

Single-cell Morphology Quality Control (coSMicQC)

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I. Erroneous outliers and analysis

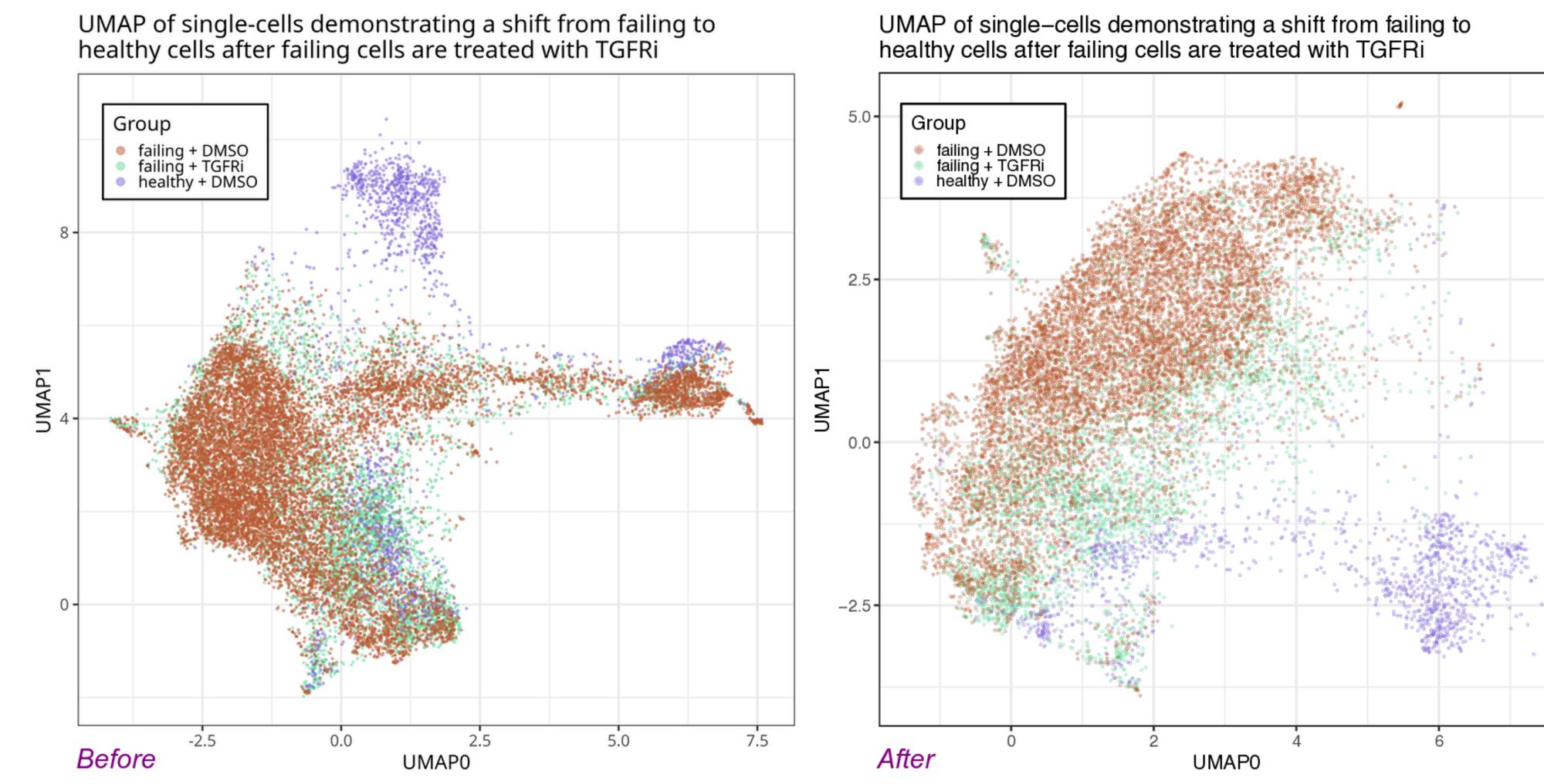
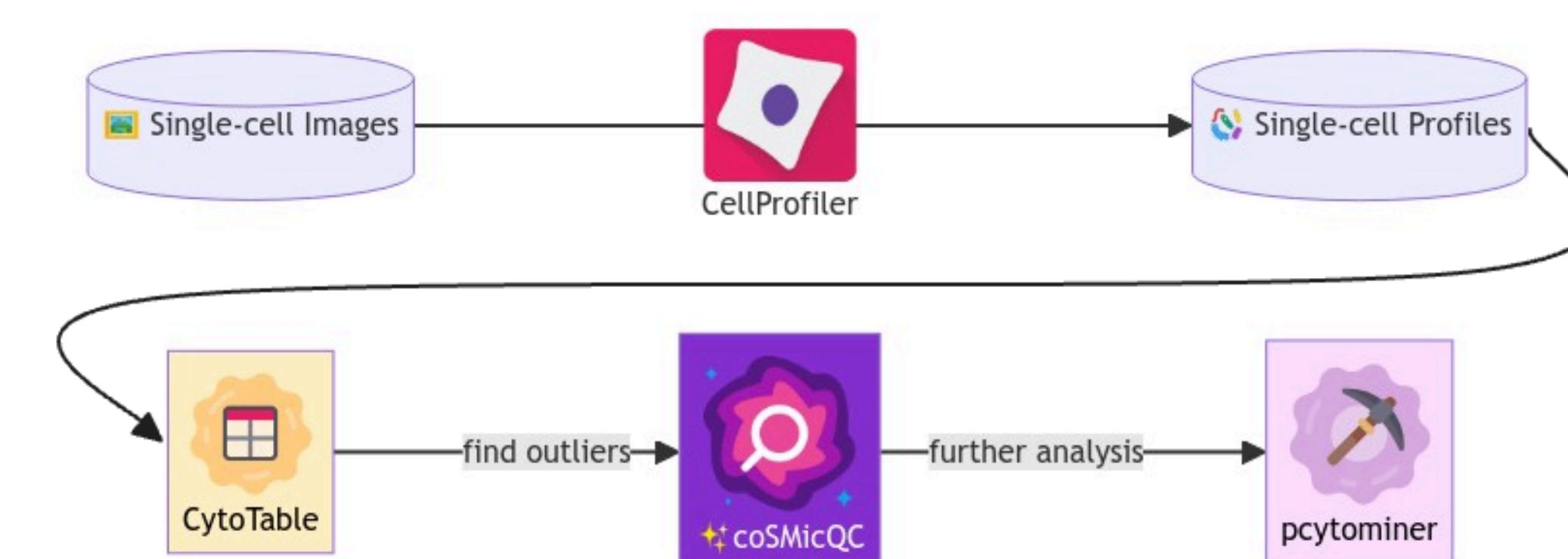


