

Tools for comparative connectomics: case studies from two sides of a larval Drosophila brain

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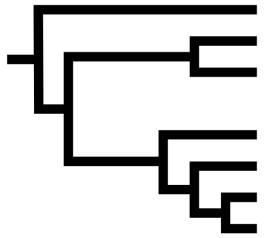
 [@bpedigod \(Twitter\)](https://twitter.com/bpedigod)

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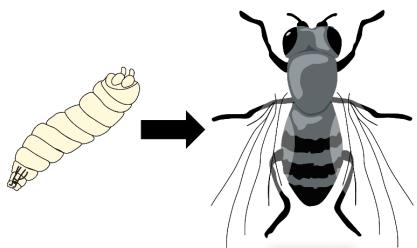
These slides at:

Connectomics is useful...

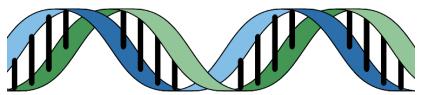
Many of the stated goals of connectomics rely on linking connectome to other domains...



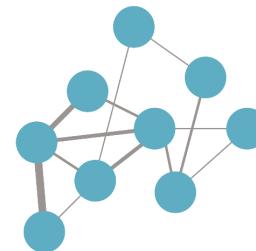
Evolution



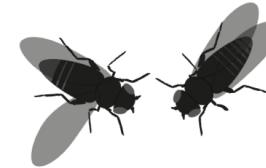
Development



Genome



Connectome



Behavior



Activity

Connectome ↔ memory

...the acquisition of wiring diagrams across multiple individuals will yield insights into how experiences shape neural connections.

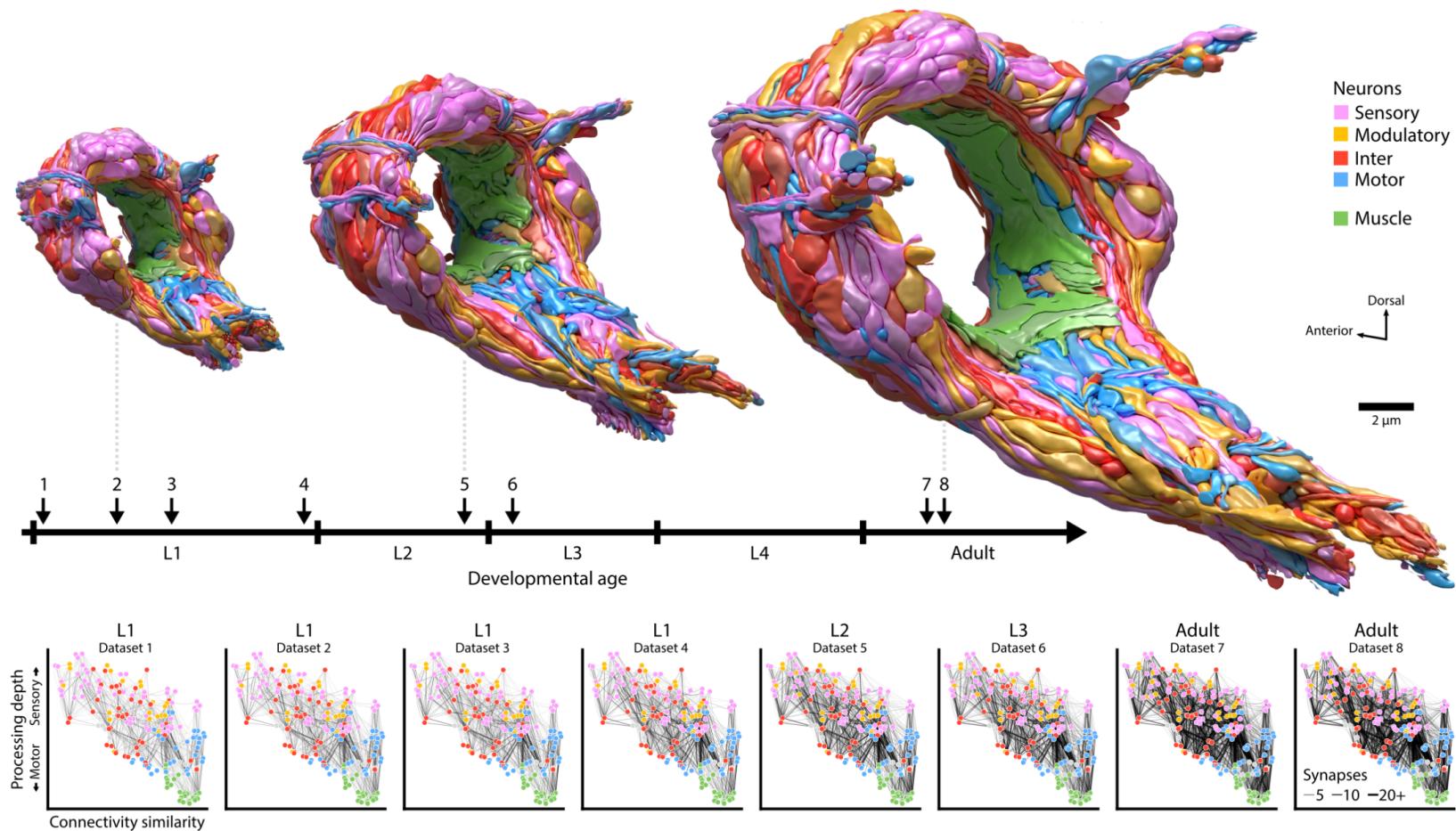
Emphasis added

Connectome ↔ evolution

Comparative connectomics of [...] species across the phylogenetic tree can infer the archetypal neural architecture of each bauplan and identify any circuits that possibly converged onto a shared and potentially optimal, structure.

Emphasis added

Connectomes across development



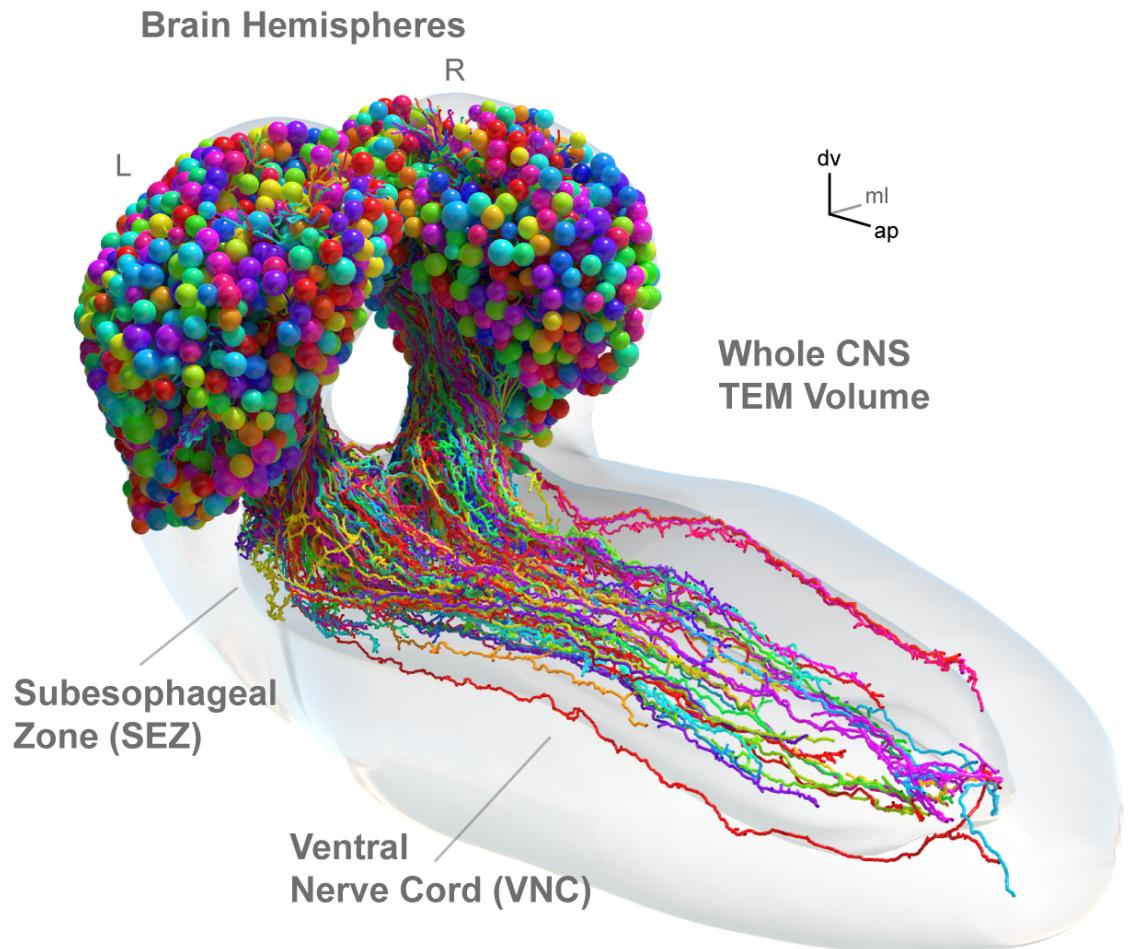
But it is methodologically hard to compare connectomes!

