



Welcome to the November 2021 Virtual CCDL scRNA-Seq Training Workshop!

Nov 1 – Nov 5, 2021
Childhood Cancer Data Lab

<https://alexslemonade.github.io/2021-november-training>



Tell us about you!

- What's your name?
- What are you studying?
- What was your favorite Halloween costume that you saw this year?



Meet your instructors



JOSH

Joshua Shapiro

Data Scientist @ the CCDL

PhD Ecology & Evolution, *UChicago*

Postdoc Integrative Genomics, *Princeton*

Research interests:

- **Evolutionary genomics**
- **Single cell workflows**



jashapiro

Meet your instructors



CHANTE

Chante Bethell

Biological Data Analyst @ the CCDL

Bachelor's in Bioinformatics from *Rowan University*

Research interests:

- **Functional motifs in the proteome**



cbethell

Meet your instructors



Ally

Ally Hawkins

Data Scientist @ the CCDL

PhD Cancer Biology *University of Michigan*

Postdoc Computational Biology *Cornell*

Research interests:

- **Single cell data analysis**
- **Origins of pediatric solid tumors**



allyhawkins

Meet your instructors



JACLYN

Jaclyn Taroni

Director @ the CCDL

PhD Genetics *Dartmouth*

Postdoc Computational Biology *UPenn*

Research interests:

- **Transcriptomics in rare, complex diseases**
- **Unsupervised pattern extraction**



jaclyn-taroni

Other staff you may see

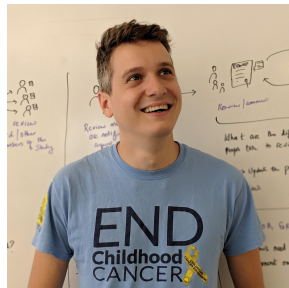


JEN

Jen O'Malley

**Scientific Community
Manager**
@CCDL

- Maintains a supportive environment for pediatric cancer researchers
- Manages Data Lab communications



KURT

Kurt Wheeler

Data Engineer
@CCDL

- Builds scalable systems
- Manages servers



DAVID

David Mejia

Developer
@CCDL

- Make websites
- Debugs

Tell us about you!

- What's your name?
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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.

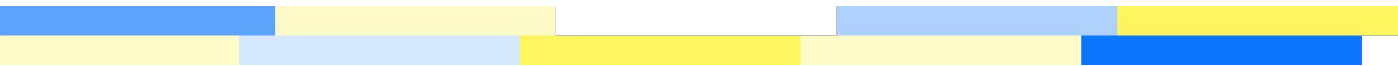
- Use welcoming and inclusive language
- Be respectful of different viewpoints and experiences
- Gracefully accept constructive criticism
- Focus on what is best for the community
- Show courtesy and respect towards other community members

Read the full Code of Conduct here:

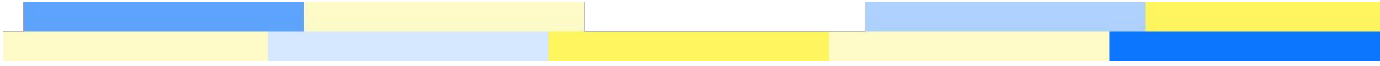
<https://alexslemonade.github.io/2021-november-training/code-of-conduct.html>



If you at any time feel harassed or treated inappropriately, please contact
ccd1@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.



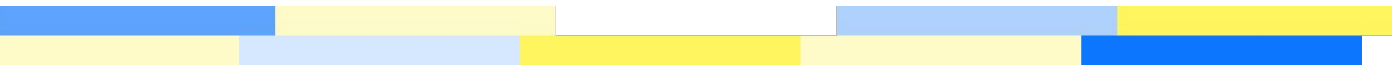
How do we pick what we teach?

