Welcome to the March 2025 Virtual Data Lab scRNA-Seq Training Workshop!

March 24-28, 2025 Childhood Cancer Data Lab https://alexslemonade.github.io/2025-march-training/







Tell us about you!

- What's your name?
- What are your pronouns?
- What are you studying?
- What's a recent piece of media (book, movie, TV show, etc.)
 you enjoyed?

Meet your instructors



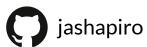
JOSH Joshua Shapiro

Senior Data Scientist @ the Data Lab

PhD Ecology & Evolution, *UChicago*Postdoc Integrative Genomics, *Princeton*

Research interests:

- Evolutionary genomics
- Single cell workflows



Meet your instructors



STEPHANIE Stephanie Spielman

Data Scientist @ the Data Lab

PhD Integrative Biology *UT Austin*Postdoc Computational Molecular Evolution *Temple*

Research interests:

- Protein evolution and reproducible genomics
- Data science and bioinformatics education



Meet your instructors



Ally Hawkins

Data Scientist @ the Data Lab

PhD Cancer Biology University of Michigan Postdoc Computational Biology Cornell

Research interests:

- Single cell data analysis
- Origins of pediatric solid tumors



Other staff you'll meet this week!



JEN Jen O'Malley

Scientific Community Manager

- Helps administer Data Lab programs such as workshops
- Engages with the research community

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Code of Conduct

Be kind, have fun

We value the involvement of everyone in the community. We are committed to creating a friendly and respectful place for learning, teaching, and contributing.

- Use welcoming and inclusive language
- Be respectful of different viewpoints and experiences
- Gracefully accept constructive criticism
- Focus on what is best for the community
- Show courtesy and respect towards other community members

Read the full Code of Conduct here:

https://alexslemonade.github.io/2025-march-training/code-of-conduct.html

If you at any time feel harassed or treated inappropriately, please contact ccdl@alexslemonade.org.

What you will learn (and what you won't)

What you will learn

We will introduce you to the R programming language, R Notebooks, and some reproducible research practices.

We cover pipelines for the quality control, pre-processing, and initial analysis of single-cell RNA-seq data almost entirely through hands-on exercises.

We generally elect to go broad and not deep.

Our overarching goals: To prepare you to perform "frontline" analyses of your own data, to get you more comfortable reading documentation/learning new methods on your own, and to give you tools to collaborate more effectively with analysts when needed

What you won't learn

We don't address experimental design (e.g., how many replicates you need).

We won't compare tools (e.g., Bioconductor vs. Seurat for single cell analysis).

We won't cover every feature (or assumption) of the tools we do present.

You may not be able to perform every analysis you need for your own work, particularly for complex experimental designs. For example, in the context of single-cell analysis, we do not cover integrating data from multiple samples.

We present analysis as a series of *linear steps*. In practice, it's **not**. It's important to consult analysis experts when you need to and to keep track of and report what you've done.

How do we pick what we teach?

We want methods to be or to have:

- Useful for a wide range of experimental designs, sample sizes
- Easy to use, well-documented, and consistently updated
- Solid tutorials, a sizeable user base, and responsive authors/maintainers

We have a preference for methods that integrate easily into a single workflow that can be run on a laptop (and our own personal biases as scientists).

Schedule

| Monday | Wednesday | Friday |
|---|--|-----------------------------|
| Workshop Intro | Single-cell RNA-seq Dimensionality reduction | Consultations Your own data |
| Intro to R RStudio Server Base R | Clustering Marker identification | Exercise notebooks |
| ggplot2 & tidyverse | Consultations | |
| Consultations Exercise notebooks | Exercise notebooks Your own data | <u>Presentations</u> |
| Tuesday | Thursday | |
| Single-cell RNA-seq Quantification & QC Filtering & Normalization | Single-cell RNA-seq Cell-type annotation Working with CITE-Seq data | |
| Consultations Exercise notebooks Your own data | Consultations Exercise notebooks Your own data | |

 $\textbf{Full schedule:} \ \underline{\texttt{https://alexslemonade.github.io/2025-march-training/workshop/SCHEDULE.html}$

Daily Schedule Outline

Instruction

Full group Lectures

Breakout

Small groups
Start exercise notebooks

Consultation Period

Exercise notebooks Your own data

- Introduce concepts and background
- Demonstrate usage
- Answer general questions
- Split up into Zoom breakout rooms
- Ask questions of instructors and other participants

- Practice what you have learned
- Work on exercises individually or in groups
- Work with your own data

Module Layout

- 00a-rstudio_guide.Rmd
- 00b-debugging_resources.Rmd
- 00c-good_scientific_coding_practices.Rmd
- 01-intro_to_base_R-live.Rmd
- 02-intro_to_ggplot2-live.Rmd
- 03-intro_to_tidyverse-live.Rmd
- exercise_01-intro_to_base_R.Rmd
- exercise_02-intro_to_R.Rmd
- exercise_03a-intro_to_tidyverse.Rmd
- exercise-03b-intro_to_tidyverse.Rmd

■ These are reference documents. Go through these on your own.

