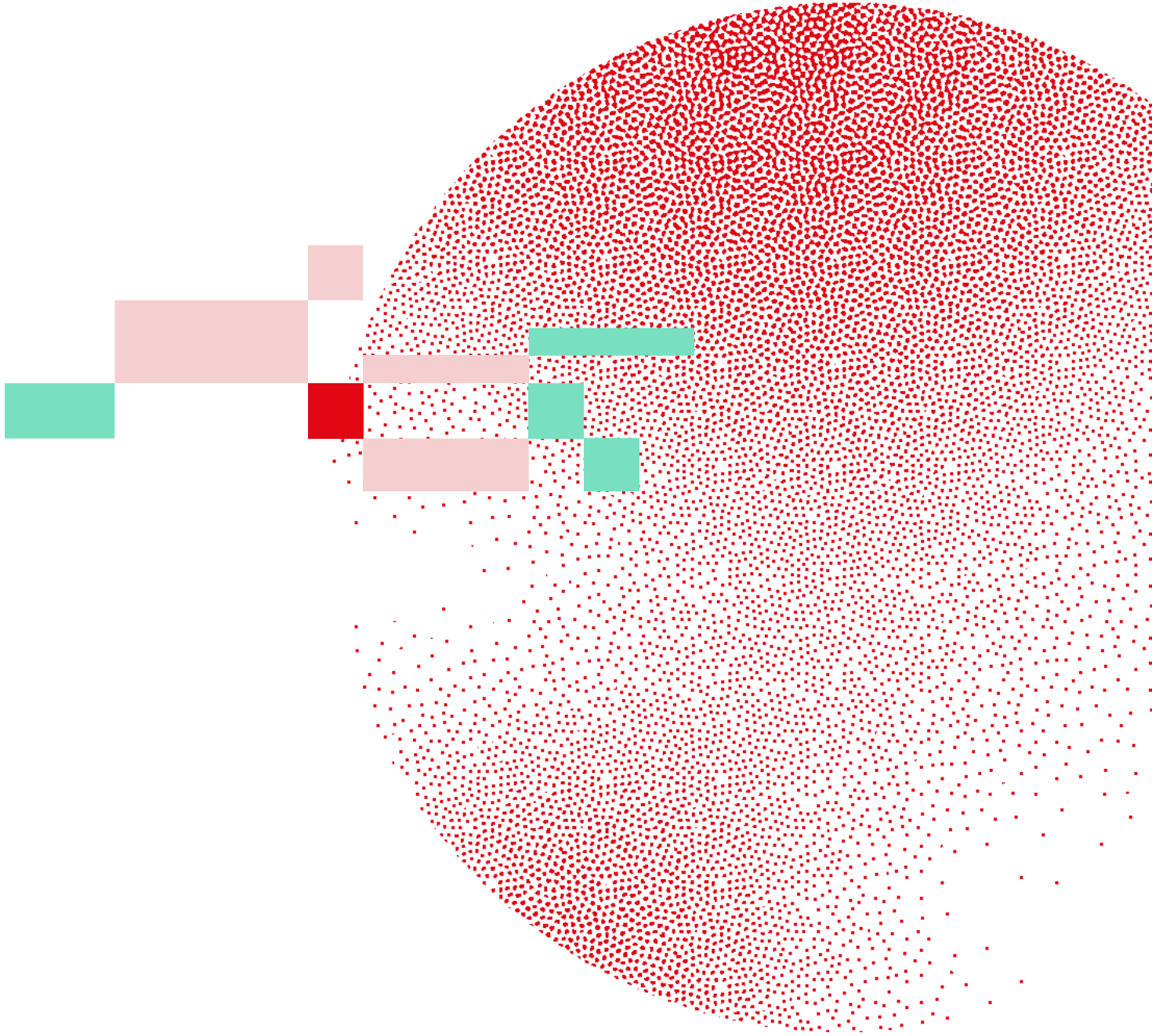
Swiss Institute of Bioinformatics



SINGLE-CELL TRANSCRIPTOMICS WITH R

Group Work

Bioinformatics trainers

July 2-4, 2025

elixir SWITZERLAND

Group Work

- **Work in groups to analyse a scRNAseq dataset**

  4 different dataset projects

- **Go through the project questions** (use the course materials)

