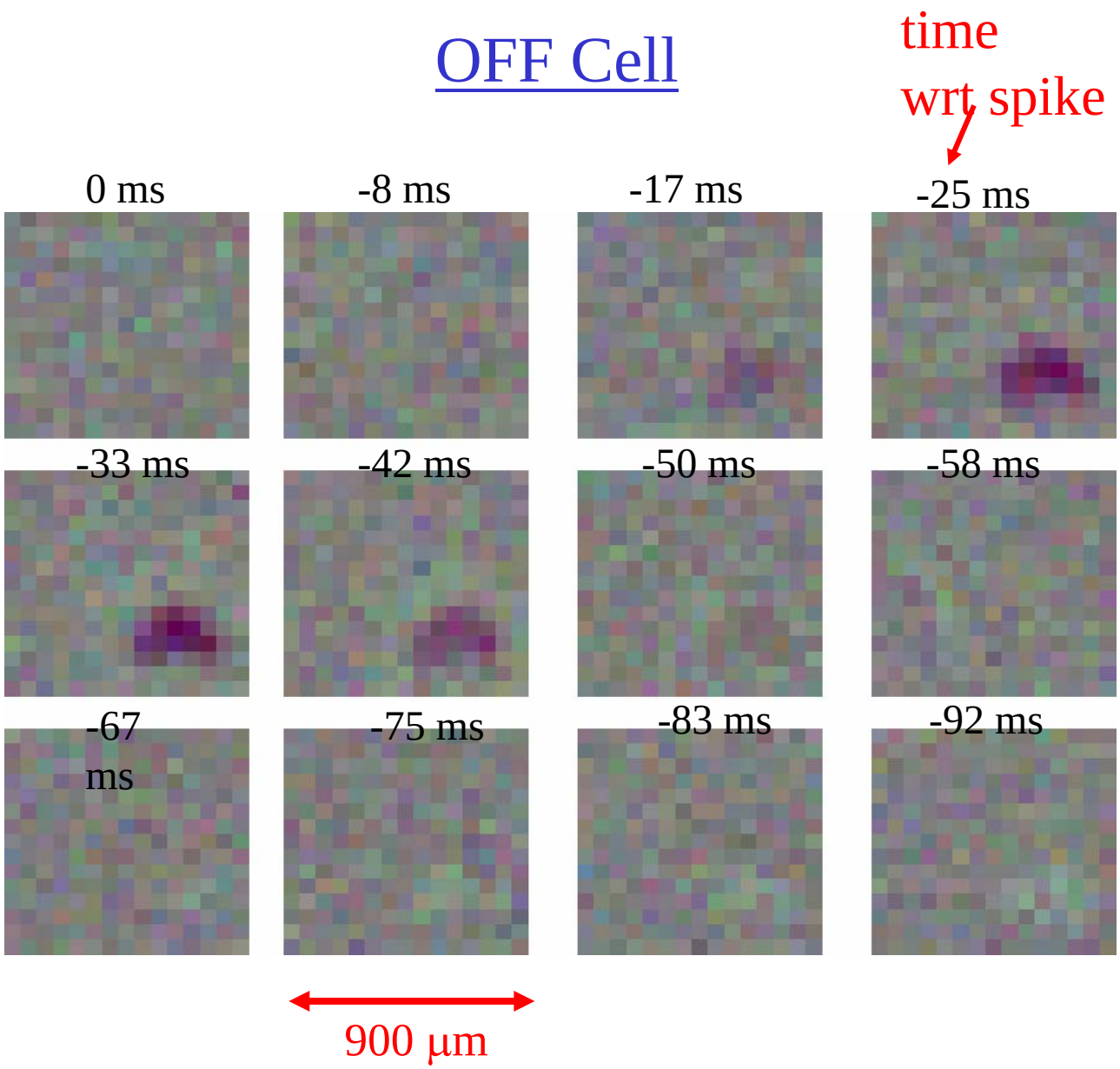
900
μm



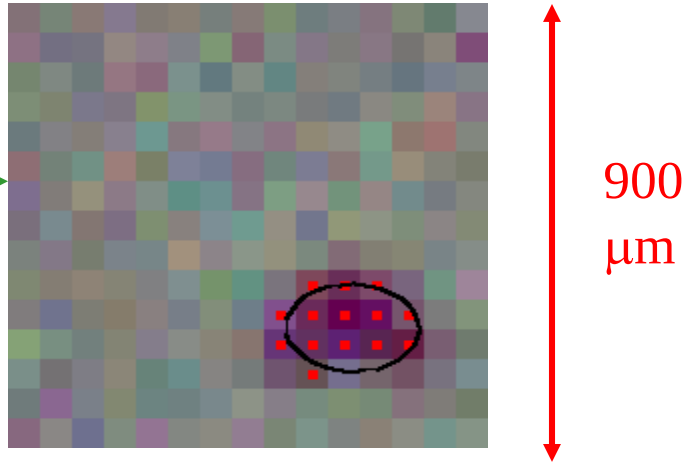


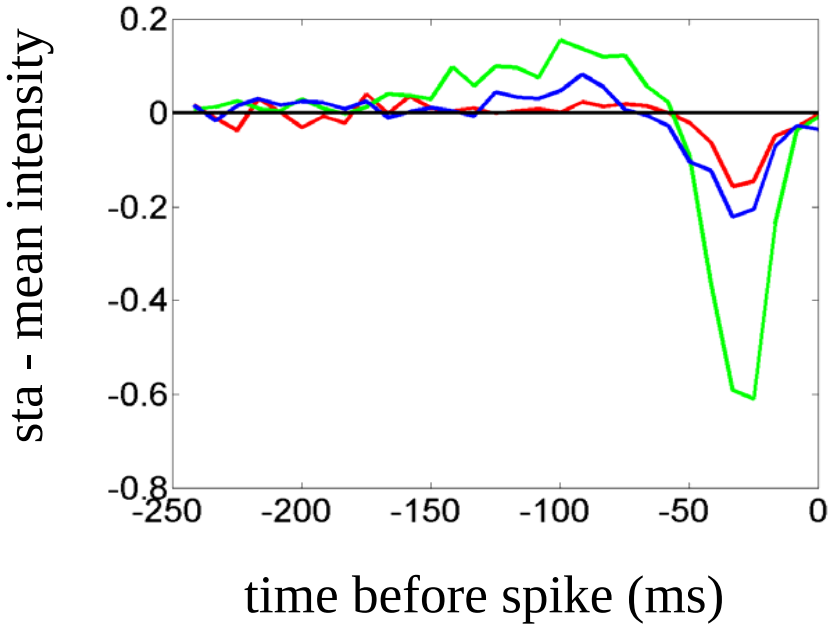
Monkey Retinal Ganglion Cell

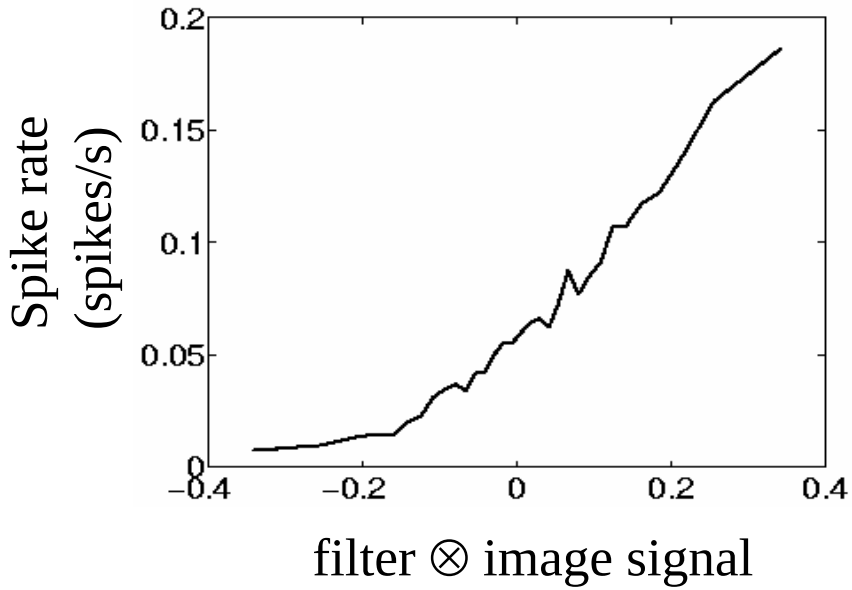
OFF Cell



