HBC
Harvard Chan Bioinformatics Core

Introduction to differential gene expression (DGE) analysis

https://tinyurl.com/hbc-dge

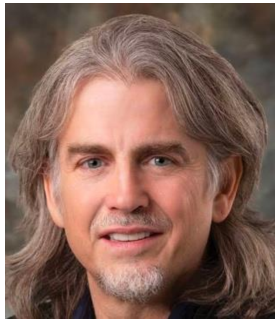
Harvard Chan Bioinformatics Core

Introductions!

Shannan Ho Sui
Director

Meeta Mistry
Associate Director

Lorena Pantano
Director of Bioinformatics Platform



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Faculty Advisor

Upen Bhattarai

Heather Wick

Will Gammerdinger

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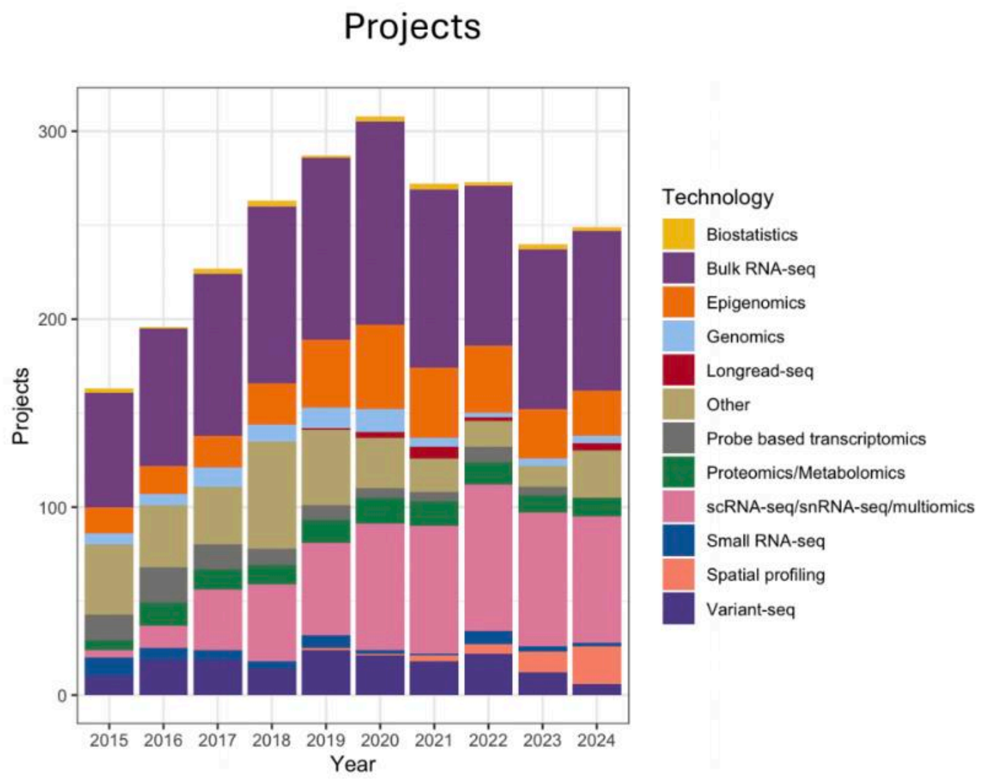
Consulting

- Transcriptomics: Bulk, single cell, small RNA
- Epigenomics: ChIP-seq, CUT&RUN, ATAC-seq, DNA methylation
- Variant discovery: WGS, resequencing, exome-seq and CNV
- Multiomics integration
- Spatial biology
- Experimental design and grant support



https://bioinformatics.sph.harvard.edu/services

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CFAR
HARVARD UNIVERSITY
CENTER FOR AIDS RESEARCH

HARVARD
T.H. CHAN
SCHOOL OF PUBLIC HEALTH

NIEHS

HARVARD
CATALYST
THE HARVARD CLINICAL AND TRANSLATIONAL SCIENCE CENTER

HARVARD
MEDICAL SCHOOL

Training

- Hands-on workshops design to reflect best practices, reproducibility and an emphasis on experimental design
  - Basic Data Skills
    - Shell
    - R
  - Advanced Topics: Analysis of high-throughput sequencing data
    - Chromatin Biology
    - Bulk RNA-seq
    - Differential Gene Expression
    - scRNA-seq
    - Variant Calling
  - Current Topics in Bioinformatics

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DF/HCC
DANA-FARBER / HARVARD CANCER CENTER