Figure 1: Extra clustering islands can be seen when looking at morphological profiles linked to poor segmentation, which when removed, better reveal patterns in the data.

Segmentation errors during single-cell morphology image analysis such as misidentifying cell compartments or artifacts as cells can lead to inaccurate single-cell measurements and *erroneous anomalies* within the data (Figure 1). If single-cell quality control is performed, it often uses bespoke methods or aggregate data into bulk profiles to avoid discrepancies caused by anomaly outliers. These techniques make it challenging to perform *quality control* on the data, impeding the potential for meaningful discoveries.

II. Single-cell quality control package



To address these challenges, we introduce **coSMicQC** (**S**ingle-cell **M**orphology **Q**uality **C**ontrol), an open source Python package designed to enhance the accuracy of single-cell morphology analysis. **coSMicQC** offers default and customizable thresholds for quality control, integrating seamlessly into both command line and Python API workflows.

III. Getting started with coSMicQC

☆ 1) Installation

```
# pip install from pypi
pip install coSMicQC

# or install directly from source
pip install git+https://github.com/WayScience/
coSMicQC.git
```

coSMicQC may be installed from PyPI or source.

☆ 2) Finding outliers

```
import cosmicqc
# find outliers from single-cell profiles
scdf = cosmicqc.analyze.find_outliers(
    df="single-cell-profiles.parquet",
    metadata_columns=[
        "Metadata_ImageNumber",
        "Image_Metadata_Plate_x"
    ],
    feature_thresholds={
        "Nuclei_AreaShape_Area": -1},
)
```

Number of outliers: 328
Outliers Range:
Nuclei_AreaShape_Area Min: 734.0
Nuclei_AreaShape_Area Max: 1904.0

| Nuclei_AreaShape_Area | Metadata_ImageNumber | Image_Metadata_Plate_x |
|-----------------------|----------------------|------------------------|
| 921.0 | 2 | Plate_2 |
| 845.0 | 2 | Plate_2 |
| 1024.0 | 2 | Plate_2 |
| 787.0 | 2 | Plate_2 |
| 1347.0 | 2 | Plate_2 |
| ... | ... | ... |

Figure 2: The `find_outliers` function in coSMicQC uses single-cell feature thresholds to provide a report on how many outliers were detected (Python API or CLI). We use z-scores to help define thresholds used throughout coSMicQC.

```
# CLI interface for coSMicQC find_outliers
$ cosmicqc find_outliers \
--df single-cell-profiles.parquet \
--metadata_columns \[Metadata_ImageNumber\] \
--feature_thresholds \{"Nuclei_AreaShape_Area": -1\}
```

Number of outliers: 328
Outliers Range:
Nuclei_AreaShape_Area Min: 734.0
...

☆ 3) Visualizing outlier distributions

```
import cosmicqc
# label and show outliers within the profiles
scdf = cosmicqc.analyze.label_outliers(
    df="single-cell-profiles.parquet",
    include_threshold_scores=True,
).show_report()
```

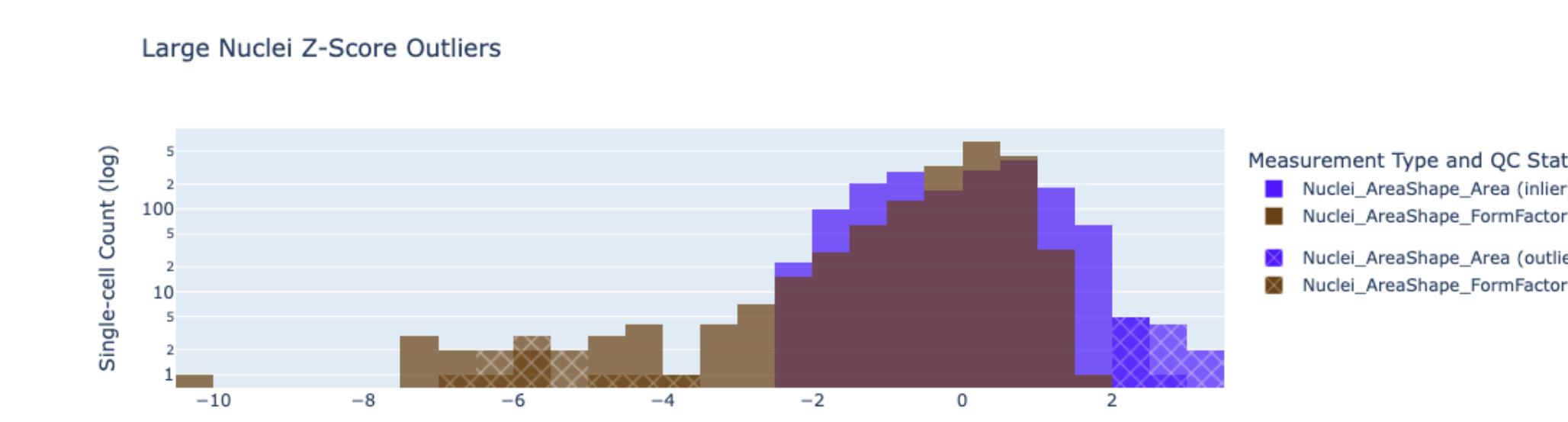


Figure 3: _coSMicQC enables erroneous anomaly analysis through the `label_outliers` function, which appends z-score data for features, and the `CytoDataFrame.show_report` method to visualize where outliers are detected within the dataset..