Spike-triggered
average image
at time of maximum
absolute intensity

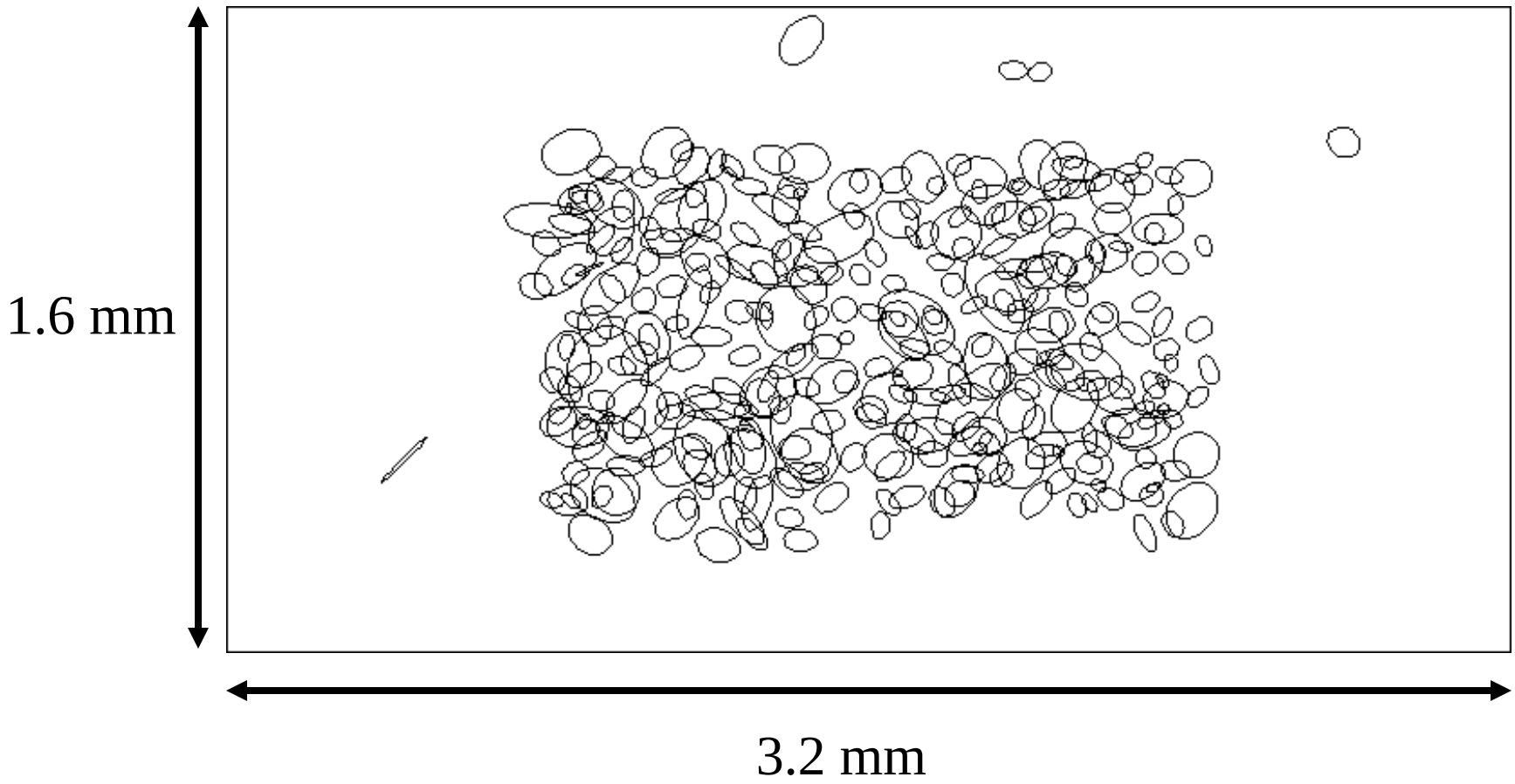






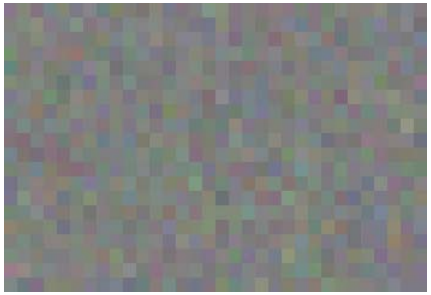
Some first (preliminary) results with monkey retina