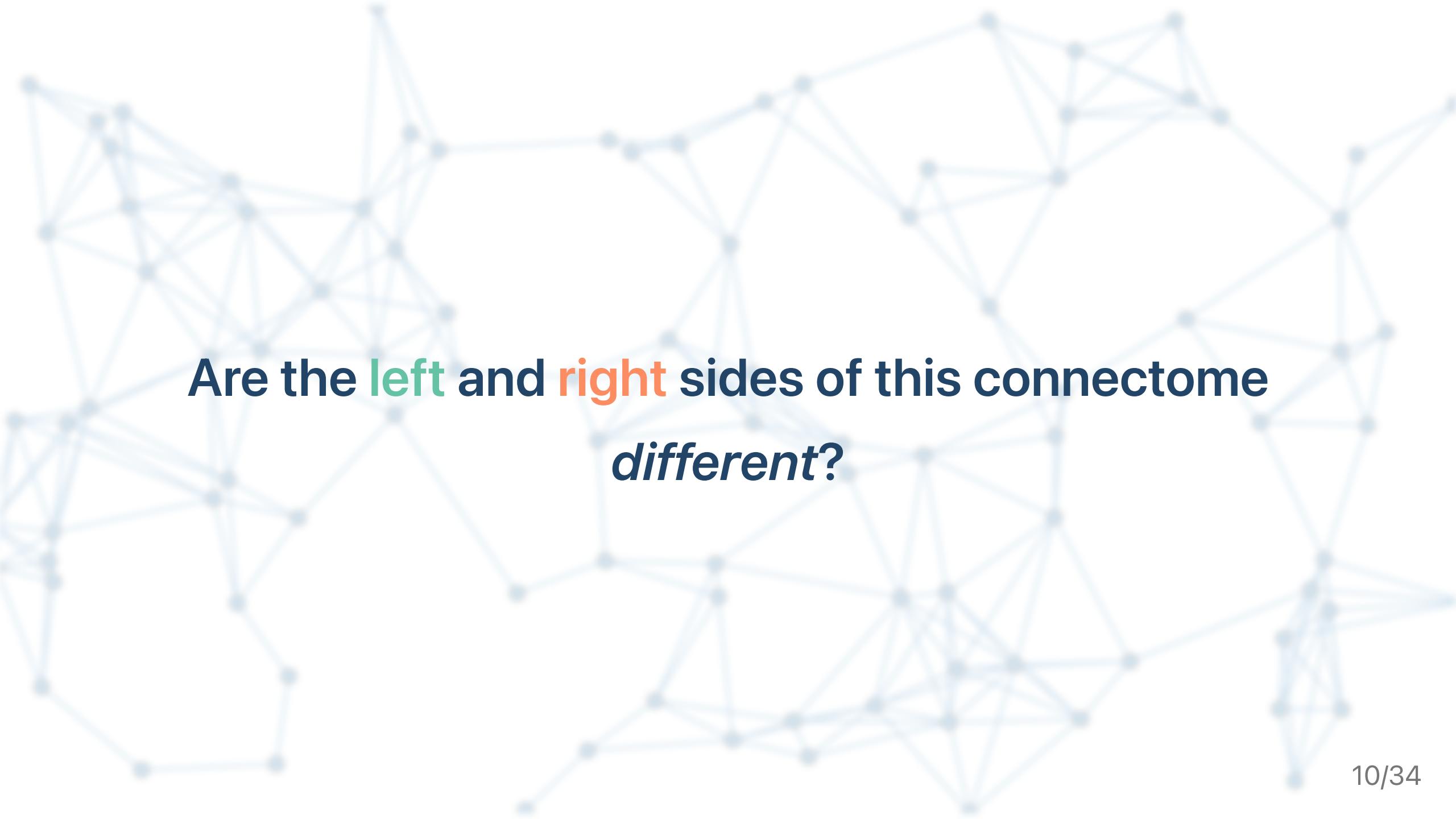
Outline for today

- Describe a dataset that I'll use for these examples throughout
- Show how connectome comparison can be framed as network hypothesis testing
- Show how we can use automated tools for predicting the correspondence of neurons across datasets
- Mention some extensions to use/combine/extend these tools that we're working on

Drosophila larva (AKA a maggot) brain connectome

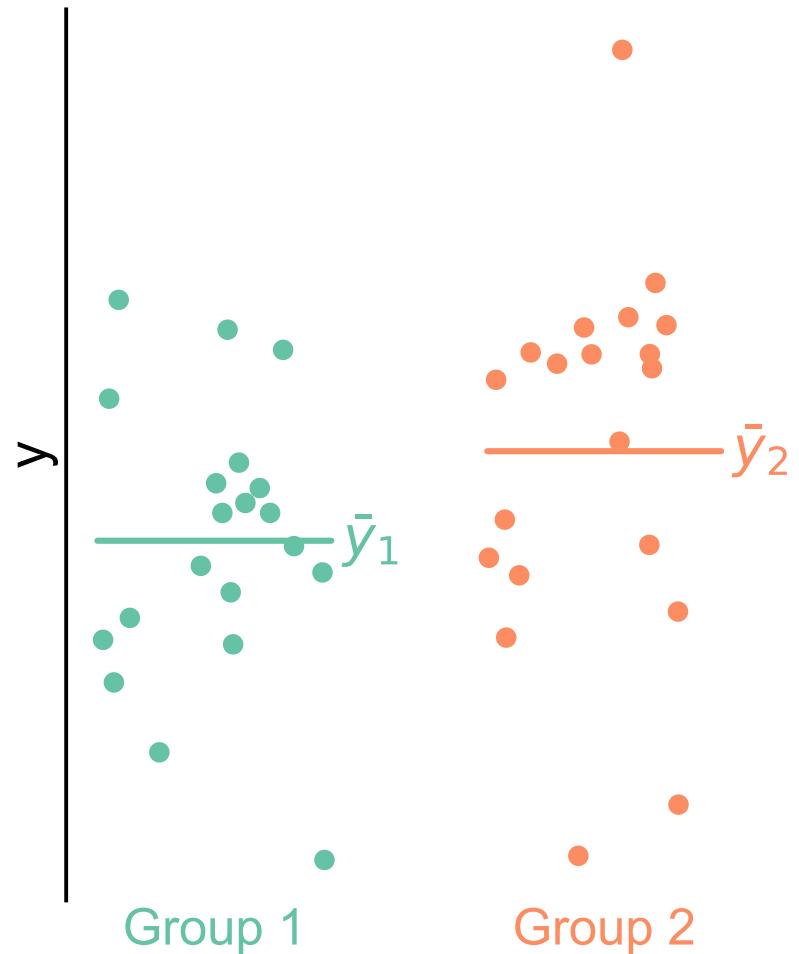
- Collaboration with Marta Zlatic/Albert Cardona's groups - led by Michael Winding
- First whole-brain, single-cell connectome of any insect
- ~3000 neurons, ~550K synapses
- Both hemispheres of the brain reconstructed