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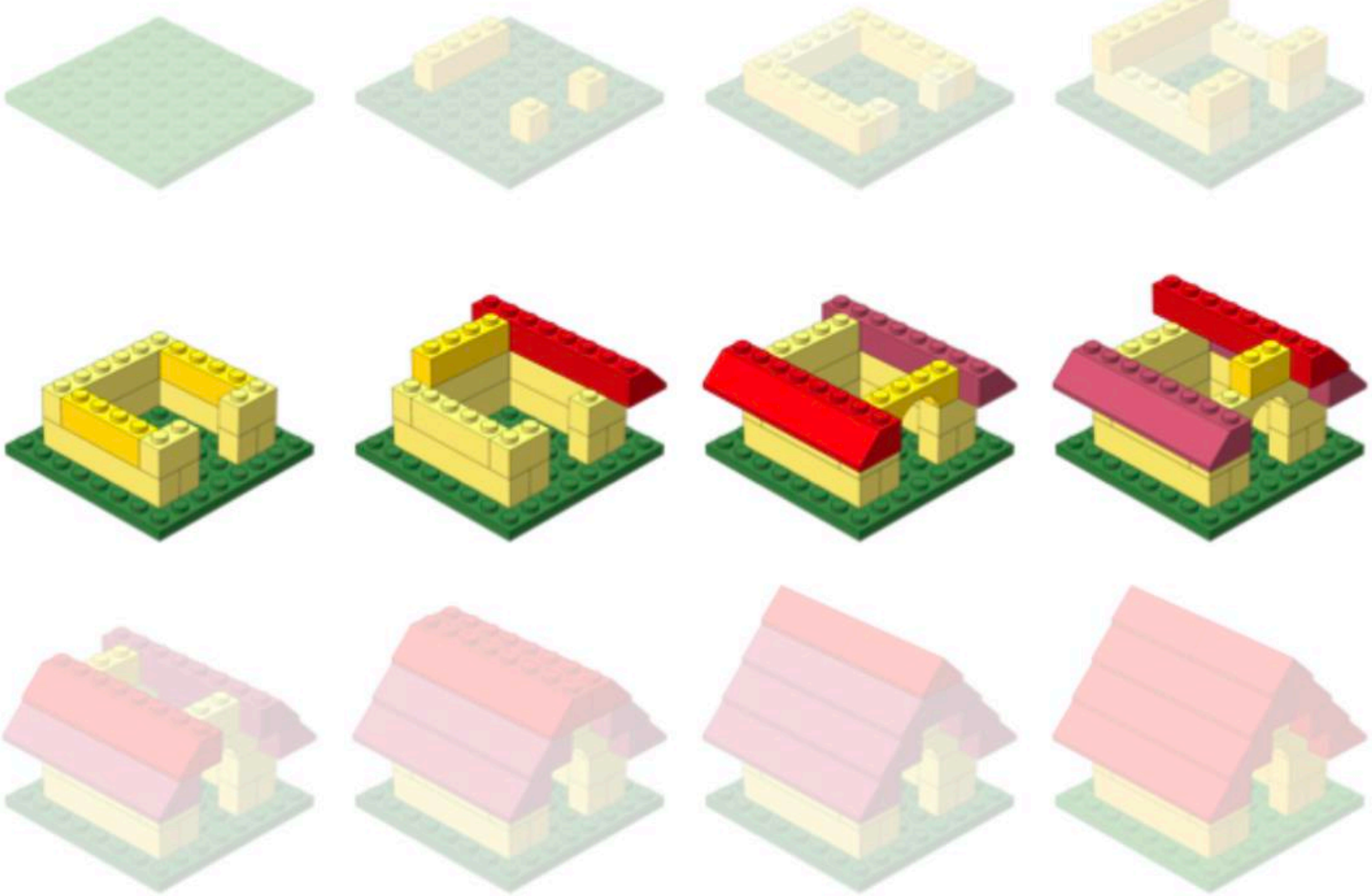
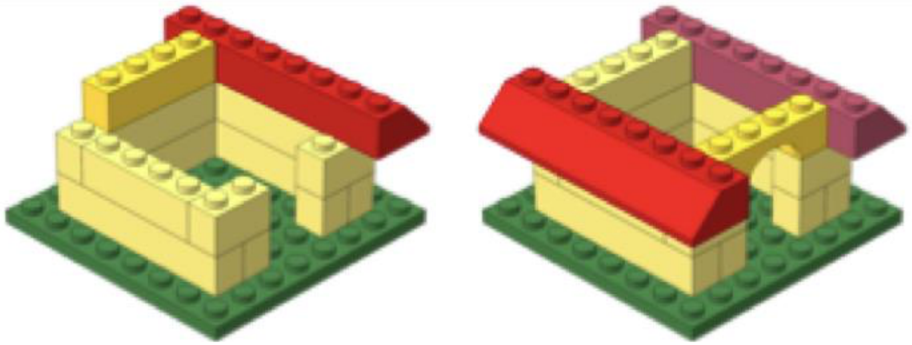
Workshop scope