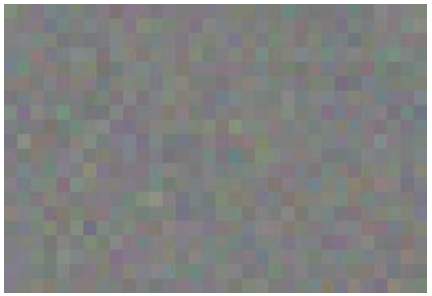
Light-sensitive regions ("receptive fields") for 338 identified neurons



Spatial/temporal response properties of individual neurons ("spike-triggered average")

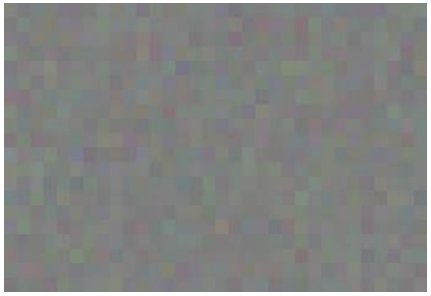
ON-parasol





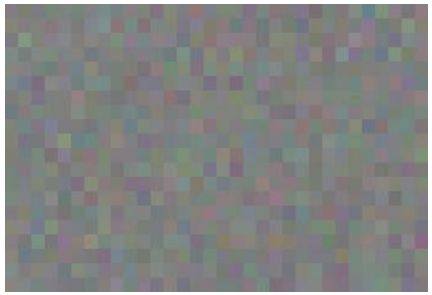
OFF-parasol

ON-midget



OFF-midget

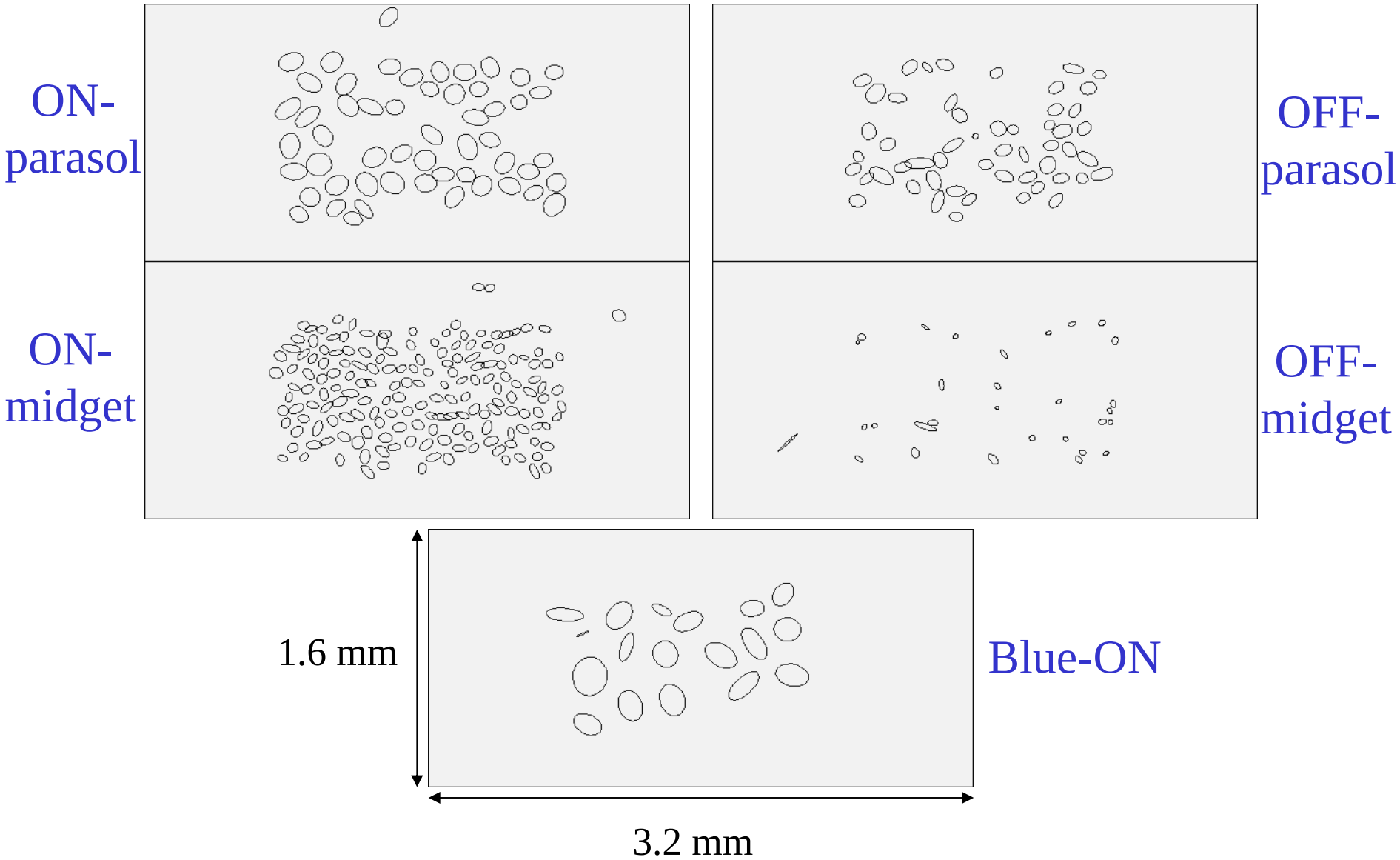
500 μm