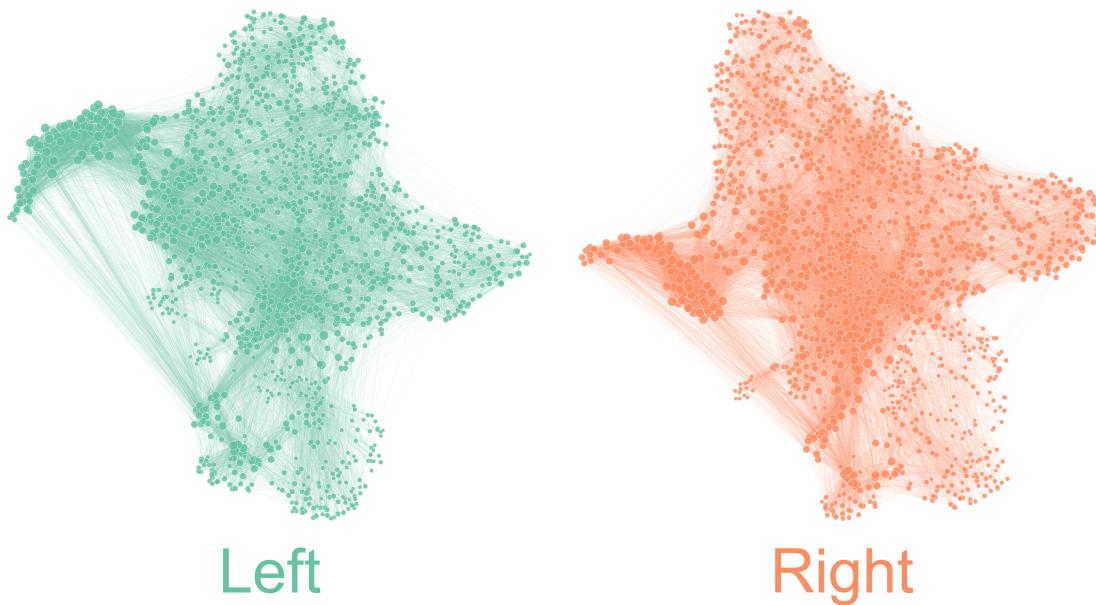
Are the **left** and **right** sides of this connectome
different?

Are these populations different?



- Known as two-sample testing
- $Y^{(1)} \sim F^{(1)}$, $Y^{(2)} \sim F^{(2)}$
- $H_0 : F^{(1)} = F^{(2)}$
 $H_A : F^{(1)} \neq F^{(2)}$

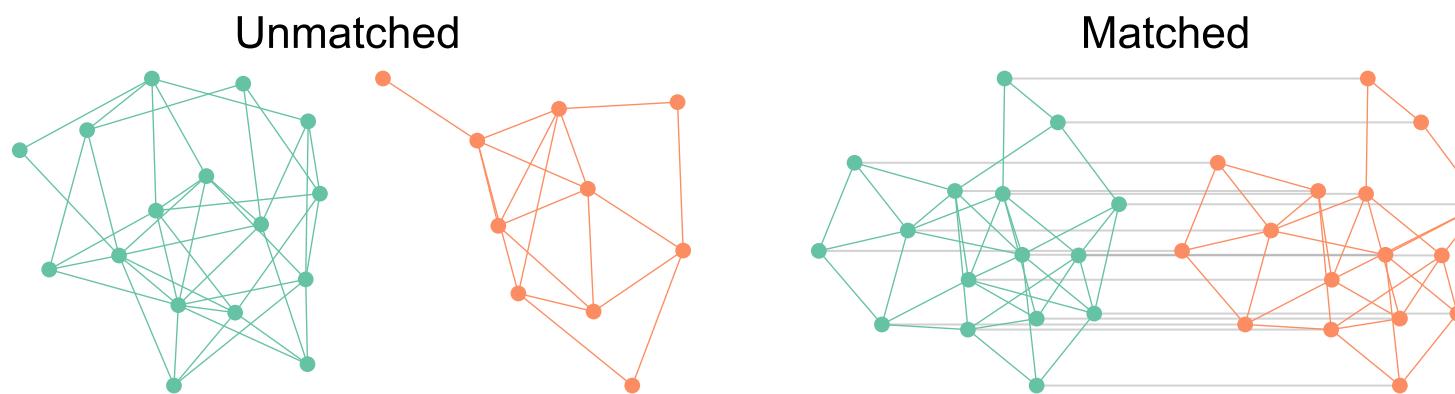
Are these two *networks* different?



- Want a two-network-sample test!
- $A^{(L)} \sim F^{(L)}$, $A^{(R)} \sim F^{(R)}$
- $H_0 : F^{(L)} = F^{(R)}$
 $H_A : F^{(L)} \neq F^{(R)}$

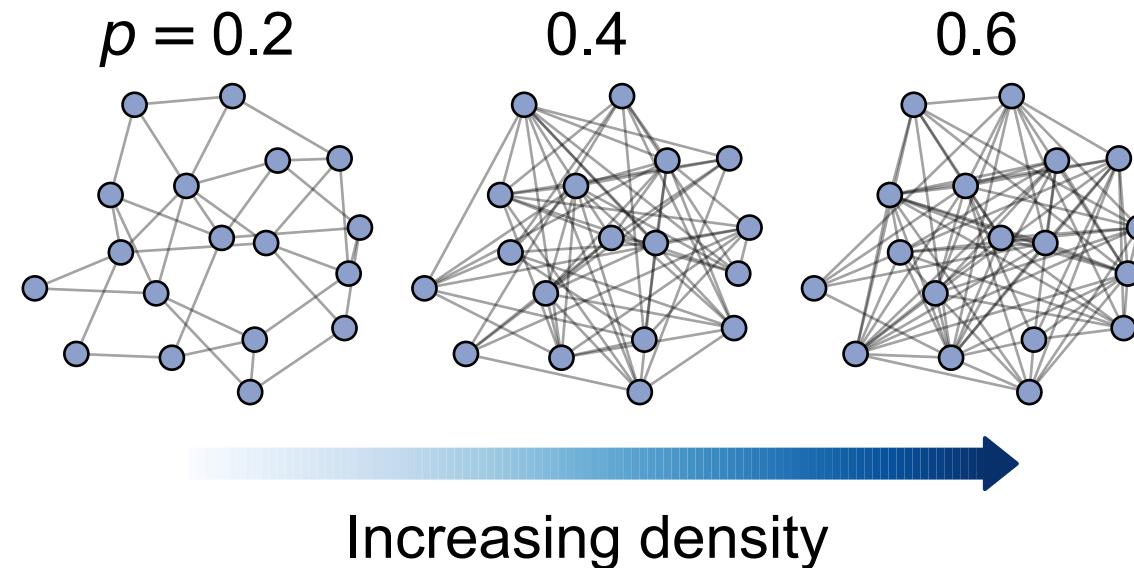
Assumptions

- We know the direction of synapses, so network is *directed*.
- For simplicity (for now), consider networks to be *unweighted*.
- For simplicity (for now), consider the **left → left** and **right → right** (*ipsilateral*) connections only.
- Not going to assume any nodes are matched