☆ 4) Understanding outlier segmentations

```
import cosmicqc
# passing image and mask dirs to display images
cosmicqc.CytoDataFrame(
    data="single-cell-profiles.parquet",
    data_context_dir="./image_directory/",
    data_mask_context_dir="./mask_directory/",
)
```

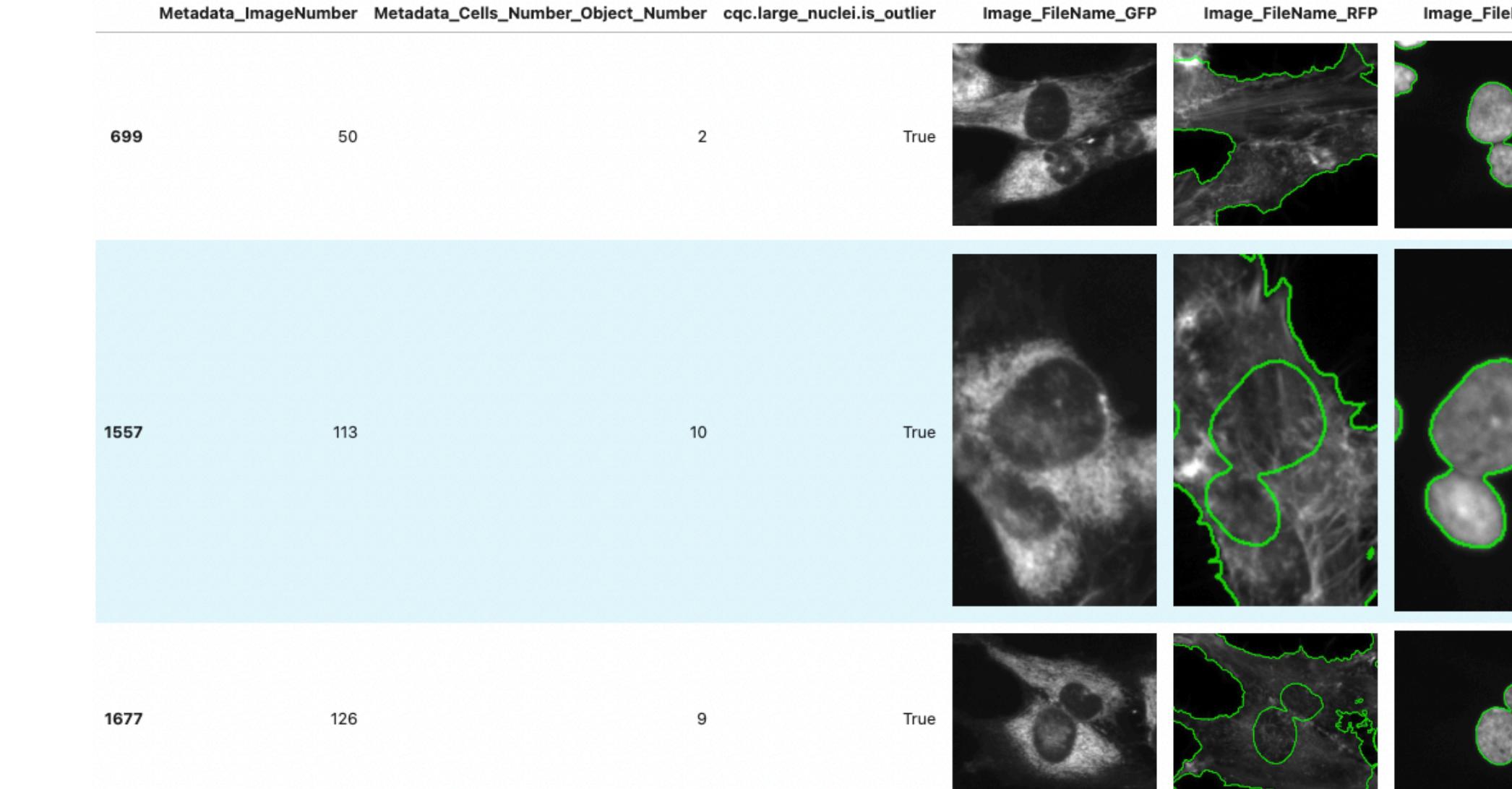


Figure 4: Interactive visualizations that help users identify outlier distributions through the `CytoDataFrame` – a novel data format that links single-cell measurements with their corresponding images and segmentation masks in real-time, enriching data analysis and interpretation.