Bioinformatics Data Analysis

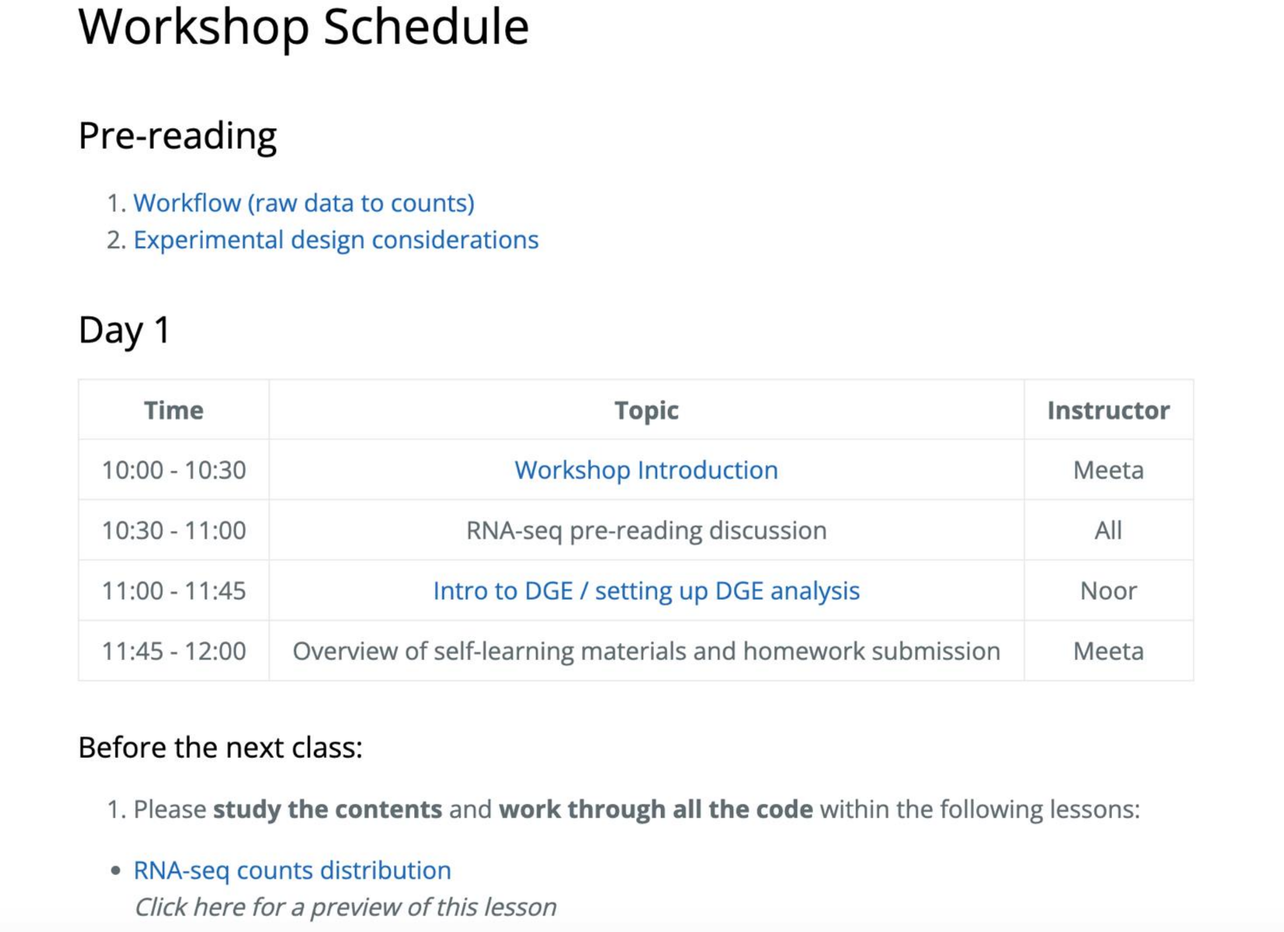
Workshop Scope



- Understand the considerations for performing statistical analysis on RNA-seq data
- Start with gene counts (after alignment and counting)
- Perform QC on count data
- Use DESeq2 to perform differential expression analysis on the count data and obtain a list of significantly different genes
- Visualize results of the analysis
- Perform functional analysis on the lists of differentially expressed genes

Logistics

Course schedule



Workshop Schedule

Pre-reading

1. Workflow (raw data to counts)
2. Experimental design considerations

Day 1

Time	Topic	Instructor
10:00 - 10:30	Workshop Introduction	Meeta
10:30 - 11:00	RNA-seq pre-reading discussion	All
11:00 - 11:45	Intro to DGE / setting up DGE analysis	Noor
11:45 - 12:00	Overview of self-learning materials and homework submission	Meeta

Before the next class:

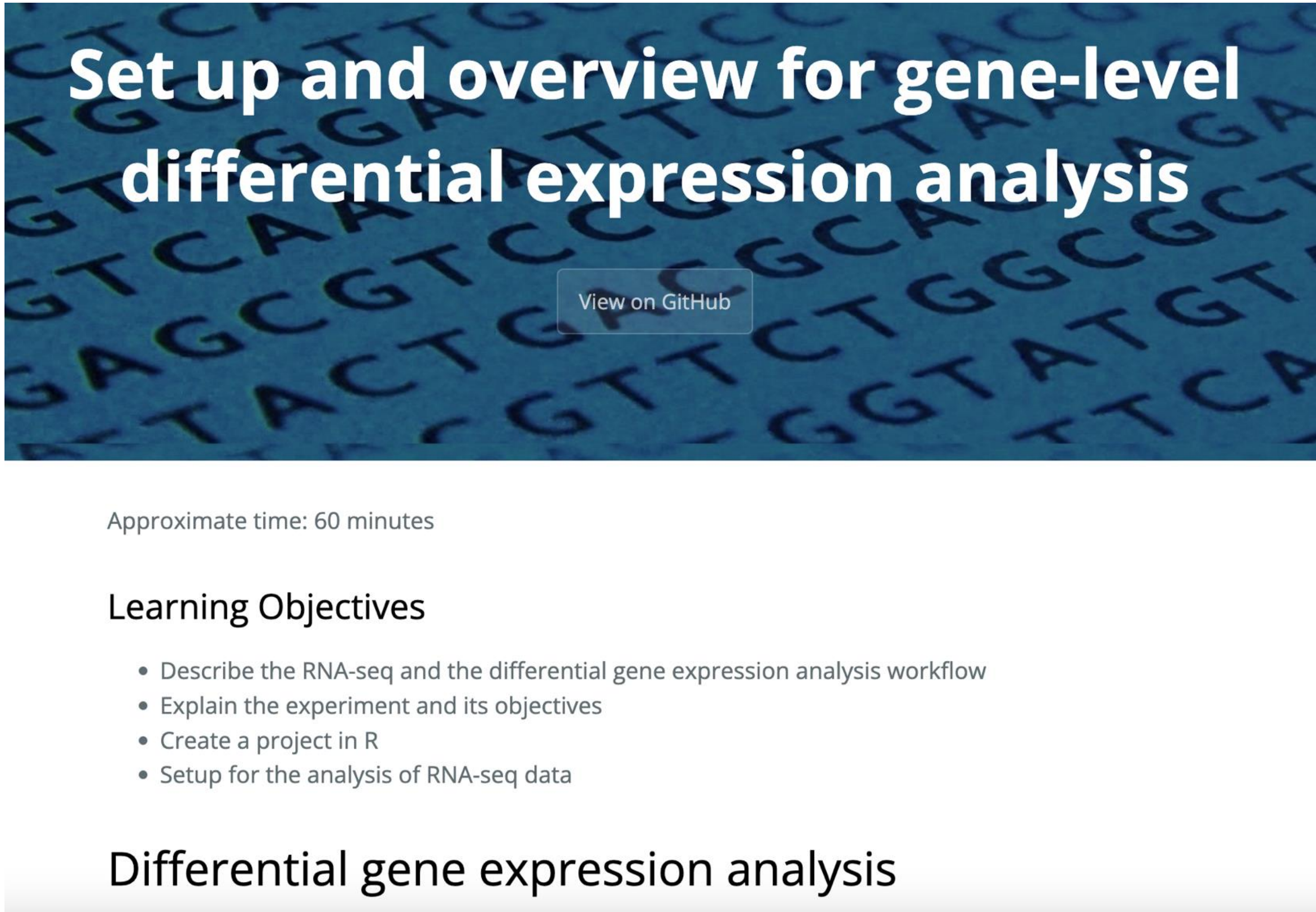
1. Please **study the contents** and **work through all the code** within the following lessons:

- RNA-seq counts distribution
  Click here for a preview of this lesson

https://tinyurl.com/hbc-dge

Course materials

- We continuously update our materials to reflect changes in the field/software



Set up and overview for gene-level differential expression analysis

Approximate time: 60 minutes

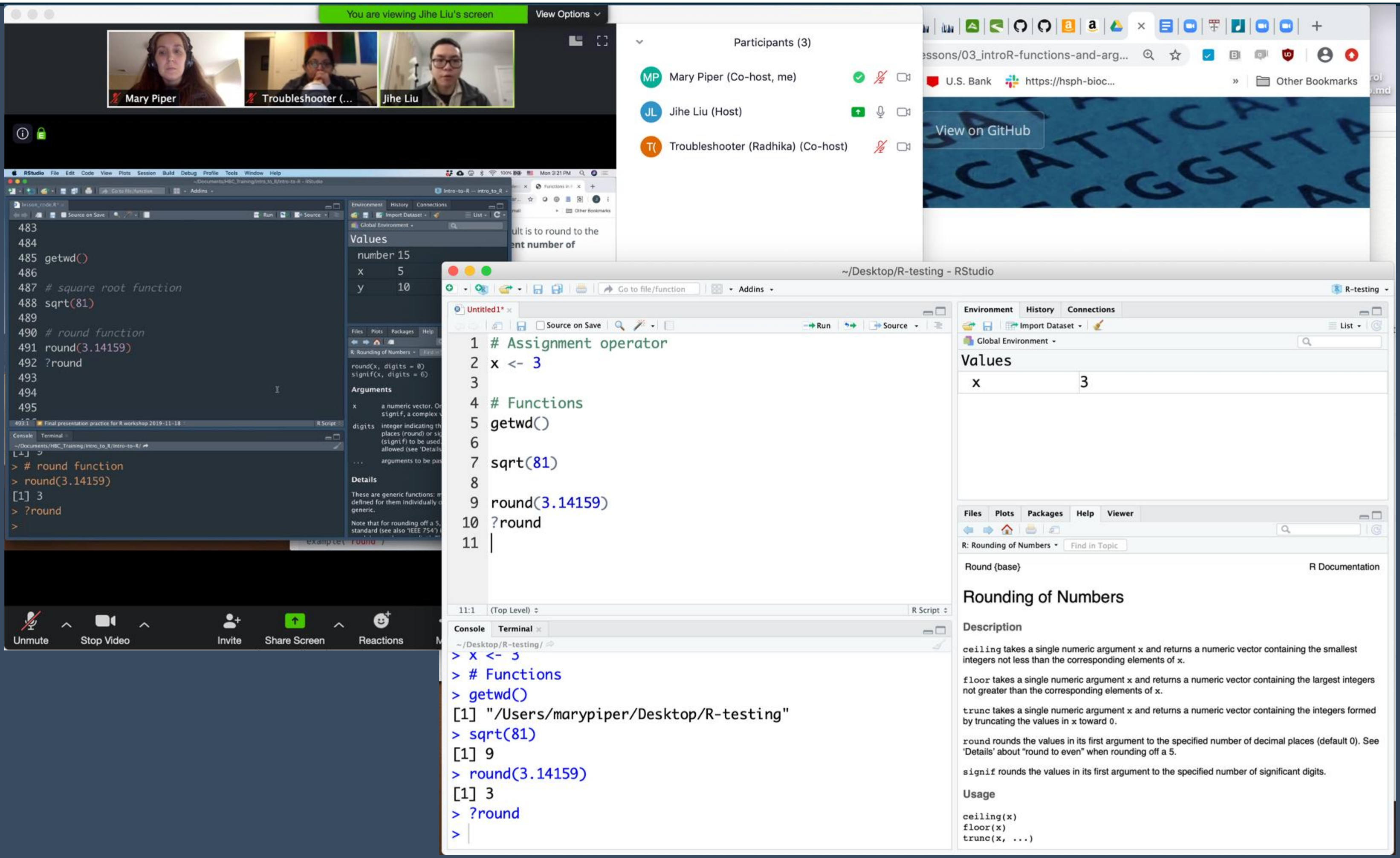
Learning Objectives

- Describe the RNA-seq and the differential gene expression analysis workflow
- Explain the experiment and its objectives
- Create a project in R
- Setup for the analysis of RNA-seq data

Differential gene expression analysis

https://tinyurl.com/hbc-dge

Single Screen & 3 Windows



```
getwd()

# square root function
sqrt(81)

# round function
round(3.14159)
?round
```

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> # round function
> round(3.14159)
[1] 3
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```
# Assignment operator
x <- 3

# Functions
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Values

x	3

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[1] "/Users/marypiper/Desktop/R-testing"
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Round {base} R Documentation

Rounding of Numbers

Description

`ceiling` takes a single numeric argument `x` and returns a numeric vector containing the smallest integers not less than the corresponding elements of `x`.

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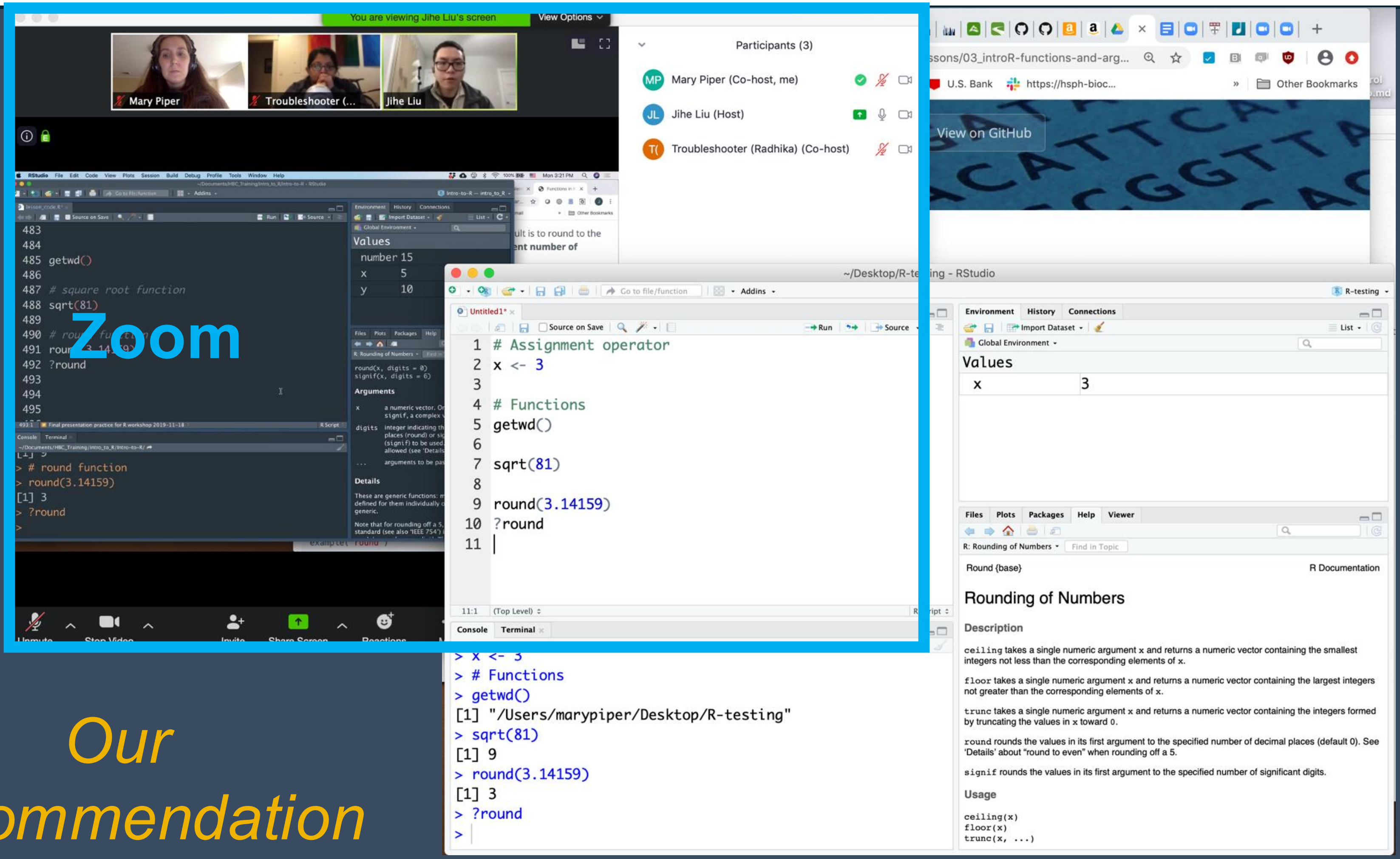
Usage

```
ceiling(x)
floor(x)
trunc(x, ...)
```