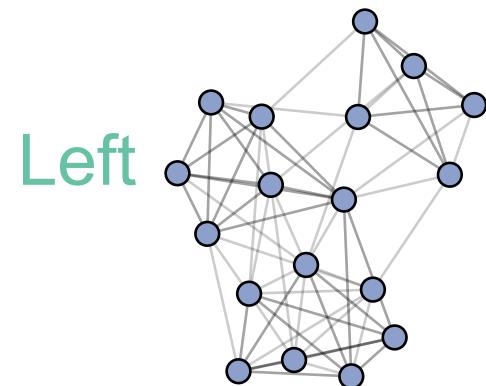
Erdos-Renyi model

- All edges are independent
- All edges generated with the same probability, p



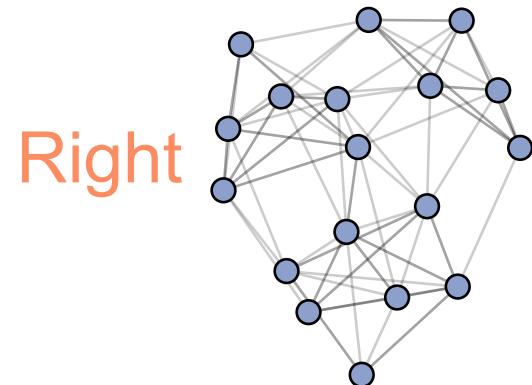
Density-based testing

Compute global
connection density



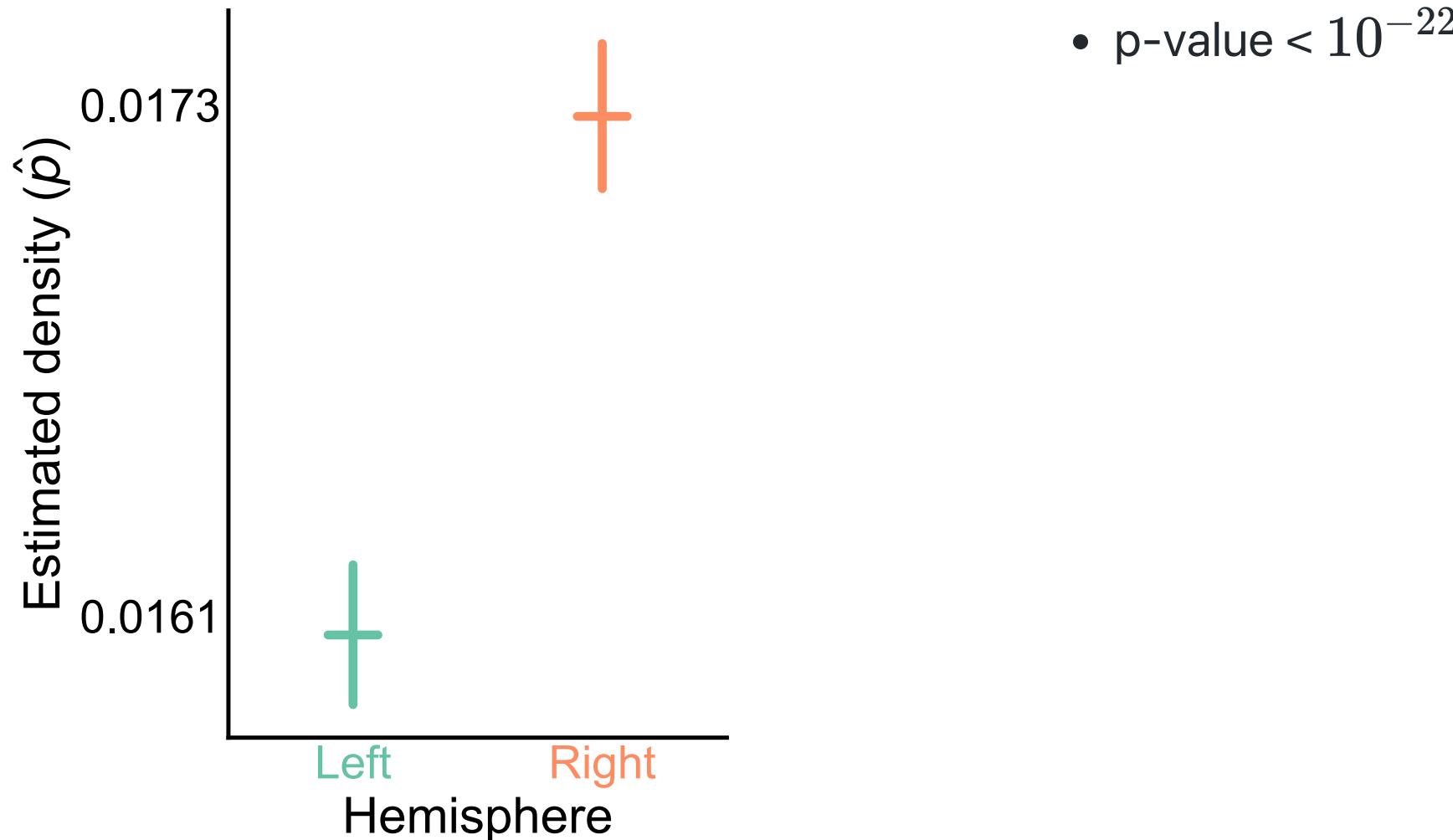
Compare ER
models

