SPATIAL OMICS DATA ANALYSIS WITH MAWA

SESSION 1: INTRODUCTION TO NIDAP. MAWA FUNDAMENTALS. SUPERVISED PHENOTYPING.

10/15/24, 1-2 PM

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All data generously provided by the David Wink lab

SESSION FORMAT

- It might be best to focus on the presentation and *not* follow along live in MAWA.

- There will be two videos of this session available (the recording and a reference video), plus the entire session script in writing, with screenshots.

- It might be better to dive into MAWA yourselves as homework (summarized at the end of the session).

- If you really want to try to follow along live in the app, start loading MAWA now by clicking this link.

- Don't stress about taking notes!

- Questions:

    - I'll periodically ask if there are questions

    - If I don't, feel free to interrupt

    - Or put it in the chat for Dante and Andrei to triage

WHAT IS MAWA?

- MAWA = Multiplex Analysis Web Apps

- New, open-source software capable of performing:

  - File handling

  - Phenotyping

  - Spatial analysis

- Includes multiple published and novel methodologies

- Extremely performant

- Aims to be user-friendly and accessible to users of all skill levels

INPUT DATA

- Spatial Transcriptomics:

    - **Technology summary**: Barcodes embedded in grids on specialized slides, RNA sequencing techniques.

    - **Raw data**: Transcript counts at each point on a grid.

    - **Processing**: Spatial deconvolution is used to obtain the gene expression profile (in counts) for each cell or spot.

- Spatial Proteomics (in the case of immunofluorescence):

    - **Technology summary**: Fluorophore-labeled antibodies bound to protein surface markers.

    - **Raw data**: Channel intensity at each pixel.

    - **Processing**: Deep-learning based image segmentation is used to obtain the protein intensity for each cell.

- Ultimately, both methods lead to text-based data with objects in rows and coordinates and numerical features in columns.

- Any data of this general format can be used in MAWA.

