

CAA2: Work development (phase 1)

1. Identification of the work and date of the report

1.1. ***Impact of 5-FU treatment on tumor cell plasticity in colorectal cancer, a scRNAseq analysis***

1.2. 09/04/2024

2. Description of the progress of the project

2.1. Degree of fulfillment of the objectives and results foreseen in the work plan:

The objectives related to the R-script and standardization of the Seurat workflow are mostly assessed. Quality Control and processing of the raw matrix is understood, and the scRNAseq analysis by each timepoint is completed, a few final adjustments are required. For the integrative analysis, still markers are needed to identify, even though the workflow and bases of the analysis are assessed.

Still, the Script reproducibility is needed. Finally, a biological explanation of the preliminary results is not clearly defined.

Also, for the following of the master thesis, the General and objectives of the study are described here:

2.1.1. **General objectives:**

2.1.1.1. Demonstrate that translational control is one of the molecular mechanisms responsible for cellular reprogramming in response to 5-FU.

2.1.1.2. Isolate relevant cellular subpopulations for study.

2.1.1.3. Detail the transcriptome of these subpopulations.

2.1.1.4. Phenotypically characterize the cellular subpopulations.

2.1.1.5. Identify the role of these subpopulations in treatment tolerance and tumor recurrence.

2.1.2. **Specific objectives:**

2.1.2.1. Investigate the role of translational control in 5-FU-induced cellular reprogramming.

2.1.2.2. Employ and optimize single-cell RNA-seq workflow using Seurat Package from R to obtain relevant cellular subpopulations.

2.1.2.3. Perform detailed analyses of the transcriptome of these subpopulations using appropriate methodologies, thanks to Seurat Package Workflow and optimization.

- 2.1.2.4. Phenotypically characterize the cellular subpopulations using cell and molecular biology techniques.
    - 2.1.2.5. Evaluate the contribution of the identified cellular subpopulations to 5-FU treatment tolerance and tumor recurrence.
  - 2.2. Justification of the changes (if necessary)
- 3. List of activities carried out.
  - 3.1. Activities planned in the work plan.
    - 3.1.1. scRNAseq analysis during different timepoints of the experiment, scRNAseq analysis by parts.
      - 3.1.1.1. Raw data processing and simplification of scRNA matrix.
      - 3.1.1.2. Identification of clusters on each timepoint.
      - 3.1.1.3. Identification of cell markers for each timepoint and cluster.
    - 3.1.2. scRNAseq integrative analysis.
      - 3.1.2.1. Raw data processing and simplification of scRNA matrix, and integration of data.
      - 3.1.2.2. Identification of clusters of the Integrative analysis.
      - 3.1.2.3. Identification of cell markers for each cluster identified.
    - 3.1.3. Comparison between integrative analysis against the non-integrative.
    - 3.1.4. Standardization of the reproducibility of the R-script and environment.
    - 3.1.5. Biological explanation of the results.
  - 3.2. Unplanned and carried out activities or programs.
- 4. List of deviations in timing and mitigation actions (if applicable) and update of the schedule (if appropriate).
- 5. List of partial results obtained so far (deliverables that are attached).
- 6. Comments from your private director if you consider it necessary.
- 7. Personal assessment