$$p = \frac{\# \text{ edges}}{\# \text{ potential edges}}$$



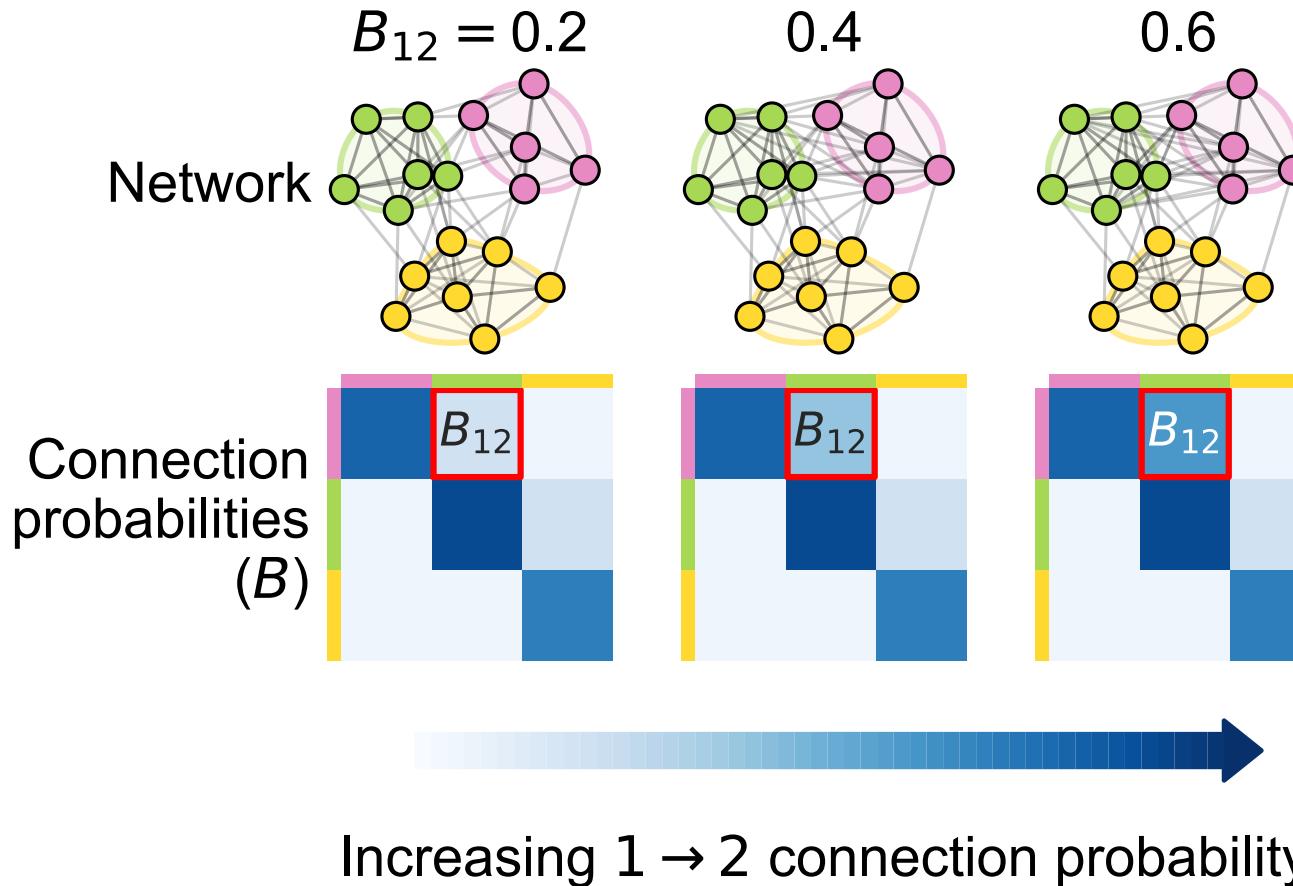
$$\begin{aligned} H_0: p^{(L)} &= p^{(R)} \\ H_A: p^{(L)} &\neq p^{(R)} \end{aligned}$$

We detect a difference in density

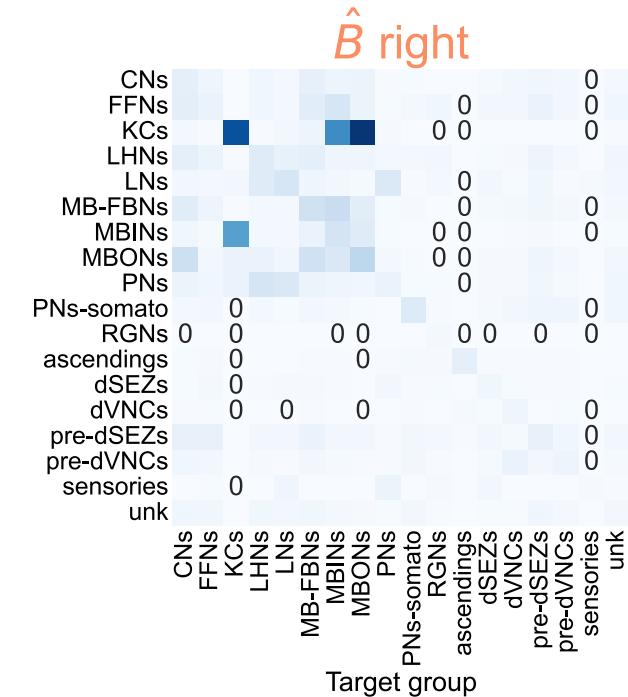
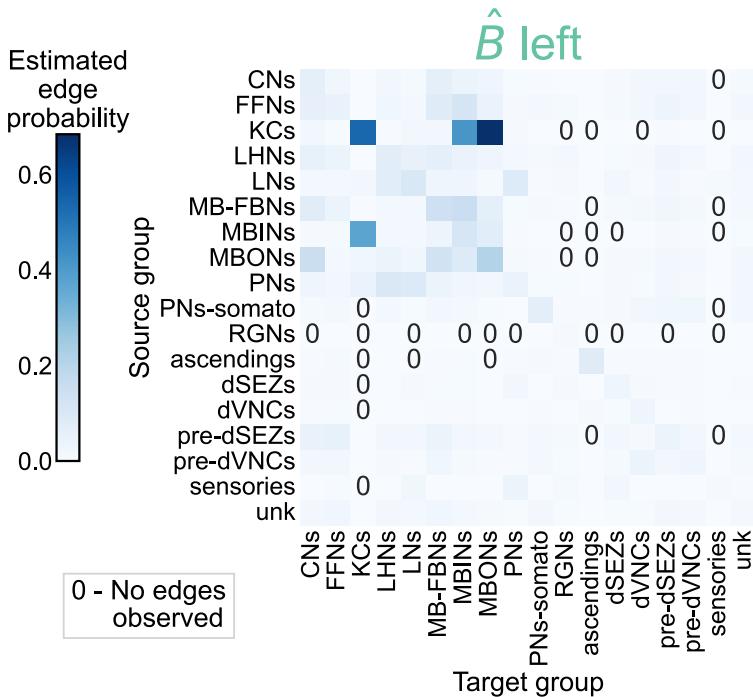
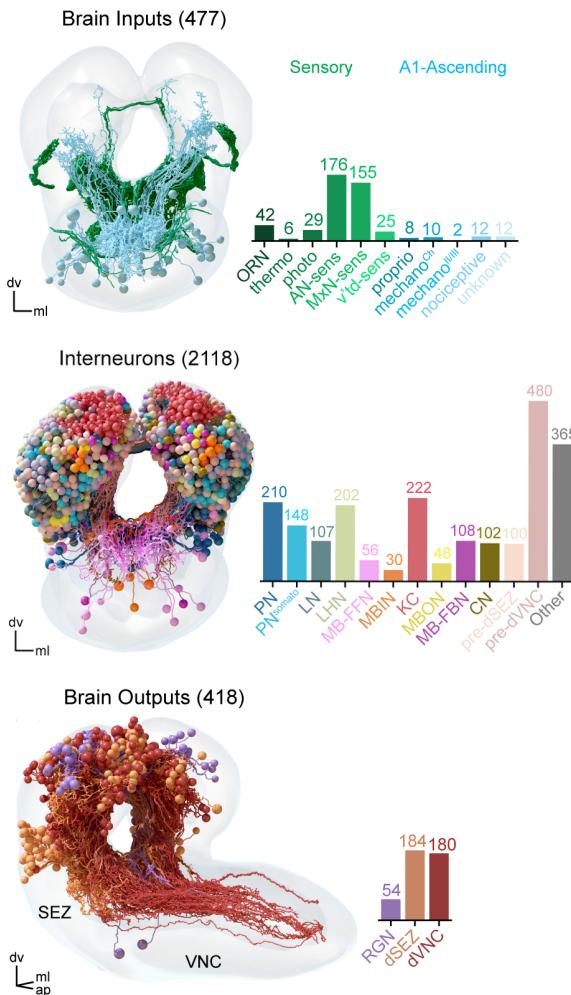