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

Workshop Intro

Intro to R

RStudio Server
Base R
ggplot2 & tidyverse

Consultations

Exercise notebooks

Wednesday

Single-cell RNA-seq

Dimensionality reduction
Clustering
Marker identification

Consultations

Exercise notebooks
Your own data

Friday

Consultations

Your own data
Exercise notebooks

Presentations

Tuesday

Single-cell RNA-seq

Quantification & QC
Filtering & Normalization

Consultations

Exercise notebooks
Your own data

Thursday

Pathway Analysis

Overrepresentation
GSEA

Consultations

Exercise notebooks
Your own data

Full schedule: <https://alexslemonade.github.io/2021-november-training/workshop/SCHEDULE.html>

Daily Schedule Outline

Instruction

Full group

Lectures

- Introduce concepts and background
- Demonstrate usage
- Answer general questions

Breakout

Small groups

Start exercise notebooks

- Split up into Zoom breakout rooms
- Ask questions of instructors and other participants

Consultation Period

Exercise notebooks

Your own data

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

Module Layout

00a-rstudio_guide.md
00b-debugging_resources.md
00c-good-scientific-coding-practices.md

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

01-intro_to_base_R-live.Rmd
01-intro_to_base_R.Rmd
01-intro_to_base_R.nb.html
02-intro_to_ggplot2-live.Rmd
02-intro_to_ggplot2.Rmd
02-intro_to_ggplot2.nb.html
03-intro_to_tidyverse-live.Rmd
03-intro_to_tidyverse.Rmd
03-intro_to_tidyverse.nb.html

These are **Instruction** notebooks.
We'll walk through these together,
step-by-step, during the workshop.

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 **Exercise** notebooks.
Use these to practice what you
have learned. We're here to help!

Module cheatsheets cover key functions

<https://github.com/AlexsLemonade/training-modules/tree/2021-november/module-cheatsheets>

dplyr

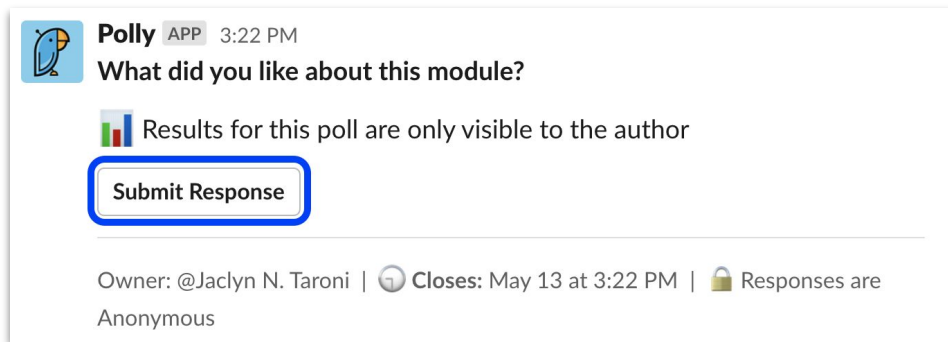
Read the `dplyr` package documentation [here](#).

A vignette on the usage of the `dplyr` package can be found [here](#).

Library/Package	Piece of code	What it's called	What it does
<code>dplyr</code>	<code>%>%</code>	Pipe operator	Funnels a <code>data.frame</code> through tidyverse operations
<code>dplyr</code>	<code>filter()</code>	Filter	Returns a subset of rows matching the conditions of the specified logical argument
<code>dplyr</code>	<code>arrange()</code>	Arrange	Reorders rows in ascending order. <code>arrange(desc())</code> would reorder rows in descending order.
<code>dplyr</code>	<code>select()</code>	Select	Selects columns that match the specified argument
<code>dplyr</code>	<code>mutate()</code>	Mutate	Adds a new column that is a function of existing columns
<code>dplyr</code>	<code>summarise()</code>	Summarise	Summarises multiple values in an object into a single value. This function can be used with other functions to retrieve a single output value for the grouped values. <code>summarize</code> and <code>summarise</code> are synonyms in this package.
<code>dplyr</code>	<code>rename()</code>	Rename	Renames designated columns while keeping all variables of the <code>data.frame</code>
<code>dplyr</code>	<code>group_by()</code>	Group By	Groups data into rows that contain the same specified value(s)
<code>dplyr</code>	<code>inner_join()</code>	Inner Join	Joins data from two data frames, retaining only the 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 screenshot shows a Slack poll interface. At the top, there's a header with a penguin icon, the name 'Polly', the label 'APP', and the time '3:22 PM'. Below this is the poll question: 'What did you like about this module?'. A small bar chart icon is followed by the text 'Results for this poll are only visible to the author'. A prominent blue-outlined button labeled 'Submit Response' is centered below the text. At the bottom, a footer line contains the text 'Owner: @Jaclyn N. Taroni | ⌚ Closes: May 13 at 3:22 PM | 🔒 Responses are Anonymous'.