- **Last day present your work**

Group Work

Group work: 15:30h – 17:00h everyday

- **Last day present your work**

Prepare few slides:

- Main methods and results (include visualisations)
- Challenges (QC, methods, interpretations..)
- Findings/conclusions

Choose a project:

Project 1 - Green Fluorescent Zebrafish

Project 2 - Drosophila on Cocaine

Project 3 - PBMCs of melanoma patients

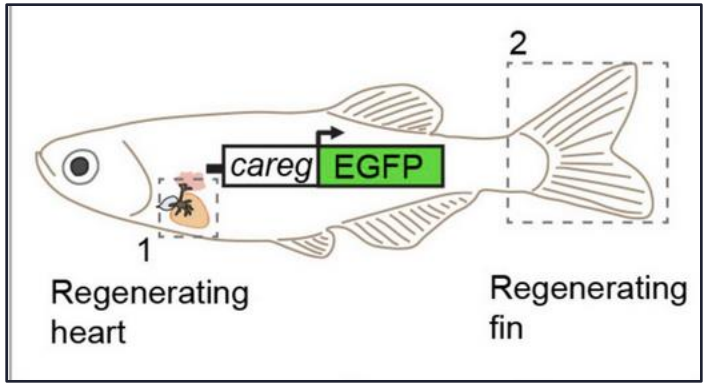
Project 4 - Cervical cancer and HPV

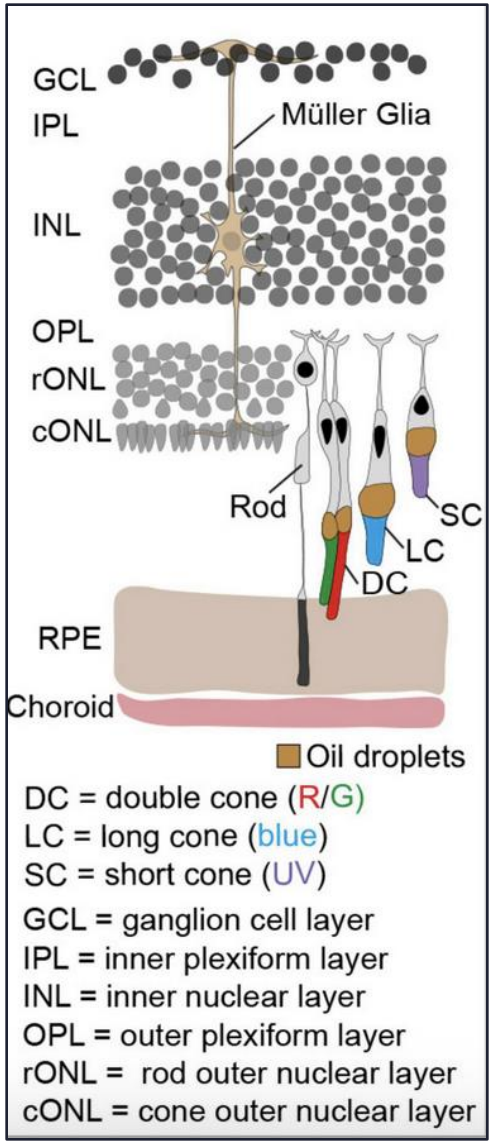
PROJECT 1

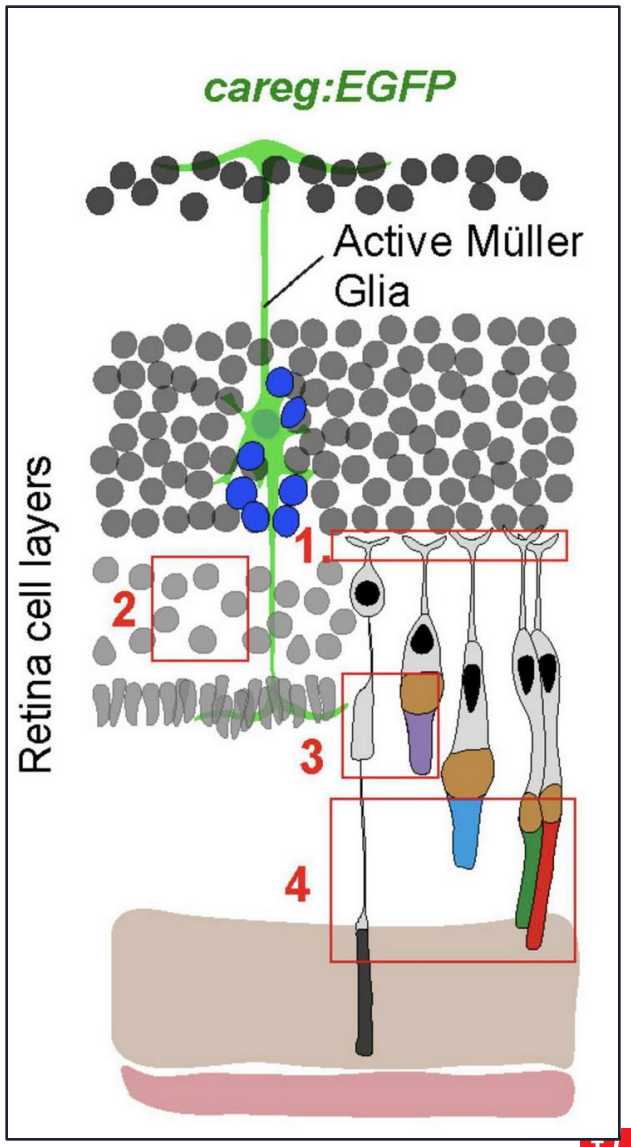
The regeneration-responsive element *careg* monitors activation of Müller glia after MNU-induced damage of photoreceptors in the zebrafish retina

Thomas Bise et al. 2023

Careg reporter detects activated MG, and provides a common marker of regeneration-competent cells