Stochastic block model

- Edge probabilities are a function of a neuron's group



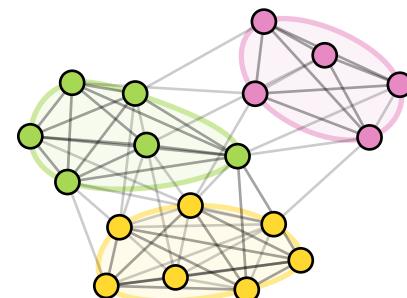
Connection probabilities between groups



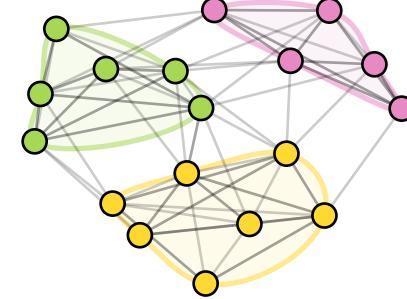
Group-based testing

Group neurons

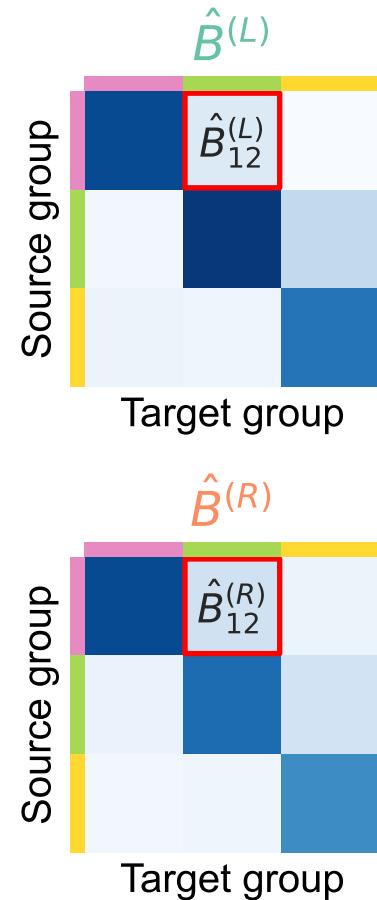
Left



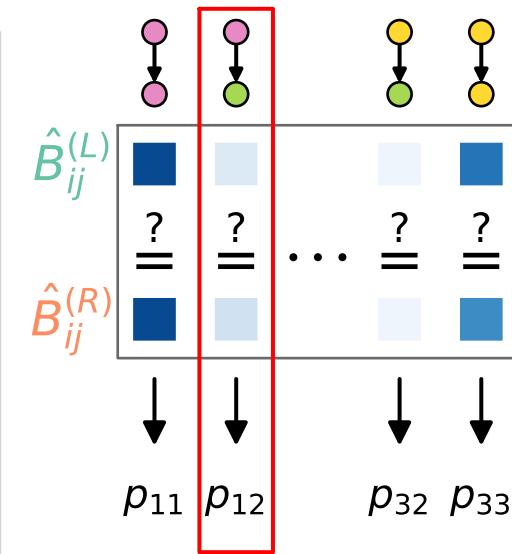
Right



Estimate group connection probabilities

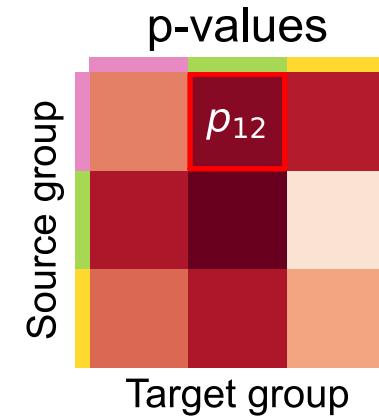


Compare probabilities, compute p-values



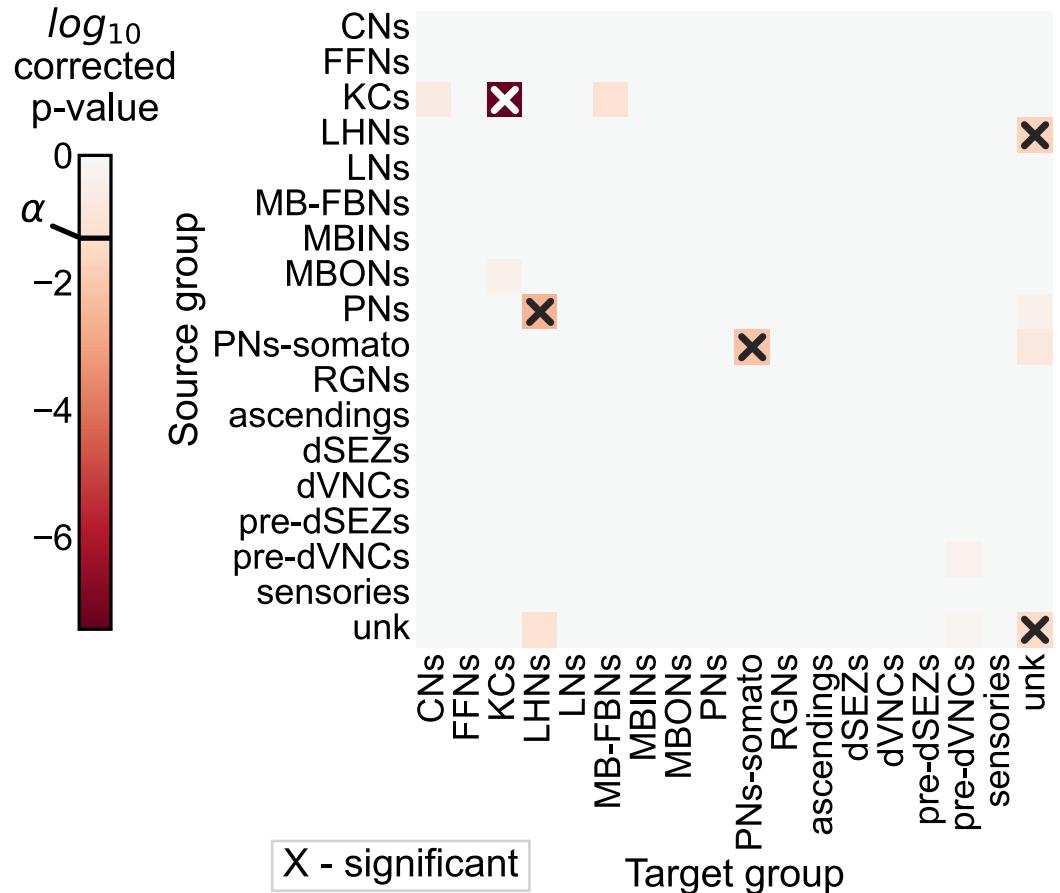
$$H_0: \hat{B}_{ij}^{(L)} = \hat{B}_{ij}^{(R)}$$
$$H_A: \hat{B}_{ij}^{(L)} \neq \hat{B}_{ij}^{(R)}$$

