Lecture overview

• What hypothesis to test in the fly?
• Quantitative data collection
• Visual physiology conventions (“Methods”)

Course 9.17: Brain Laboratory, Brain and Cognitive Sciences
Lecture overview

• What hypothesis to test in the fly?
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General situation in vision encoding studies:

Independent variable: pattern of photons
Dependent variable: pattern of spikes from one or more neurons

A specific case (e.g. for H1 in the fly):

Independent variable: direction of a moving grating
Dependent variable: spike rate during each the presentation of each grating

spikes per unit time (spikes/sec)
Overall goal of the fly labs: the basics of carrying out a complete, quantitative neurophysiology experiment.

- Design visual stimuli to test a hypothesis
- Setup a prep to record from relevant neurons
- Collect digital data during that presentation
- Isolate individual spikes in that data
- Analyze the relationship between spike responses and visual stimuli
- Document your findings

MATLAB proj 2
Design lab
FLY wet lab 1
FLY wet lab 2
FLY wet lab 2
MATLAB proj 1
MATLAB proj 3
Data analysis lab
Lab Report 2

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Example of quantitative neurophysiology: orientation tuning a neuron in primary visual cortex (area V1) of the monkey.

One of our overarching goals is to teach you how these kind of plots are produced. You will do all the steps yourself!

Lecture overview

- What hypothesis to test in the fly?
- Quantitative data collection
- Visual physiology conventions ("Methods")
Basic electrophysiological setup

- Photon stimulation (retinal receptors)
- Shielding
- Preamplifier and filters
- Analog to digital device (A to D)
- Data collection computer
- Digital oscilloscope
- Audio monitor
- Observed signal
- Trigger pulse (start A to D)
- Synchronization pulses
- LCD screen
- Display computer

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We must have some way of saving the voltage signal so that we can find the spikes (action potentials) later.

A single action potential

Analog to digital device (A to D)

Number of samples taken every second:
- 1000 (1 kHz sampling)
- 2000 (2 kHz sampling)
- 4000 (4 kHz sampling)

What sampling rate is optimal?

Are there downsides to sampling faster?

*Sample at 2x the highest frequency in the signal to get a perfect reproduction of the signal (= “Nyquist sampling rate”).*
Basic electrophysiological setup

Photon stimulation (retinal receptors)

Preamplifier and filters

Digital oscilloscope

Audio monitor

Data collection computer

Synchronization pulses

Display computer

trigger pulse (start A to D)

S

Analog to digital device (A to D)

BNC

ground

LCD screen

shielding

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We must have some way of aligning (synchronizing) the visual stimulus with the recorded voltage.

Visual stimulus:

Voltage near neuron:

Times of action potentials (‘spikes’):
First steps to quantitative physiology (science):

- Ability to accurately repeat conditions.
- Control of variables -- only change one thing at a time!

**Stimulus run 1:**

**Stimulus run 2:**
Example voltage traces

Stimulus:

Voltage near neuron on run 1:

Voltage near neuron on run 2:
VoltageMatrixUV(1,:) = row 1 of matrix = Voltage data collected near neuron during run 1 of movie

VoltageMatrixUV(2,:) = row 2 of matrix = Voltage data collected near neuron during run 2 of movie

VoltageMatrixUV =

SpikeTimesMS{1} = [ 23 56 103 ... ]

SpikeTimesMS{2} = [ 10 12 150 160 175 ... ]

... 

SpikeTimesMS{10} = [ ... ]

TimesMS =

0 0.05 0.1 0.015 0.02 0.025 ... 10999.95 11000

time from movie start (ms) →
**Spike sorting:**

You learned how to do spike detection in Matlab Tutorial 1. However, that was relatively clean data. Your data file may be much messier and may contain many artifacts that you should try to deal with. For example, if the fly moves, this creates a lot of noise in the voltage trace, and a simple spike detector might count this noise as spikes. Thus, you should NOT blindly run a spike detector, but should examine your data closely and make sure the spike detector is doing something reasonable.
Another spike waveform

Your spike waveform of interest

Also, look out for noise bursts!
H1 cell    Post-stimulus time histogram (PSTH)

Neuronal spiking is variable:

"Post-stimulus spike histogram"

"Spike rasters"
What is a key analysis parameter here?

What are key underlying assumptions that allow us to collapse the 10 trials together?

"Post-stimulus spike histogram"

"Spike rasters"
If you compute a histogram for each of your conditions, you get this…

Effect of each condition on mean firing rate

mean over what?

Matlab project 3 due a week from Tuesday

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Example of quantitative neurophysiology: orientation tuning a neuron in primary visual cortex (area V1) of the monkey.

One of our overarching goals is to teach you how these kind of plots are produced. You will do all the steps yourself!

‘Direction-of-reach’ tuning curve for a neuron in primary motor cortex (area M1) of the monkey.

Figure 1.6 (A) Recordings from the primary motor cortex of a monkey performing an arm-reaching task. The hand of the monkey started from a central resting location, and reaching movements were made in the directions indicated by the arrows. The rasters for each direction show action potentials fired on five trials. (B) Average firing rate plotted as a function of the direction in which the monkey moved its arm. The curve is a fit using the function 1.15 with parameters $r_{\text{max}} = 54.69$ Hz, $r_0 = 32.34$ Hz, and $s_{\text{max}} = 161.25^\circ$. (A adapted from Georgopoulos et al., 1982, which is also the source of the data points in B.)

Can we make a tuning curve?

Inferotemporal Face Cell
Desimone et al. 1984

Tuning curves: what are they good for?

• Show the data (nice summary)
• Describe trends in the data (function fit)
• Predict response to other stimuli?
• Describe the neuron’s role in behavior???

Tuning curves: downsides

- Emphasize mean firing rates. Msec-scale timing of individual spikes is ignored
- Not everything we are interested in can be plotted along a single axis
Neuronal spiking is **stochastic**. Why?

Stimulus:

Voltage near neuron on run 1:

Voltage near neuron on run 2:
Lecture overview

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**Standard units of visual stimuli**

**Stimulus size: degrees of visual angle (deg)**

E.g. “The white square subtended approximately 4 deg x 4 deg of visual angle”

**Speed of motion: degrees of visual angle covered per unit time (deg/sec)**

E.g. “The white square was moved from left to right at a speed of 40 deg/s.”
Standard units of visual stimuli:
receptive field maps in vertebrates

- Center of receptive field
- Size (diameter) of receptive field
- Center of gaze (center of the retina)

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Standard units of visual stimuli: receptive field map

Receptive field map of a neuron

Center of visual axis of the fly

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motion RF of the H1 neuron

![Graph showing motion RF of the H1 neuron with azimuth and elevation axes, and a shaded region indicating preferred motion direction.]

Your monitor?


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Krapp and Hengstenberg, Vision Res. (1997)
Fly visual field: state your conventions!

Video monitor

'above'

'behind'

'in front'

'below'

Directly in front of fly when in real world

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Luminance
Basic units of visual stimuli

- **luminance**: amount of light (~photons) emitted from a particular area per unit time (units: candela/m²)

- **contrast**: range of luminance relative to the total luminance (e.g. Michelson contrast = \( \frac{L_{\text{max}} - L_{\text{min}}}{L_{\text{max}} + L_{\text{min}}} \))

For your lab report:

assume that your display monitor has:

maximum luminance of 100 cd/m² (full white, pixel value = 255 in Matlab matrix)

minimum luminance of 1 cd/m² (deepest black, pixel value = 0 in Matlab matrix)
9.17 Systems Neuroscience Lab
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