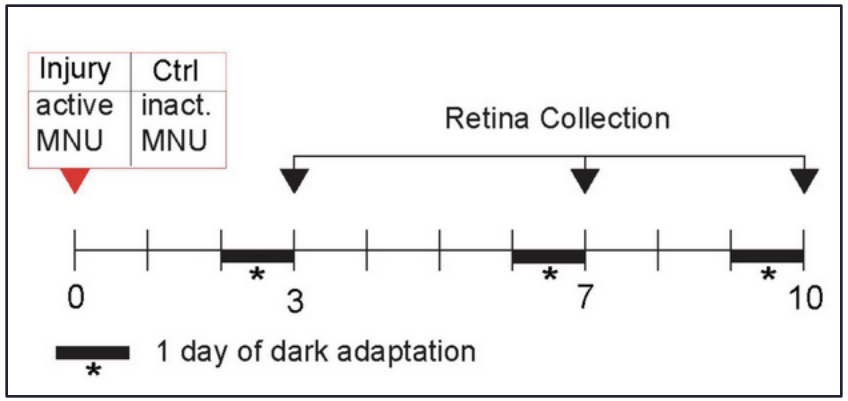


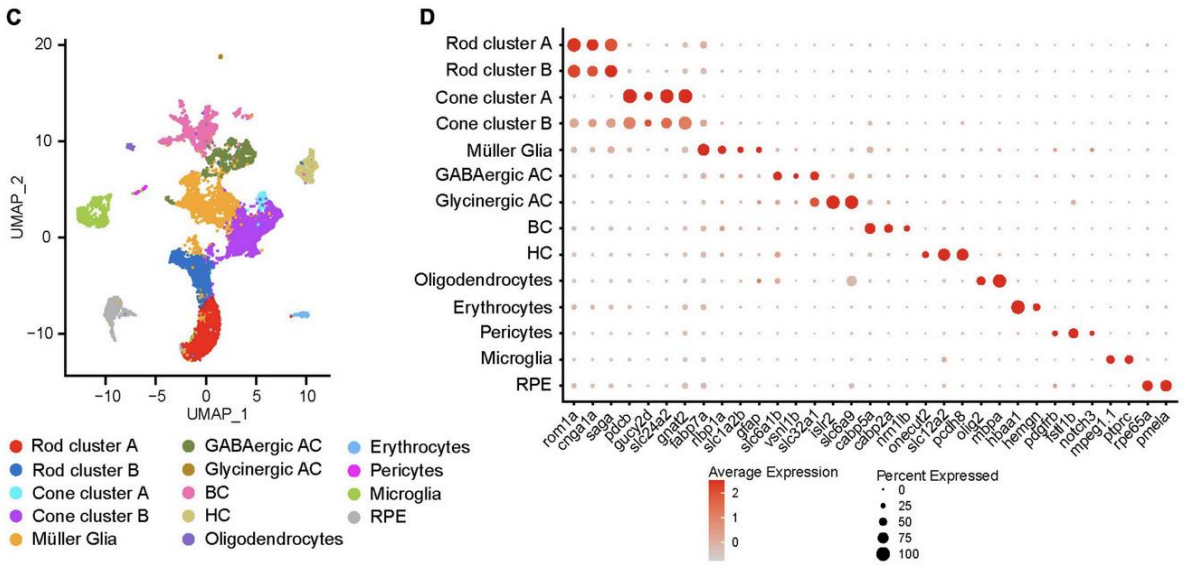




PROJECT 1

Species: Danio Rerio
Tissue: Retina
Conditions: Control, damaged tissue (3,7,10 days)





- Can you annotate the main cell types of interest: MG, cons and rods?
- Identify activated MG in the different conditions