IV. Real-world applications

Bootstrap ROC AUC Distributions

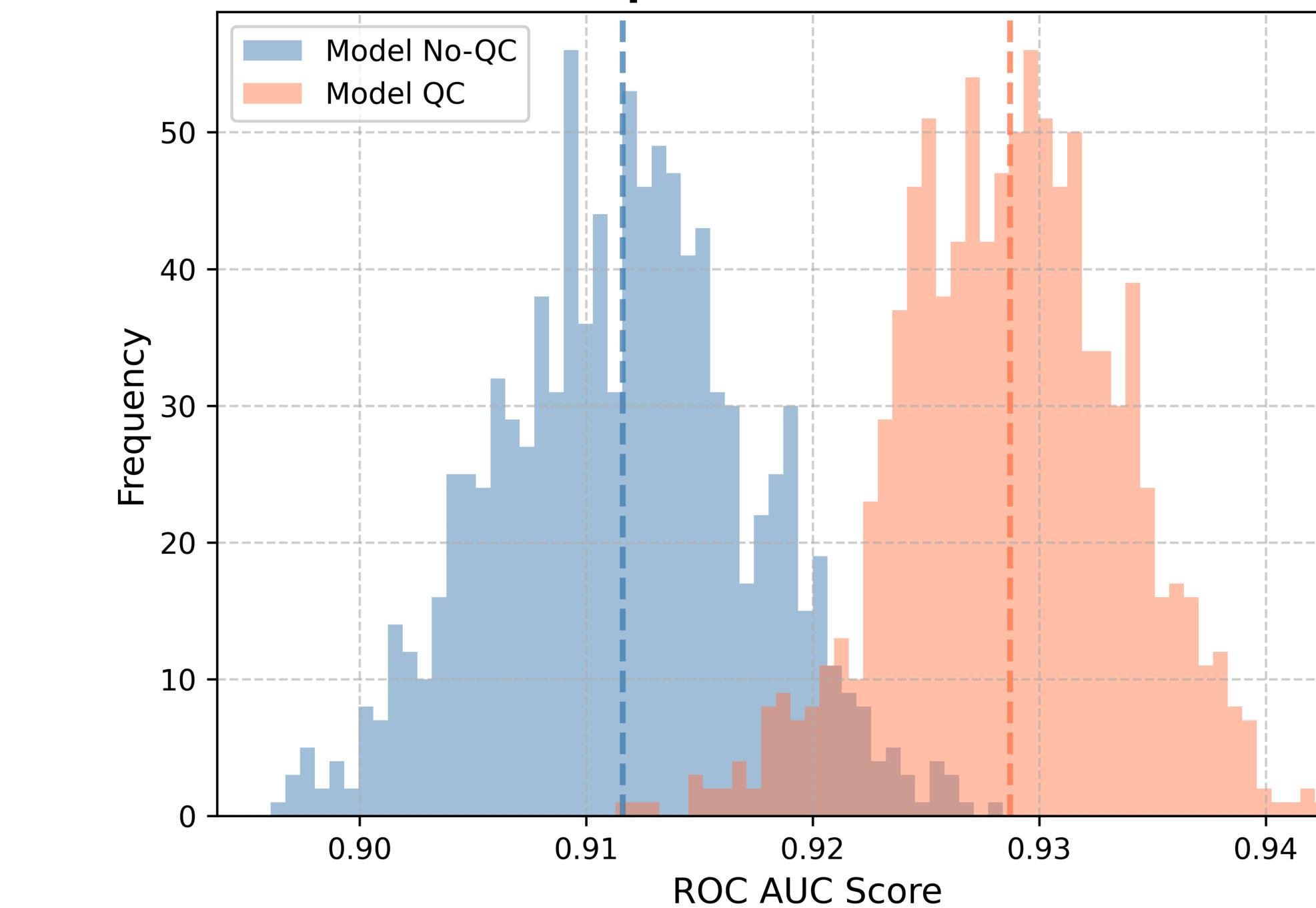


Figure 5: A representation of ROC AUC scores from multiple random samples taken from a holdout dataset (that has had QC applied) being applied to the QC trained model and no-QC trained model. The QC model outperforms the no-QC model, and the average performance of the QC model is significantly higher than the no-QC model (t -statistic = -72.1, p -value = 0.0).