Single Screen & 3 Windows



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Our Recommendation

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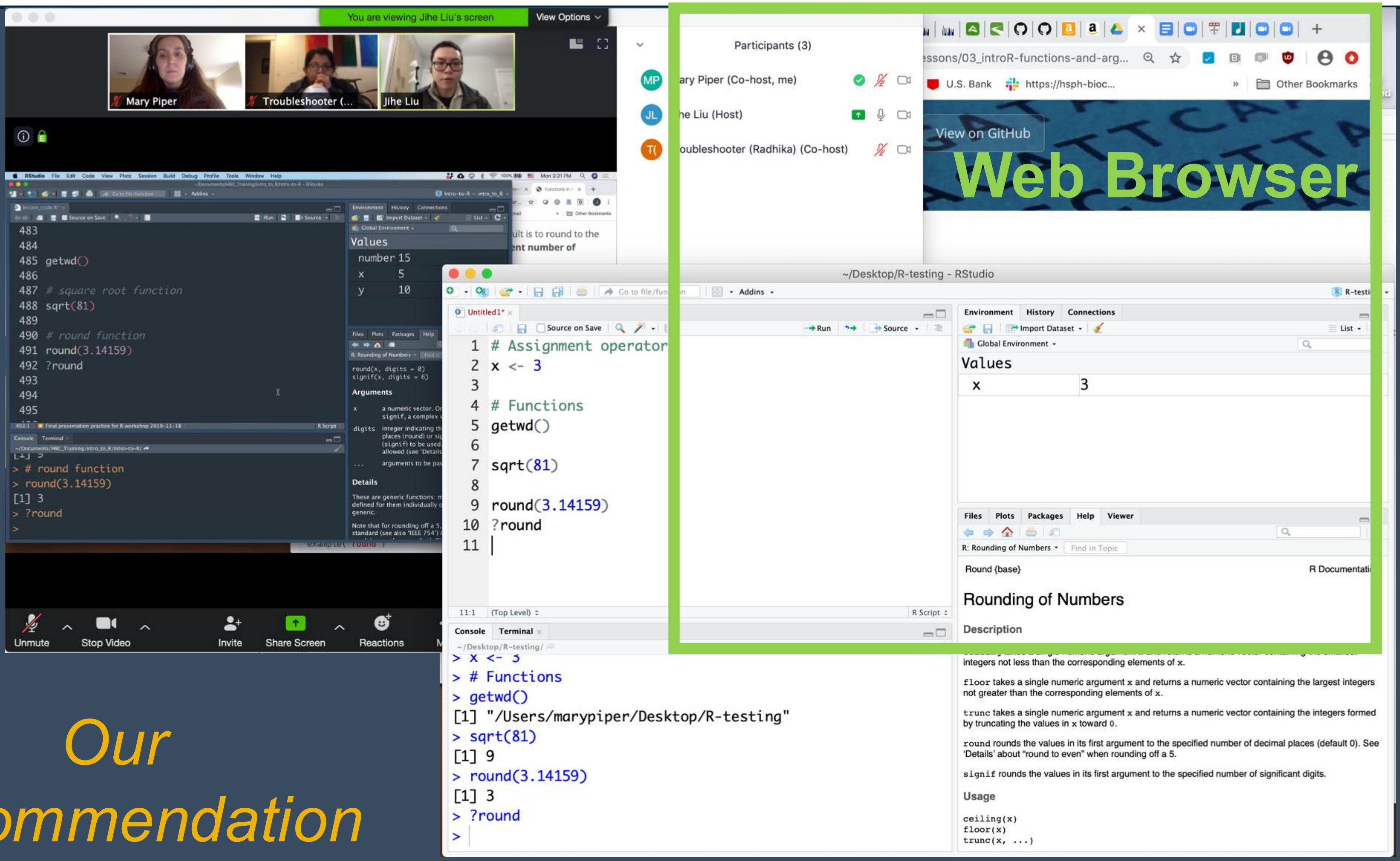