- Look at molecular differences between injured and uninjured cons or rods

Project 2: Drosophila on cocaine

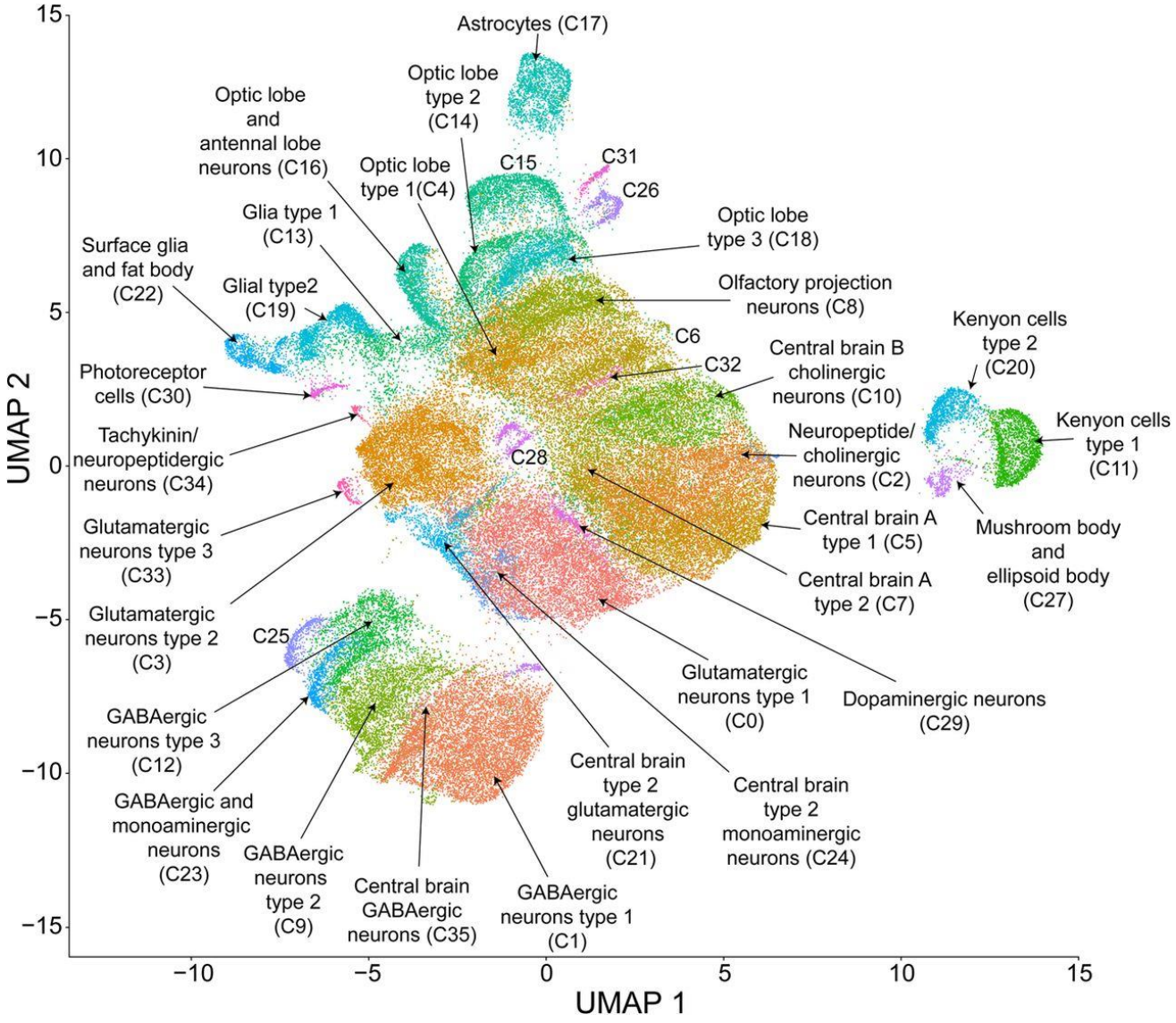
Study Focus: Investigating **single-cell transcriptional responses** in the Drosophila brain after acute cocaine exposure.