MAWA DEPLOYMENT PLATFORMS
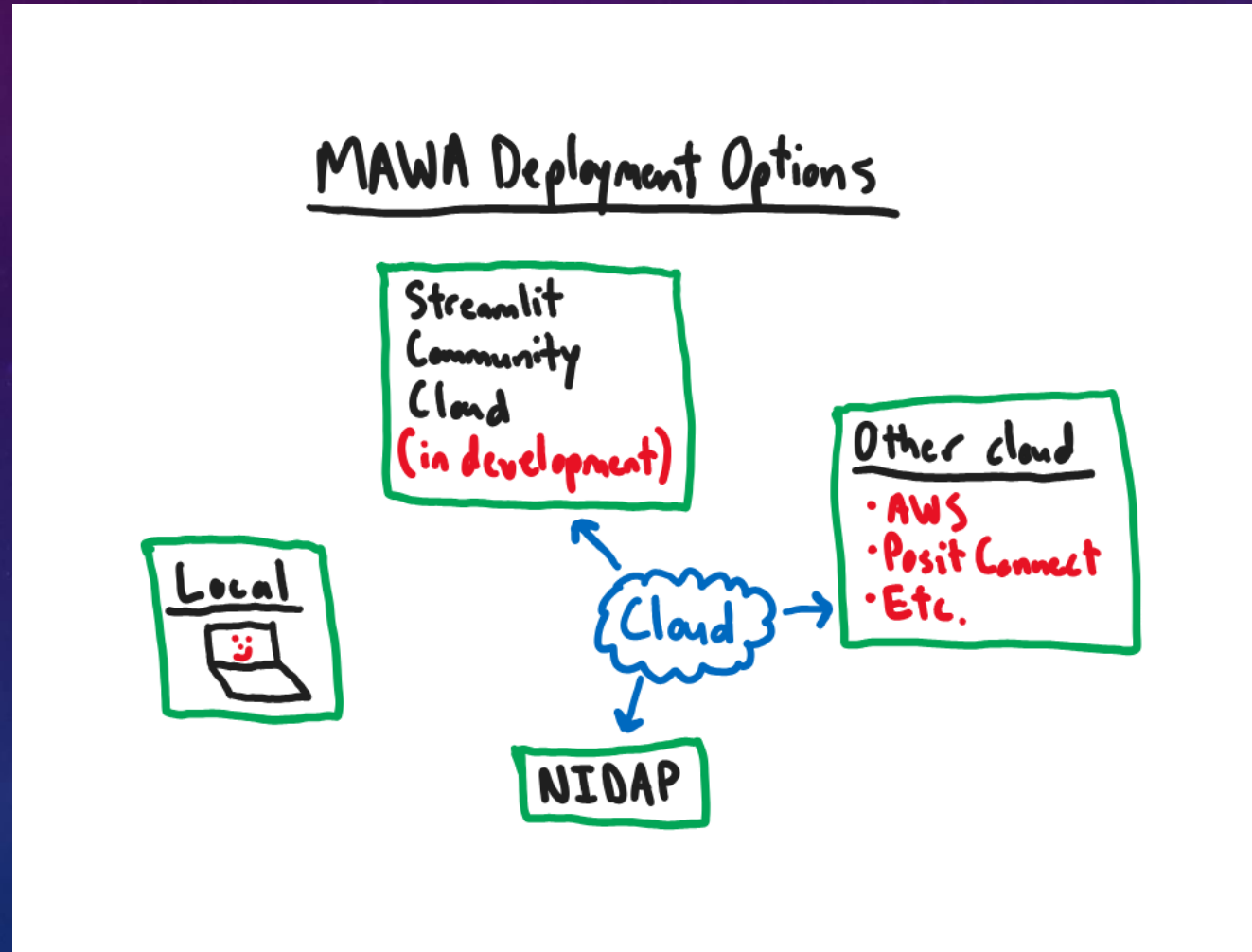
NIDAP

- NIDAP = NIH Integrated Data Analysis Platform

- Filesystem is organized into Projects, which usually correspond to research groups

- Each project can contain both user data and instances of NIDAP applications

- We have deployed MAWA in a public project on NIDAP for use during this workshop

- To create a secure deployment for your group, contact us (andrew.weisman@nih.gov) to have MAWA set up in your project
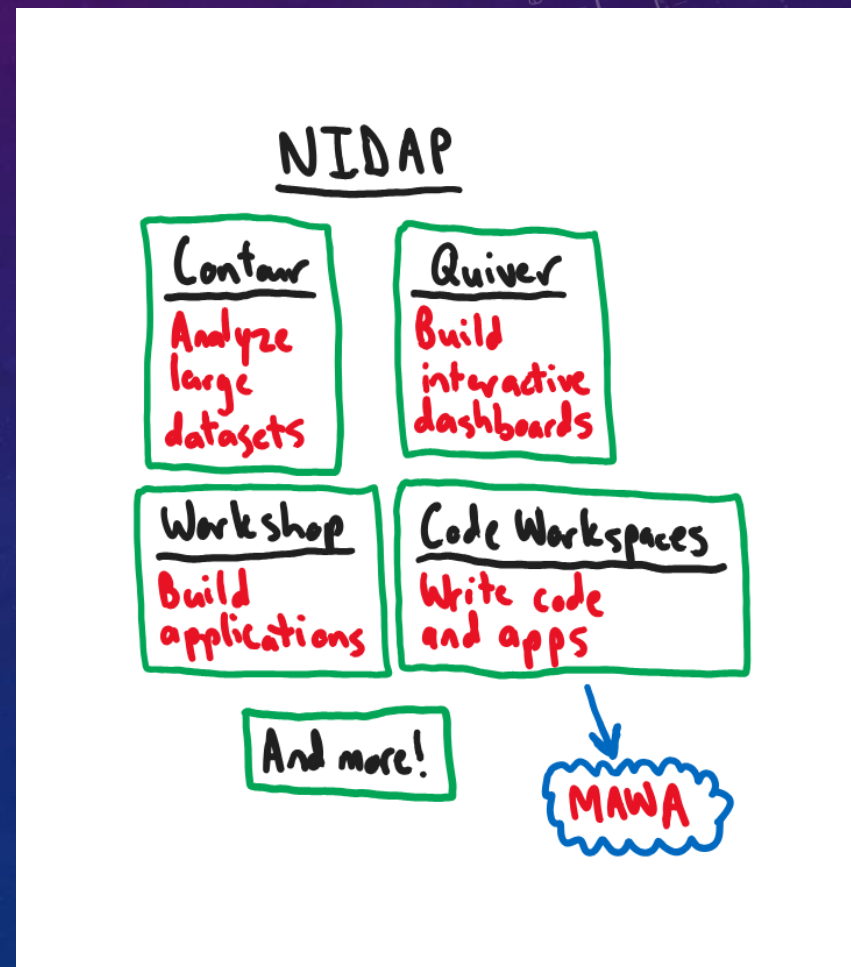
THRESHOLDED PHENOTYPING OPTIONS