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number 15
x 5
y 10

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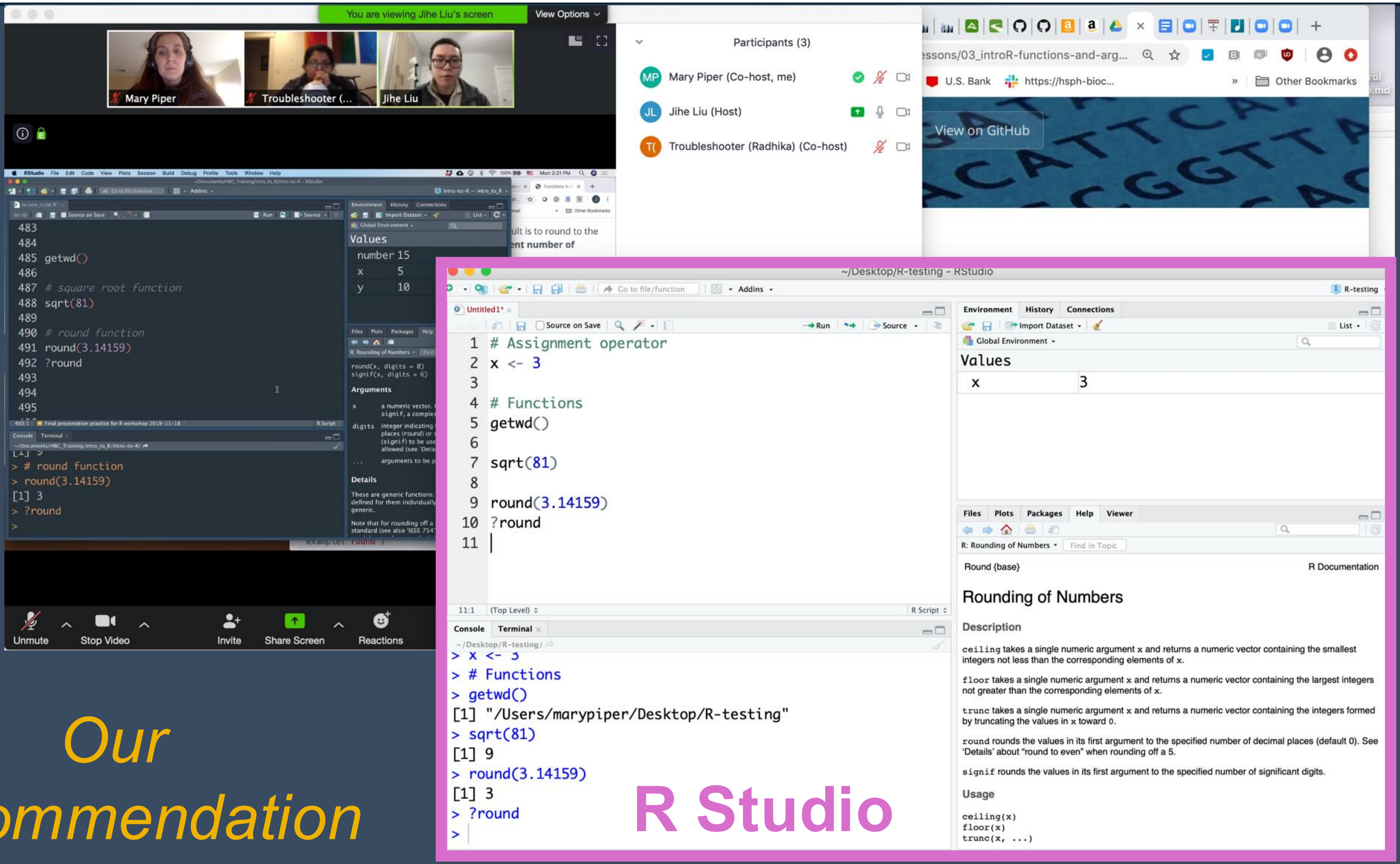