Unsupervised clustering of 86,224 cells revealed **36 distinct clusters**, representing major neuronal and glial cell types across brain regions.

Profound Sexual Dimorphism: Males exhibited more pronounced and widespread transcriptional changes to cocaine than females.

Available Data: Pre-processed single-cell count matrices (GEO GSE152495) for male/female, cocaine/sucrose conditions, ready for analysis.

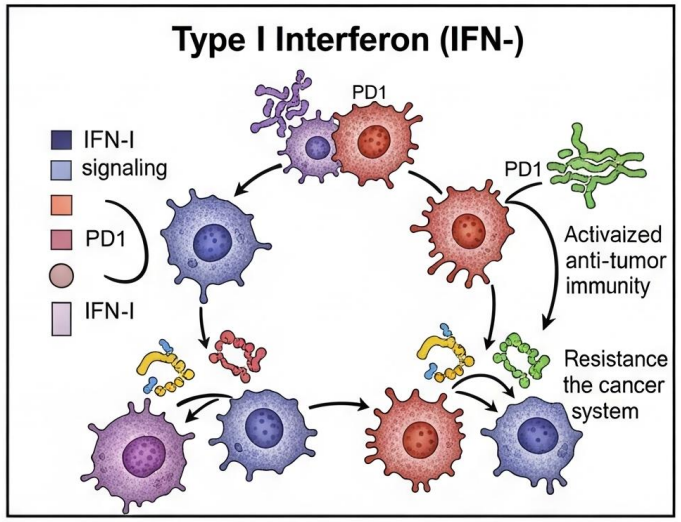
UMAP visualization and clustering



This figure will be very useful when you are performing the "Dimensionality Reduction" and "Unsupervised Clustering" steps, and especially for "Cluster Annotation" to compare your results and ensure you are identifying similar cell populations and their spatial relationships in the UMAP embedding.

Brandon M. Baker et al. Genome Res. 2021;31:1927-1937

Project 3 - IFN-I Responsiveness & PD1 Blockade



"PD1 Blockade" refers to a specific type of cancer immunotherapy that aims to reactivate immune cells to fight cancer.

IFN-I are alarm signals that cells release when they detect threats like viruses or certain aspects of cancer.

Question: Why some cancer patients respond well to a type of immunotherapy called PD1 blockade, while others don’t?

The Problem: Immunotherapy like PD1 blockade can be very effective against cancer, but it doesn't work for everyone.

Project 3 - IFN-I Responsiveness & PD1 Blockade

Our Approach: Analyzing single-cell RNA sequencing (scRNA-seq) data from healthy donors and 8 treated patients.

Key Finding: Patients with lower pre-therapy IFN-I responsiveness in CD4/CD8 effector T cells (Teff) showed:

- Signatures of improved immune function.
- Better therapy outcomes.

Available Data: Pre-processed scRNA-seq count matrices from GEO GSE199994.