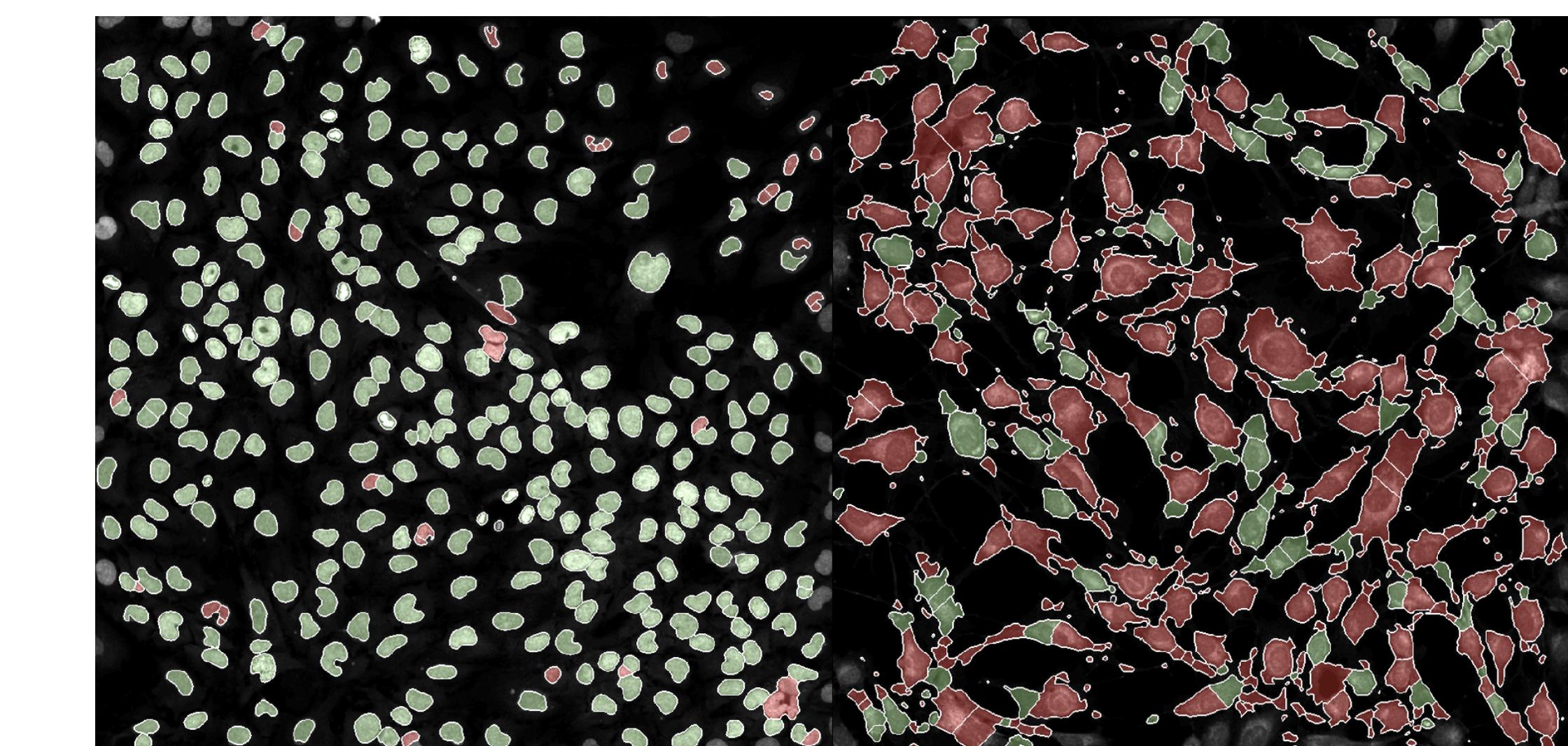


Figure 6: Single-cell segmentations were evaluated with coSMicQC, identifying which passed (green) or failed (red) quality control (QC) criteria. The left panel showcases field-of-view (FOV) images displaying nuclei from a more standard phenotype while the right panel shows nuclei from a sample with an unusual phenotype. These results illustrate how coSMicQC effectively distinguishes between high- and low-quality segmentations, aiding in the accurate identification of outliers and ensuring the reliability of downstream analysis for complex biological datasets.