Blue-ON

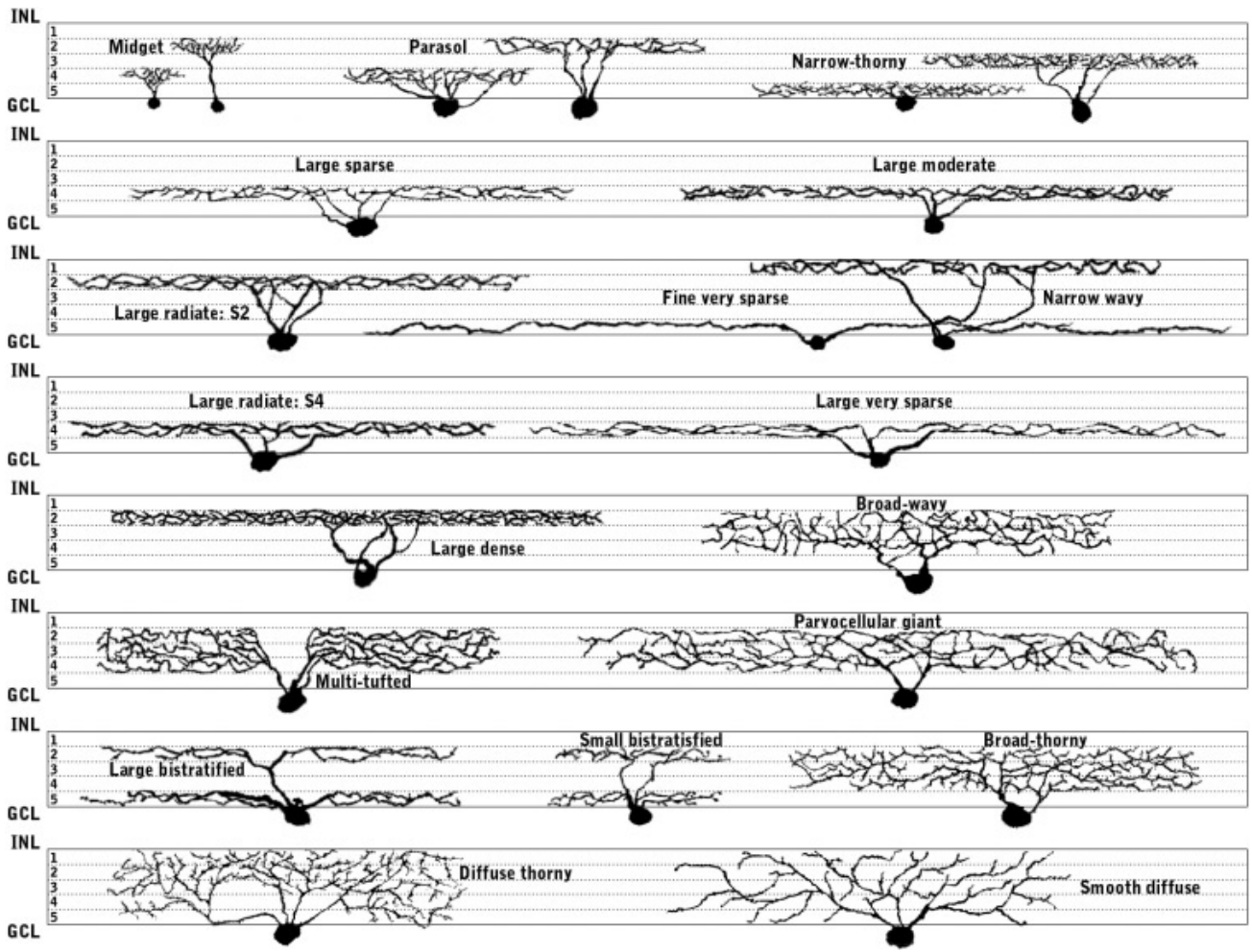
800 μm

(8.3 ms/frame)


ON-
parasol
OFF-
parasol
ON-
midget
OFF-
midget
1.6 mm
Blue-ON
3.2 mm

Five identified monkey RGC classes (already well-known), but this is just the tip of the iceberg.

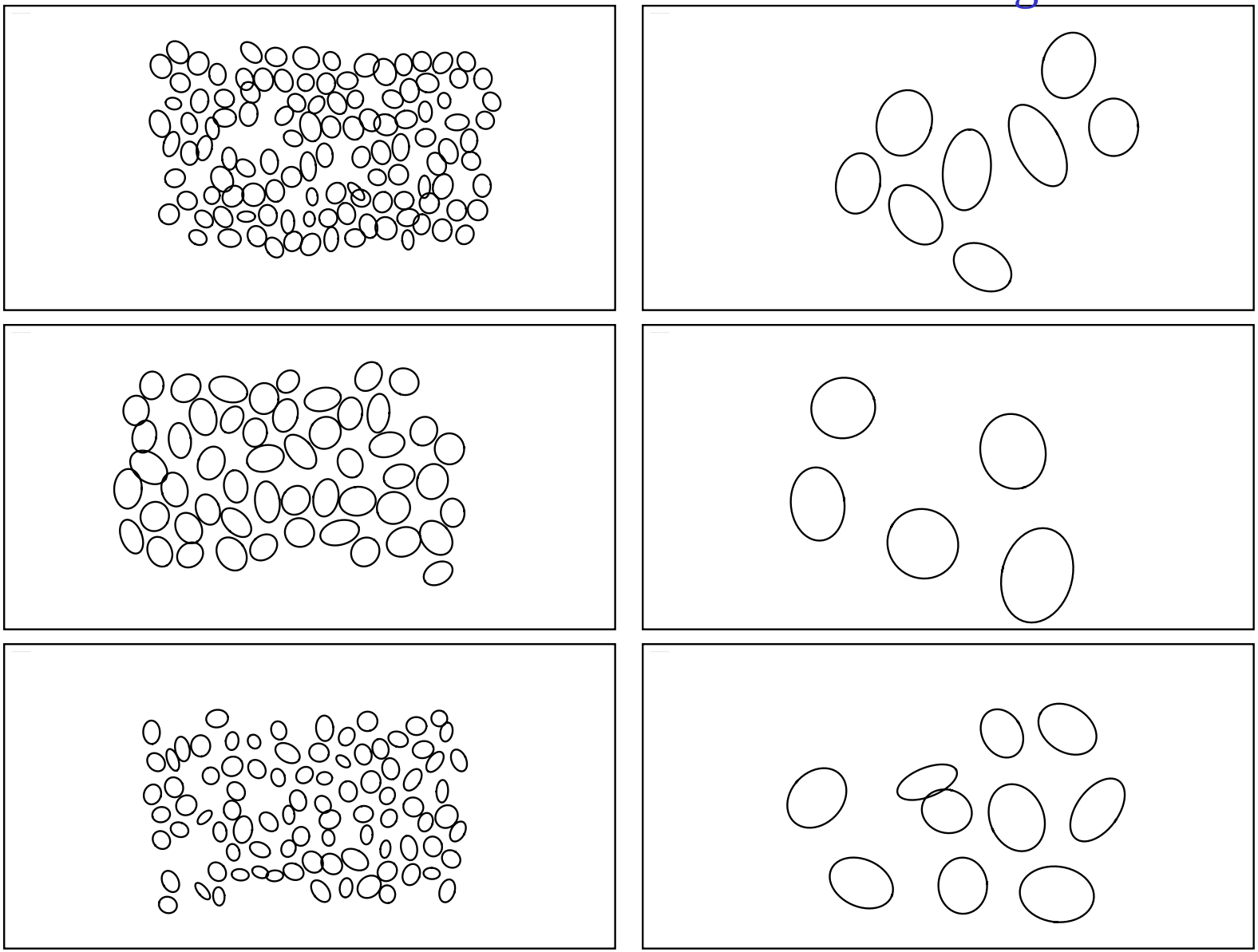
From anatomical studies, it is estimated that there are at least 22 distinct types of monkey RGCs.