| x coord	y coord	CD8	CD163	CK	FOXP3	"Species" phenotype
x1	y1	1	0	0	0	CD8+
x2	y2	0	1	1	0	CD163+ CK+

x coord	y coord	CD8	CD163	CK	FOXP3	"Custom" phenotype
x1	y1	1	0	0	0	CTL
x2	y2	0	1	1	0	Tumor M2

x coord	y coord	CD8	CD163	CK	FOXP3	"Marker" phenotype
x1	y1	1	0	0	0	CD8
x2	y2	0	1	1	0	CD163
x2	y2					CK

x coord	y coord	CD8	CD163	CK	FOXP3	Phenotypes		
						Species	Custom	Marker
x1	y1	1	0	0	0	CD8+	CTL	CD8
x2	y2	0	1	1	0	CD163+ CK+	Tumor M2	CD163
x2	y2							CK

MUTUALLY EXCLUSIVE PHENOTYPES
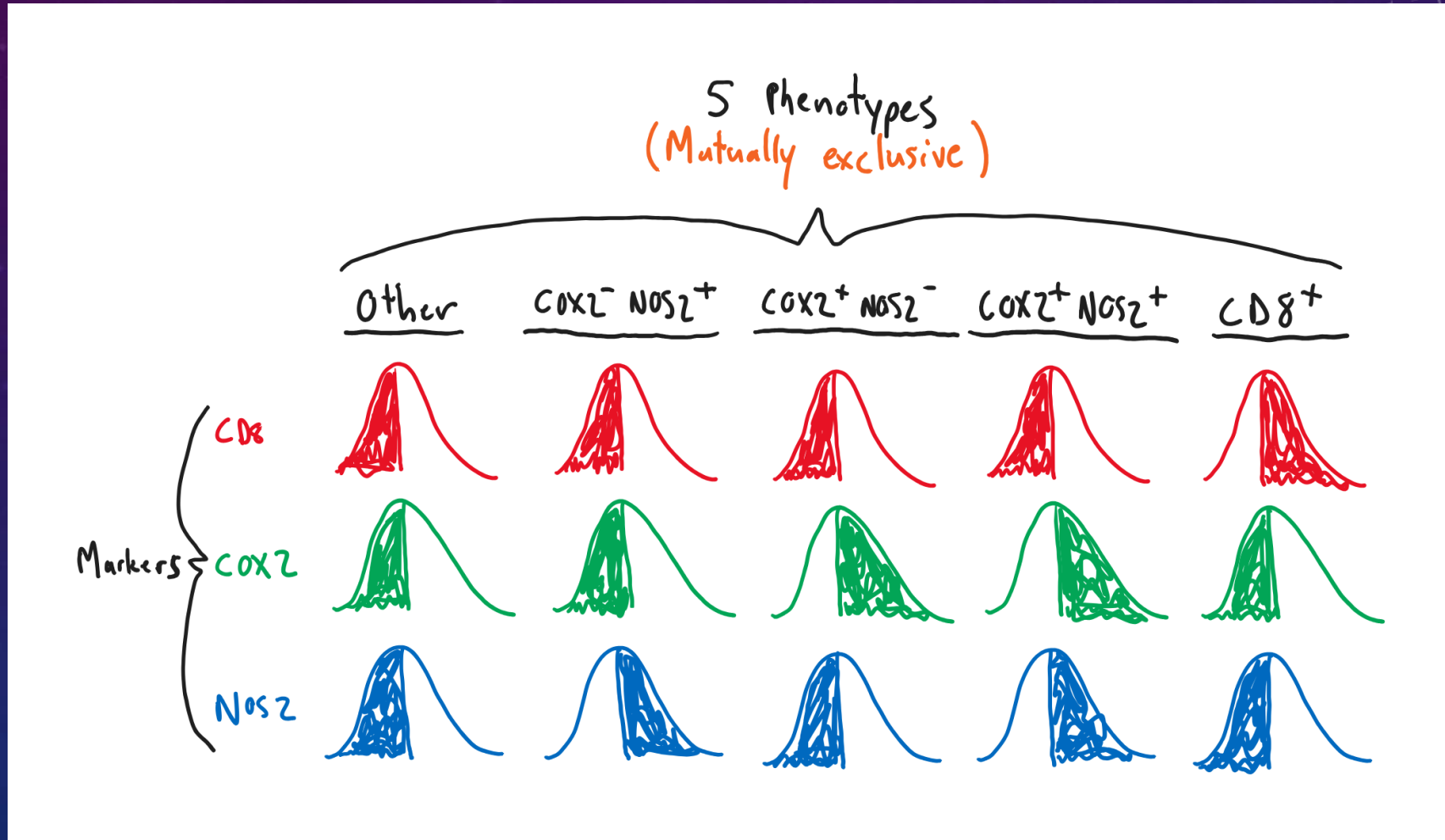