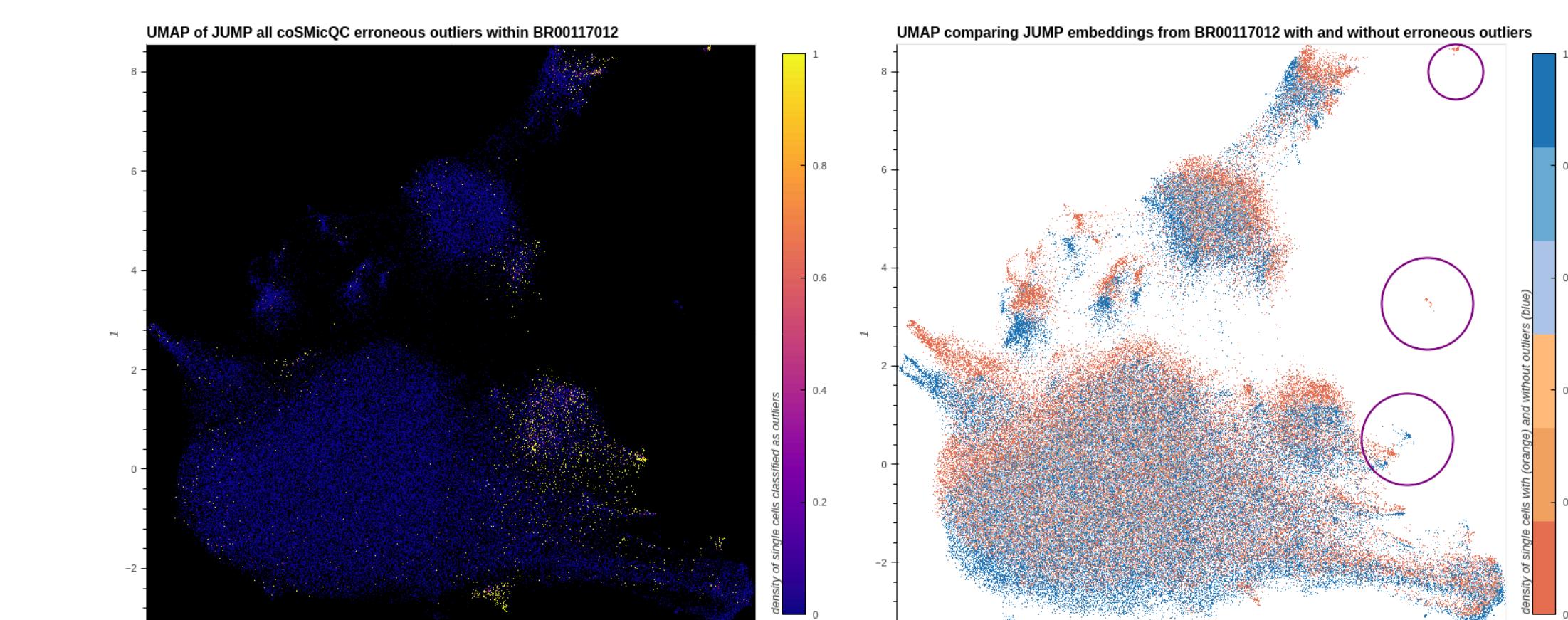


Figure 7: Applying coSMicQC with the JUMP dataset BR00117012 (cpg0000) reveals erroneous outliers, which are highlighted in yellow in the left panel. These outliers significantly impact the UMAP embeddings by altering the spatial distribution of data points. Specifically, the presence of outliers causes shifts in cluster locations or even their removal from the embeddings. In the right panel, orange points represent UMAP embeddings that include these outliers, while blue points denote embeddings generated after removing outliers. This comparison demonstrates the crucial role of coSMicQC in identifying and mitigating outliers, thereby ensuring more accurate and meaningful clustering in UMAP visualizations.

V. Acknowledgements

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