Yamada, Bordt, and Marshak, Visual Neuroscience 22 (2005) 383.

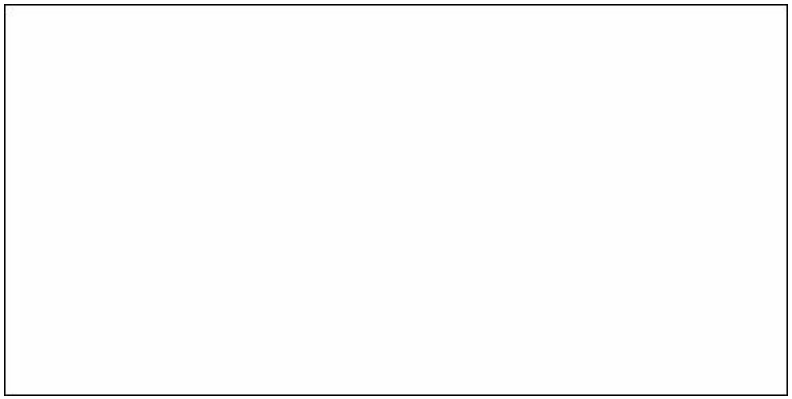
OFF Parasol

OFF Large

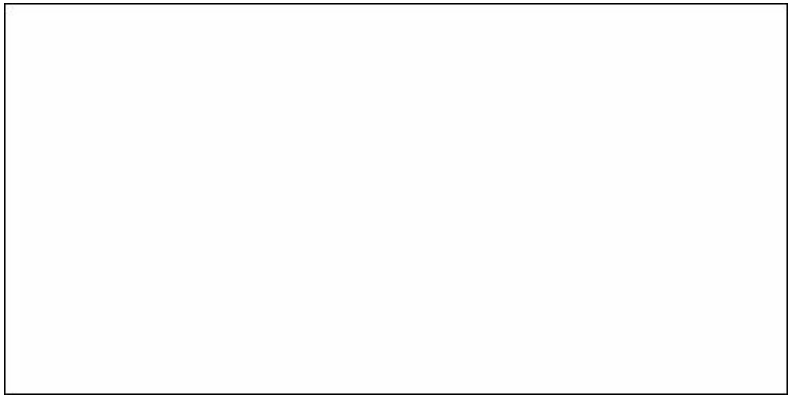


D. Petrusca et al., Soc. for Neuroscience annual meeting (2005)

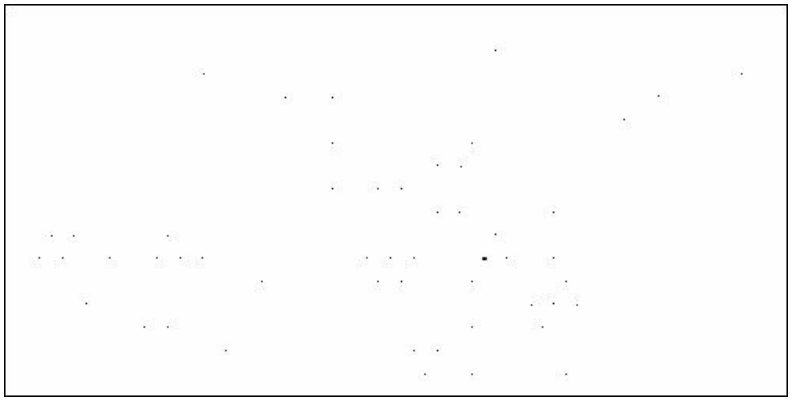
OFF Parasol



OFF Large



Amacrine

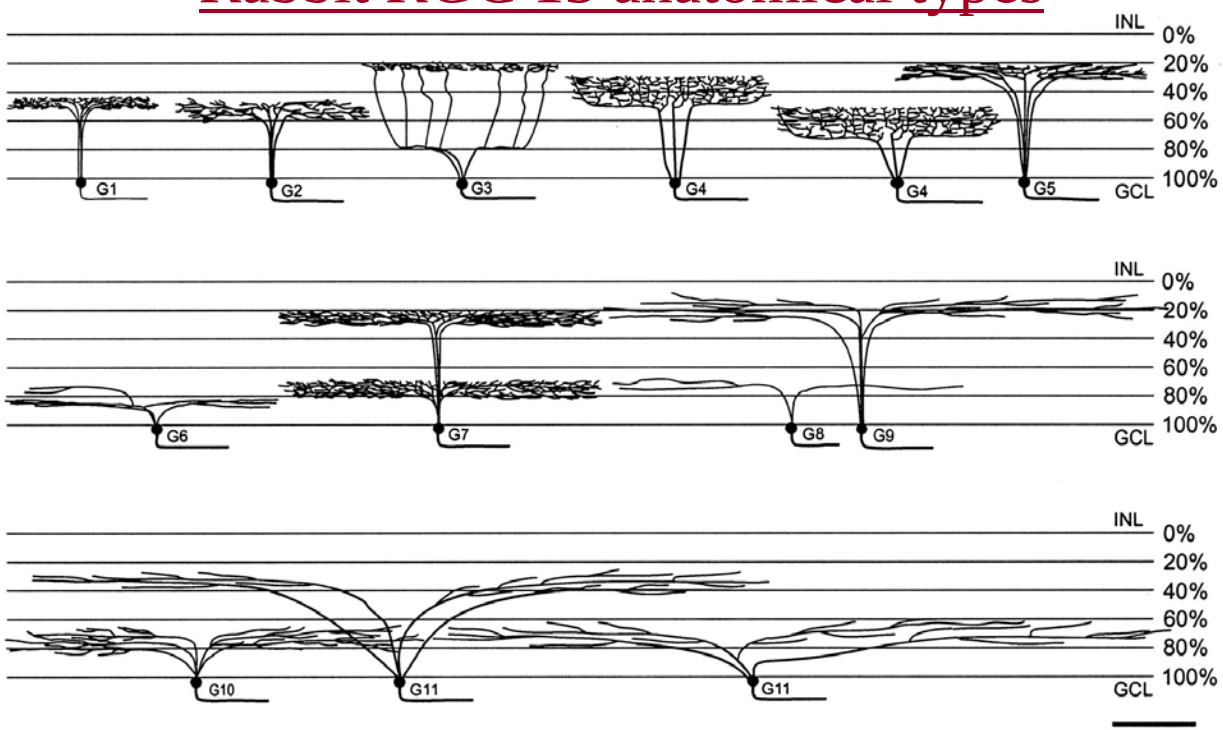


D. Petrusca et al.

What about the guinea pig retinal ganglion cells?