Polly APP 3:22 PM

What did you like about this module?

Results for this poll are only visible to the author

Submit Response

Owner: @Jaclyn N. Taroni | ⌚ Closes: May 13 at 3:22 PM | 🔒 Responses are Anonymous

- 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 CCDL staff and each other.

Presentations

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

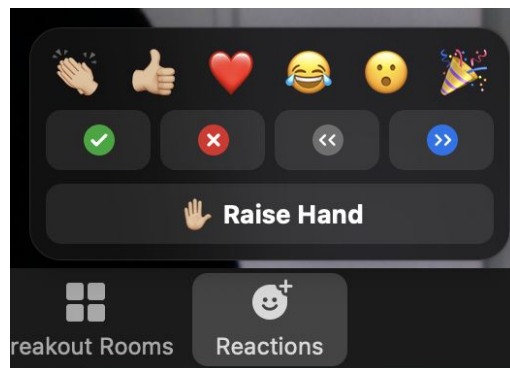
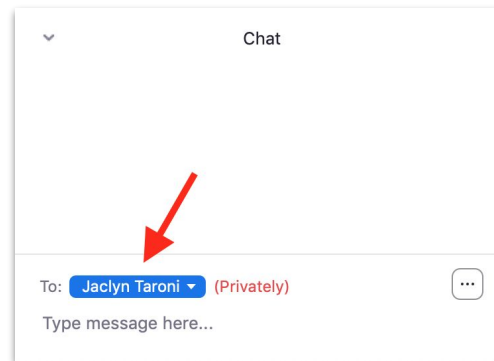


Virtual Training Procedures



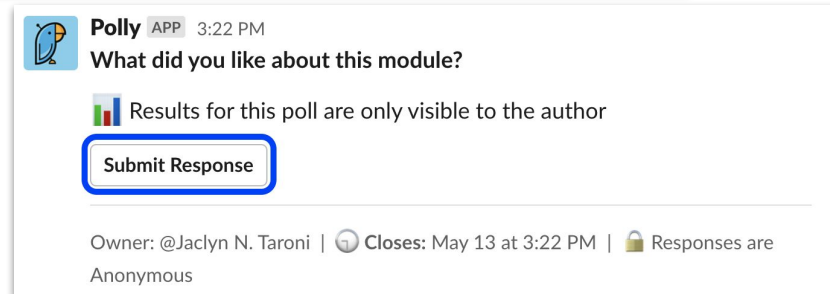
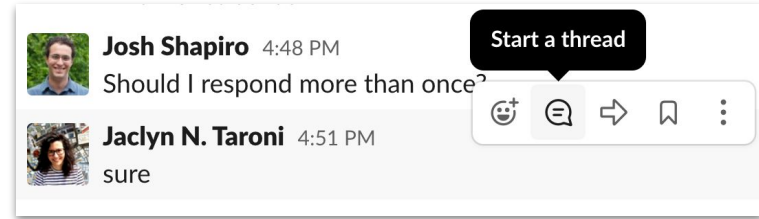
General Zoom Etiquette

- Keep your microphone muted
- Type questions in the Chat window (directed to the host)
 - 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 **#2021-november-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 CCDL staff member



- I have an **general question** that does not need an answer right away.
 - **Post** in #2021-november-training
- I'm having trouble **logging in** to RStudio Server
 - **Direct Message** a CCDL 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 #2021-november-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 #2021-november-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 CCDL staff member
- I'm having trouble **logging in** to RStudio Server
 - **Direct Message** a CCDL staff member

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