Combine p-values for overall test



$$H_0: \hat{B}^{(L)} = \hat{B}^{(R)}$$
$$H_A: \hat{B}^{(L)} \neq \hat{B}^{(R)}$$

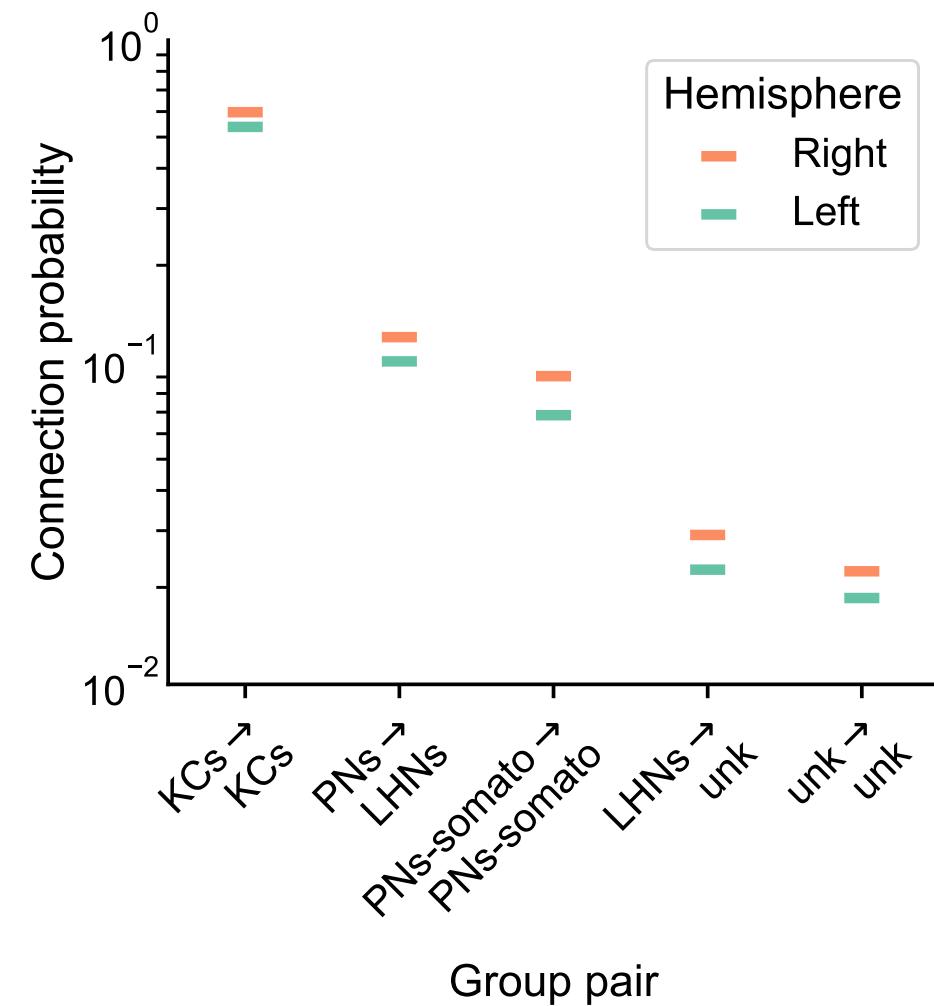
We detect a difference in group-to-group connection probabilities



- After multiple comparison, find 5 group-to-group connections which are significantly different
- Combine (uncorrected) p-values (like a meta-analysis), leads to p-value for overall test of $< 10^{-7}$

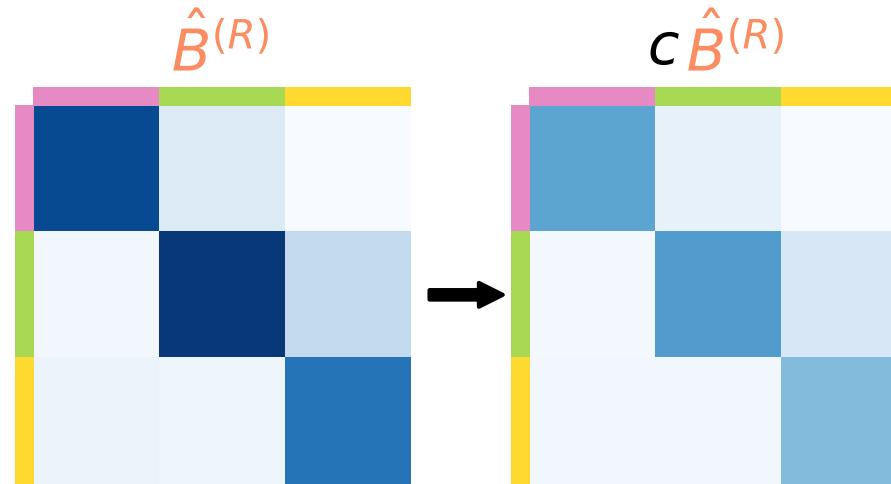
Should we be surprised?

- Already saw that even the overall densities were different
- For all significant comparisons, probabilities on the right hemisphere were higher
- Maybe the right is just a "scaled up" version of the left?
 - $H_0 : B^{(L)} = cB^{(R)}$ where c is a density-adjusting constant, $\frac{p^{(L)}}{p^{(R)}}$