Not well studied, anatomically or physiologically. Much better studied is the rabbit retina. It is known that there are at least 13 distinct anatomical types and ~11-15 physiological classes of rabbit RGCs (only ~8 anatomical types have been well-matched with physiological classes)

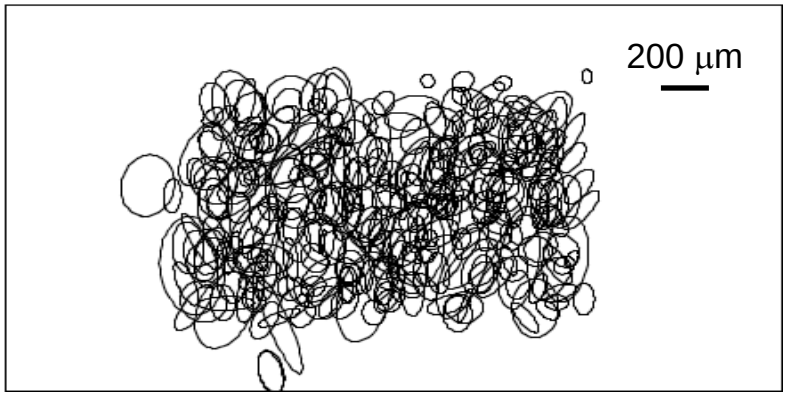
Rabbit RGC 13 anatomical types



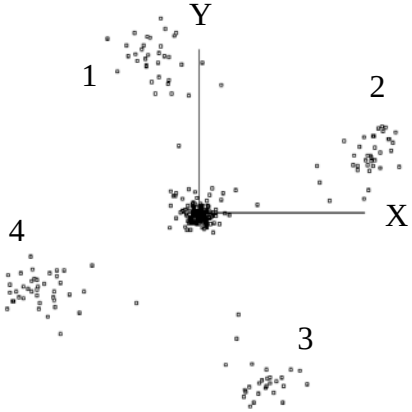
Rockhill et al., J. Neuroscience 22 (2002) 3831