These are **Instruction** notebooks.

We'll walk through these together, step-by-step, during the workshop.

These are **Exercise** notebooks.

Use these to practice what you've learned. We're here to help!

Module cheatsheets cover key functions

https://github.com/AlexsLemonade/training-modules/tree/2025-march/module-cheatsheets

dplyr

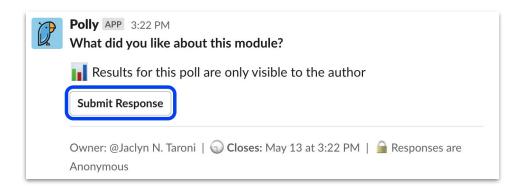
Read the dplyr package documentation here.

A vignette on the usage of the dplyr package can be found here.

What it's Library/Package Piece of code What it does called Pipe dplyr 8>8 Funnels a data.frame through tidyverse operations operator Returns a subset of rows matching the conditions of filter() Filter dplyr the specified logical argument Reorders rows in ascending order. arrange(desc()) dplyr arrange() Arrange would reorder rows in descending order. dplyr select() Select Selects columns that match the specified argument Adds a new column that is a function of existing dplyr mutate() Mutate columns Summarises multiple values in an object into a single value. This function can be used with other dplyr summarise() Summarise functions to retrieve a single output value for the grouped values. summarize and summarise are synonyms in this package. Renames designated columns while keeping all dplyr rename() Rename variables of the data.frame Groups data into rows that contain the same dplyr group by() Group By specified value(s) Joins data from two data frames, retaining only the dplyr inner join() Inner Join rows that are in both datasets.

We want your feedback!

At the end of each module, we will post a few questions in the Slack channel.



- The most difficult or confusing point of the module ("muddiest point")
 We will post additional material answering your questions the next day
 Responses to this question will appear in the channel anonymously
- What did you like about the module?
- How we can improve the module?
 These responses will be collected anonymously (and not posted)

Friday

Your own projects Exercise notebooks

Spend Friday working with your own data, getting assistance as needed from Data Lab staff and each other.

Presentations

Present what you worked on during the consultation times to the group!

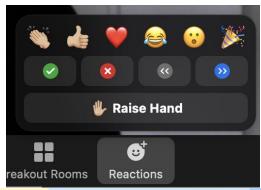
Virtual Training Procedures



General Zoom Etiquette

- Keep your microphone muted
- Type questions in the Chat window (directed to the host)

- To: Jaclyn Taroni ▼ (Privately)
 Type message here...
- Click on the "Chat" button at the bottom of your window to open the chat.
- Use the Zoom reactions to tell us how you are doing!

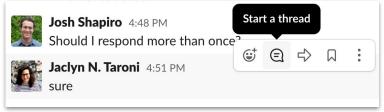


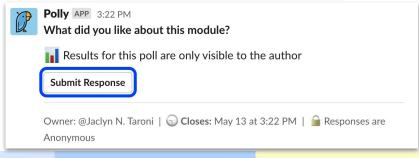


Using Slack

- Use the #2025-march-training Slack channel
- Post public questions, get help with errors and debugging, make comments, and help your fellow participants!
 - Use threads to keep related content together

Watch out for feedback polls!





Communication during instruction



- I have an urgent question that needs an answer before moving on:
 - Raise Hand or Chat with the room host
- I'm stuck with an error and can't proceed with the hands-on exercise
 - *Chat* with meeting host: Request 1:1 and you will be placed in a breakout room with a Data Lab staff member



- I have an general question that does not need an answer right away.
 - **Post** in #2025-march-training
- I'm having trouble logging in to RStudio Server
 Direct Message a Data Lab staff member (not the current host or instructor)

Trouble logging into Zoom and Slack? *Email* training@ccdatalab.org

Communication at other times (consultation periods)



- I have questions about previous instruction or exercise notebooks
 - **Post** in #2025-march-training
 - If you need to share your screen, we will set up a 1:1 or group Zoom call
- I would like to be paired up with other participants
 - **Post** in #2025-march-training; we can set you up in a Zoom breakout room

- I have a question that is highly specific to my data
 - **Direct Message** a Data Lab staff member
- I'm having trouble logging in to RStudio Server
 - **Direct Message** a Data Lab staff member

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