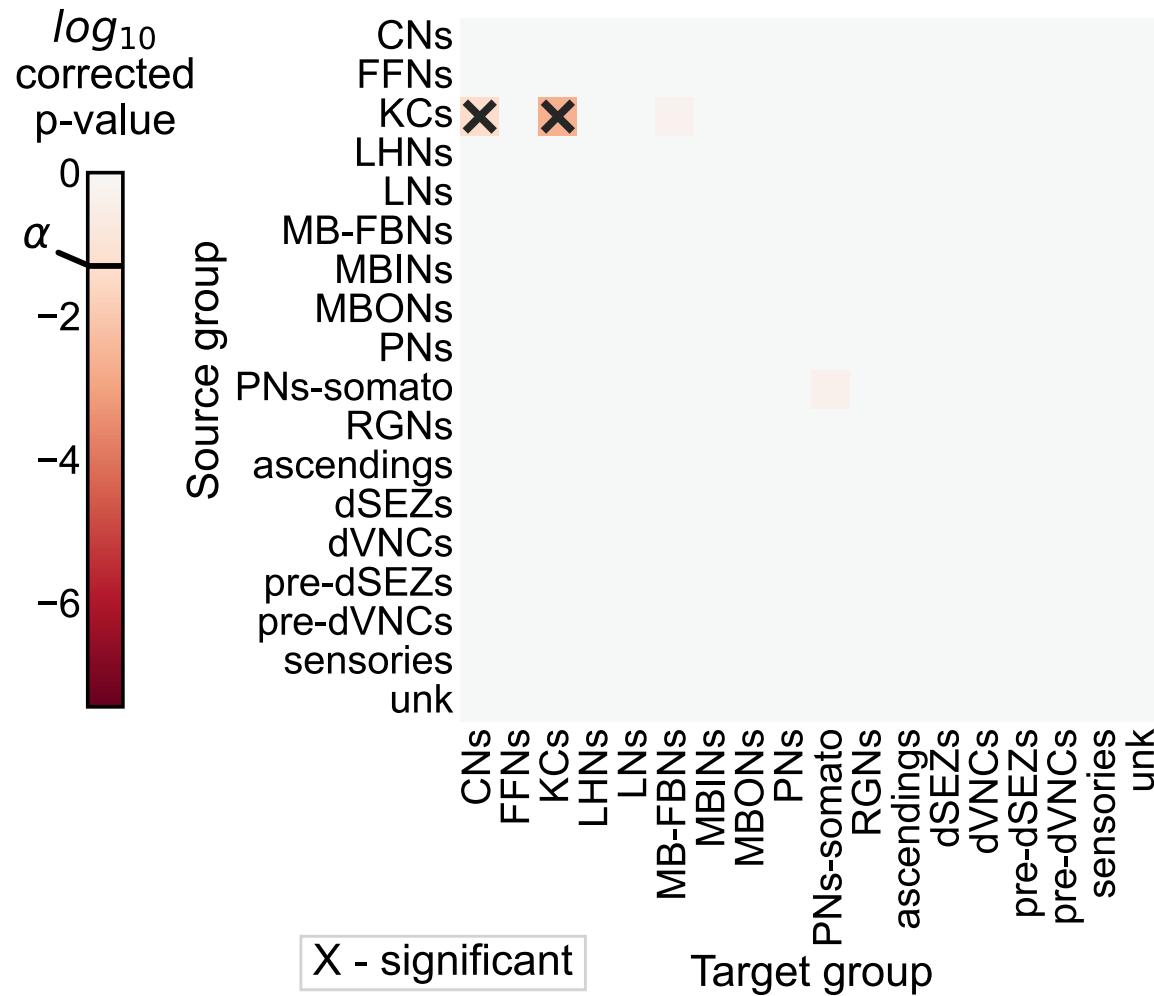
Adjusting for a difference in density

Scale connection probabilities
to match densities



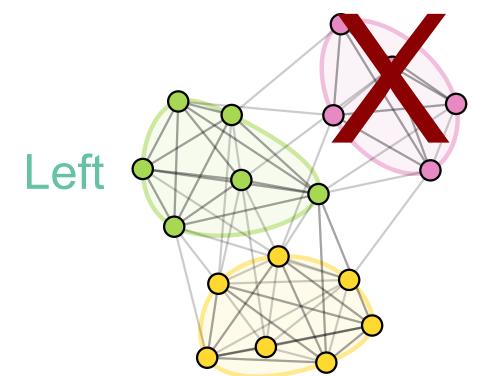
$$\begin{aligned} H_0: B^{(L)} &= cB^{(R)} \\ H_A: B^{(L)} &\neq cB^{(R)} \end{aligned}$$

Remaining differences are isolated to KCs

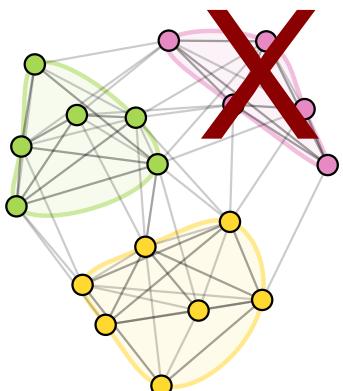


So the Kenyon cells (KCs) are the only group where we detect remaining differences...

Remove Kenyon cells



Right



Re-run all tests

Density

$$\begin{aligned} H_0: p^{(L)} &= p^{(R)} \\ H_A: p^{(L)} &\neq p^{(R)} \end{aligned}$$

Group connection

$$\begin{aligned} H_0: B^{(L)} &= B^{(R)} \\ H_A: B^{(L)} &\neq B^{(R)} \end{aligned}$$

Density-adjusted
group connection

$$\begin{aligned} H_0: B^{(L)} &= cB^{(R)} \\ H_A: B^{(L)} &\neq cB^{(R)} \end{aligned}$$

- ER test: $p < 10^{-26}$
- SBM test: $p \approx 0.003$
- Adjusted SBM test: $p \approx 0.43$

To sum up...

Model	H_0 (vs. $H_A \neq$)	KCs	p-value	Interpretation
ER	$p^{(L)} = p^{(R)}$	+	$< 10^{-23}$	Reject densities the same
SBM	$B^{(L)} = B^{(R)}$	+	$< 10^{-7}$	Reject group connection probabilities the same
aSBM	$B^{(L)} = cB^{(R)}$	+	≈ 0.002	Reject above even after accounting for density
ER	$p^{(L)} = p^{(R)}$	-	$< 10^{-26}$	Reject densities the same (w/o KCs)
SBM	$B^{(L)} = B^{(R)}$	-	≈ 0.003	Reject group connection probabilities the same (w/o KCs)
aSBM	$B^{(L)} = cB^{(R)}$	-	≈ 0.43	Don't reject above after density adjustment (w/o KCs)

Matching neurons

Extensions and ongoing work

matched versions of our tests

- we think greater power, basically

testing for a significant matching

- evaluate stereotypy at a single neuron level, basically

the value of pairs - looking at models

- show the hierarchical clustering
- SBM cross validation curve

Summary

How to use these tools?

graspologic

bilateral repo

bgm

get in touch!

Acknowledgements

Team



Michael
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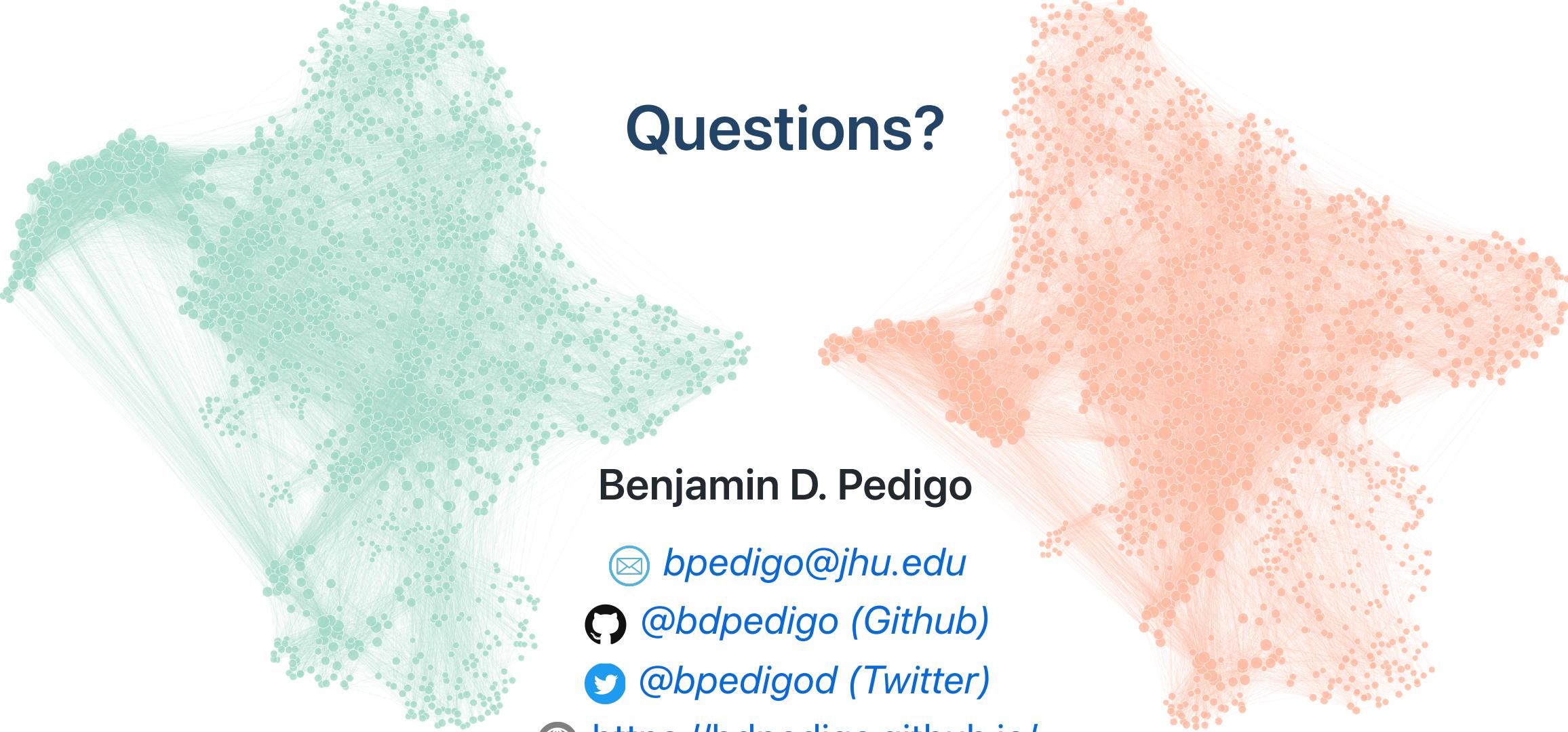
Marta
Zlatic

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Cardona

Carey
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Joshua
Vogelstein

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Questions?

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Left

Right