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R Studio

Our Recommendation

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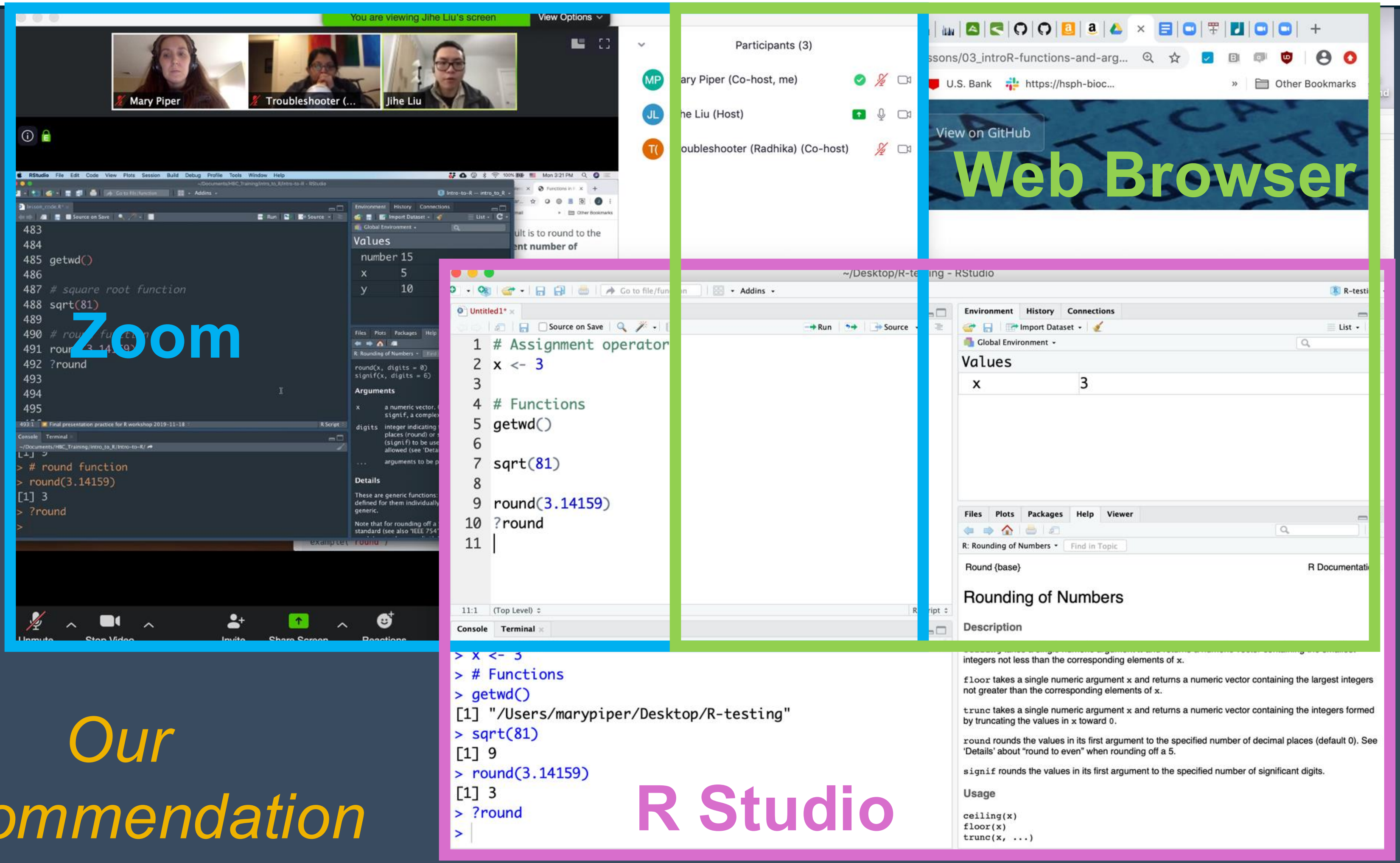

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Our Recommendation

Course participation

- Mandatory review of self-learning lessons and assignments
- Attendance required for all classes
- Your questions and active participation drive learning
- **We look forward to all of your questions!**

Course participation

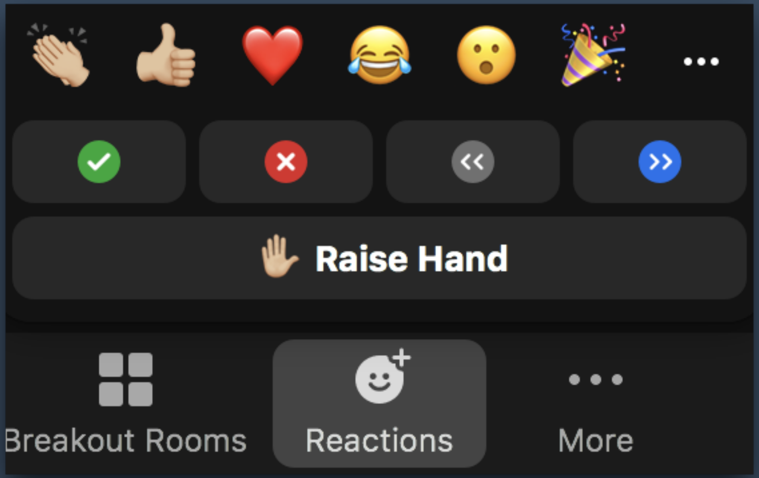
- At-home lessons and exercises after each session
- Cover material not previously discussed
- Provides us feedback to help pace the course appropriately
- 3-5 hours to complete
- Homework load is heavier in the beginning of this workshop series and tapers off

Using AI for Assignments

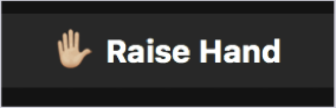
- Do
  - Try to resolve error messages with it
  - Test code written by AI on a dataset where you have expected results
  - Take the time to review the generated code line-by-line
- Don’t
  - Implement it in replacement to learning
  - Write code that you don’t understand
  - Assume the output from an AI process is correct

Odds & Ends

- Quit/minimize all applications that are not required for class
- Are you all set?
  - ✓ = "agree", "I'm all set"
  - ✗ = "disagree", "I need help"



Odds & Ends

- Questions for the presenter?
  - Post the question in the Chat window OR
  -  when the presenter asks for questions
  - Let the Moderator know

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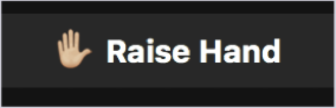
    -  Raise Hand when the presenter asks for questions
    - Let the Moderator know
- Technical difficulties with software?
    - Start a private chat with the Troubleshooter with a description of the problem

Contact Us

- *HBC training team:* hbctraining@hsph.harvard.edu
- *HBC consulting:* bioinformatics@hsph.harvard.edu