NON-MUTUALLY EXCLUSIVE PHENOTYPES
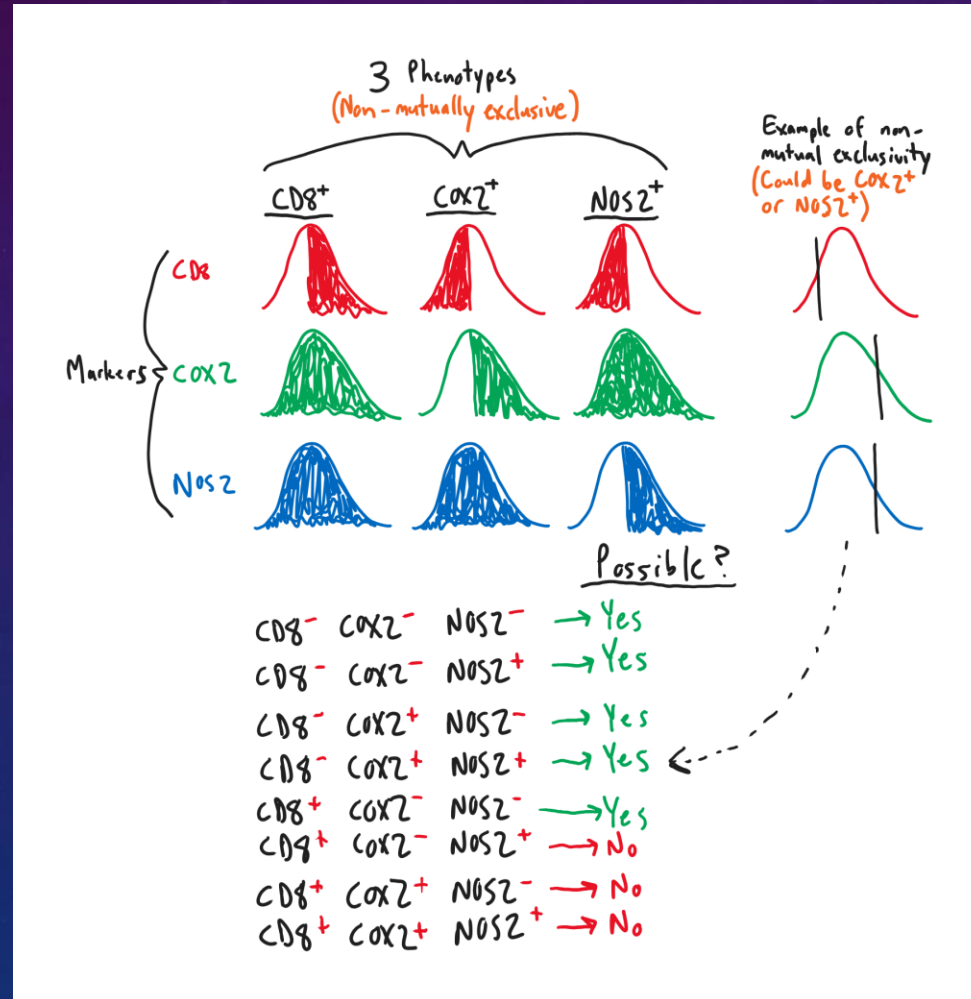
NEXT STEPS

- Homework:
  - Review the video and script for this session of this workshop.
  - Follow along step-by-step in the training MAWA deployment.
- Plan to attend the next sessions:
  - Tue 10/22, 1-2 PM
    - Unsupervised phenotyping.
    - Andrei Bombin, Ph.D.
  - Tue 10/29, 1-2 PM
    - Pairwise spatial analysis using hypothesis testing.
    - Andrew Weisman, Ph.D.
  - Wed 11/6, 11-12 PM
    - Neighborhood analysis using spatial UMAP.
    - Dante Smith, Ph.D.