Project 4 – Cervical Cancer Dataset

Aim 1 (paper 1): Understand the mechanisms of cervical cancer development mediated by HPV infection

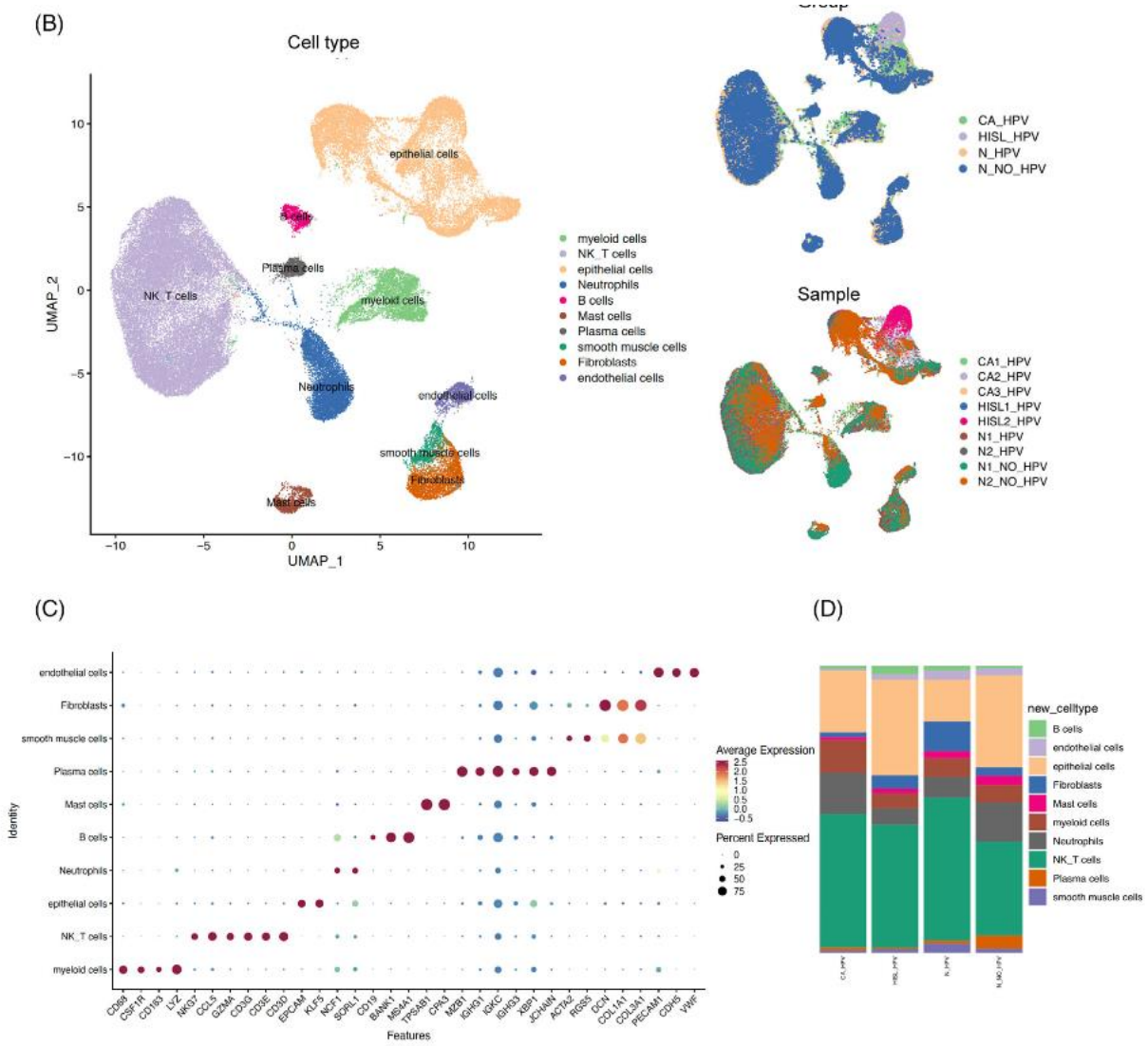
Aim 2 (paper 2): Understand the mechanisms of immune remodelling in cervical cancer

Data:

- Normal cervix (no HPV) (n = 2)
- Cervical cancer (with HPV) (n = 2)
- Normal cervix (with HPV) (n=2)
- Pre-cancer cervix (with HPV) (n=2)

} Use these samples only

Key Figure (try to reproduce)



Source: Guo (2023) - *Paper 1*

Choose a project (google form):

https://docs.google.com/forms/d/e/1FAIpQLSeQRkWostANryxYCKDG-UUYH1l3J98x249zrb89VHzY-3T8sw/viewform?usp=header

Project 1 - Green Fluorescent Zebrafish

Project 2 - Drosophila on Cocaine

Project 3 - PBMCs of melanoma patients

Project 4 - Cervical cancer and HPV