Guinea Pig Retinal Ganglion Cells: OFF cells

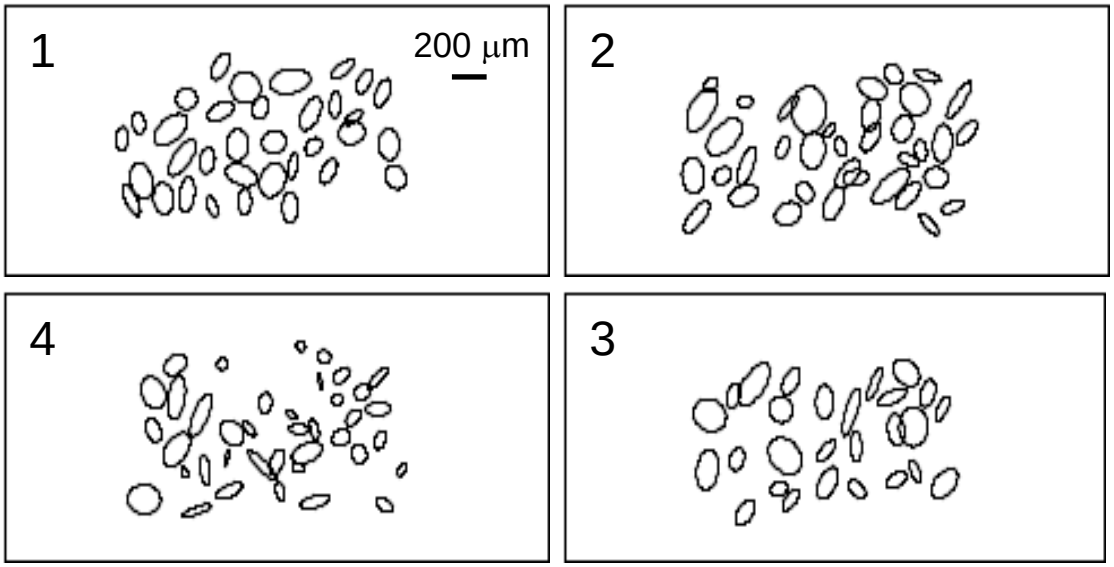
RF mosaic for 311 OFF cells



Direction selectivity for drifting sinusoidal gratings

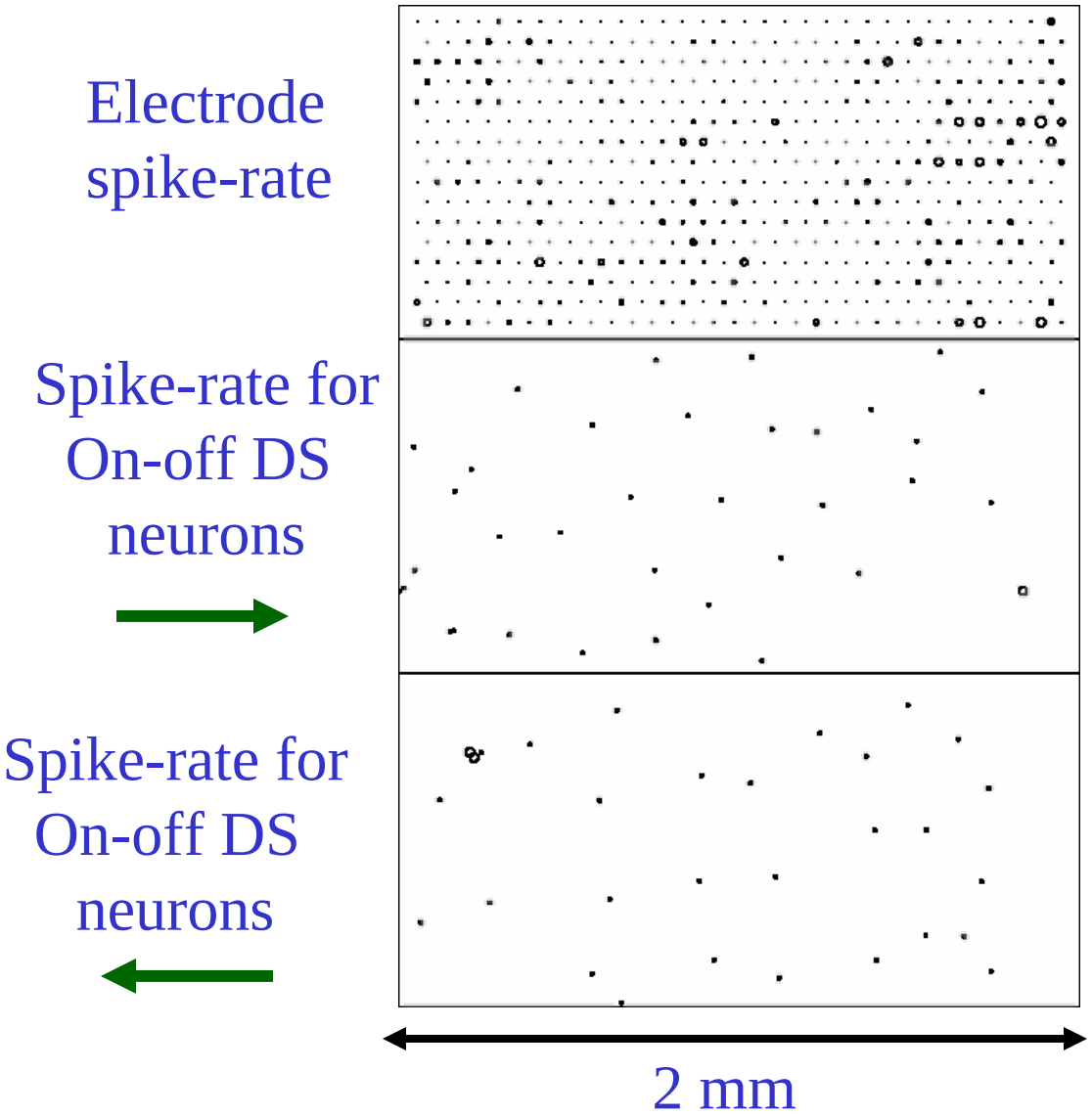


RF mosaics for clusters 1-4



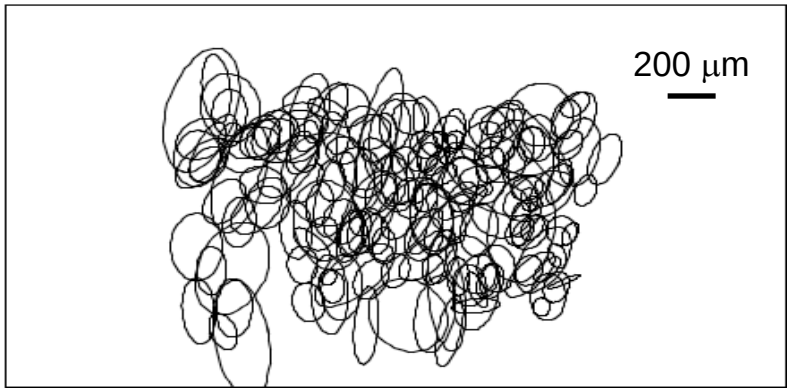
Neural activity recorded with 512-electrode system as image of vertical moving bar is focused on a section of guinea pig retina

(Animation repeats after 2 sweeps)

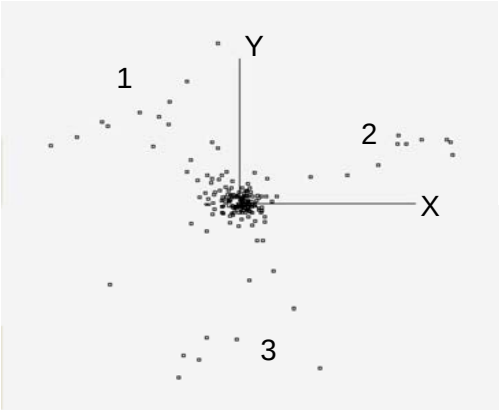


Guinea Pig Retinal Ganglion Cells: ON cells

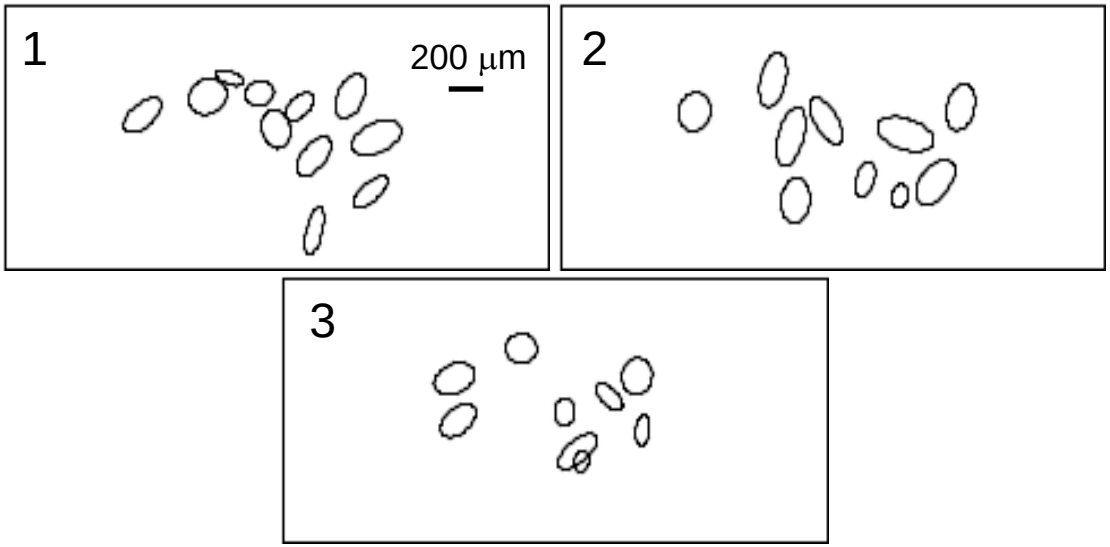
RF mosaic for 169 ON cells



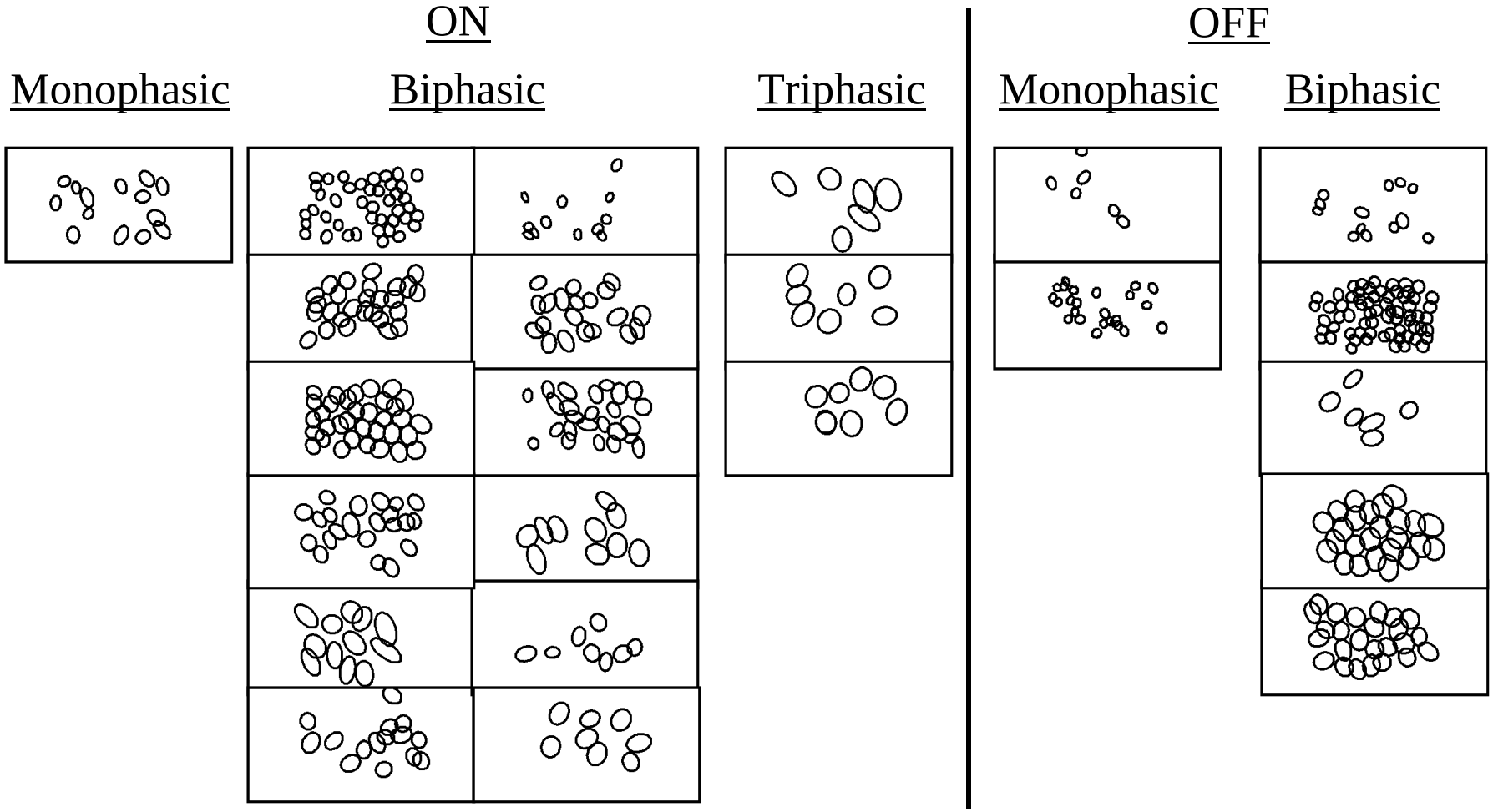
Direction selectivity for drifting sinusoidal gratings



RF mosaics for clusters 1-3



Guinea Pig Retinal Ganglion Cells: Non-Direction Selective



⇒ There are at least 30 distinct and highly specialized guinea pig retinal ganglion cell types

M. Grivich et al., Society for Neuroscience annual meeting, 2005

Summary and Additional Applications

- We have developed a multielectrode system for the large scale recording of retinal ganglion cell activity
- Experimental data has been obtained with live guinea pig and monkey retinas
- For the first time, it has become possible to study image processing and encoding by the retina in terms of the correlated activity of hundreds of neurons
- There are numerous physiological classes of retinal ganglion cells, each of which appears to tile the visual field, and each of which appears to send a separate image to the brain
- Additional applications include studies of:
  - ⇒ retinal development
  - ⇒ retinal stimulation for retinal prosthesis
  - ⇒ dynamical neural network activity in slices of brain tissue
  - ⇒ brain activity in awake, naturally-behaving animals