BioSET - Biomarker-based Spatial co-Expression analysis in Tumor environments

Chahat Kalsi[1]* Yuancheng Shen[1] Sophia Gaupp[2] Luca Reichmann[2]
Meri Rogava[5] Michael Krone[4] Saeed Boorboor[3] Robert Krüger[1],
[1]New York University [2]University of Tübingen [3]University of Illinois Chicago
[4]Stuttgart University of Applied Sciences [5]Zucker School of Medicine at Hofstra/Northwell

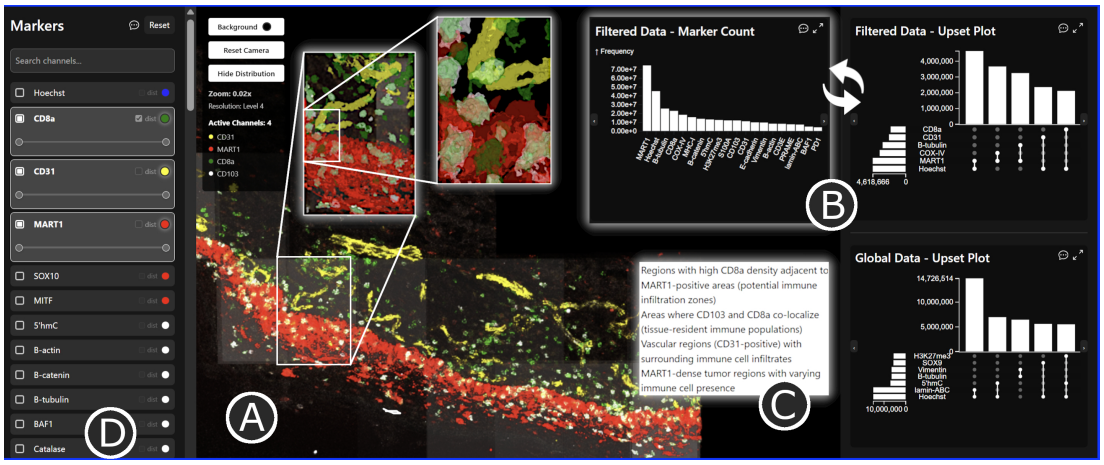

Figure 1: **BioSET. (A)** Multi-volume rendering of selected protein channels with distance-adaptive resolution. T-cells (CD8a, green), are migrating out of blood vessels (CD31, yellow) to infiltrate and fight melanoma cells (MART1, red); a tile-based heatmap (gray) guides users to regions with high co-location of these protein markers. Hierarchical regional selections reveal details on cellular scale with combined mesh+volume rendering: Presence of CD103 (white) suggests that some t-cells are 'tissue-resident', rather than entering from the bloodstream. **(B)** UpSet plots summarize frequencies of most intersecting channel sets both globally and for the selected markers. Plots can also be toggled to individual channel-wise counts. **(C)** Biomni AI agent provides textual interpretation. **(D)** Selection of channels and visualization parameters.

1 Introduction

This abstract reports on our solution to the IEEE VIS 2025 Bio+MedVis 3D Microscopy Imaging Challenge with the tasks of identification and visualization of cell-cell interactions in 3D Cyclic Immunoflorescence (CyCIF) data. The provided 3D melanoma tissue data [11] comprises 70 channels (volumes) across 6 levels of resolution, the highest having $194\times5{,}508\times10{,}908$ voxels per channel (1.48 TB) and the lowest having $194\times172\times340$ voxels per channel (1.48 GB).

For this dataset, the challenge defined **three tasks**: **(T1)** Detect ROIs containing user-specified bio-marker combinations; **(T2)** Visualize these ROIs globally to assess extent and tissue context; **(T3)** Enable cellular-level drill-down within selected ROIs.

Few (mostly proprietary) existing tools such as Imaris [5] can visualize multi-volumetric data at scale, and being visual display-focused, their spatial analysis capabilities are limited to manual exploration. While integrated analytics approaches, e.g., [10], have been proposed for 2D multiplexed imaging data, methods that handle volumetric datasets are rare. Most related to our work is Cell2Cell by Mörth *et al.* [6], a novel segmentation-free approach that assumes equally sized spherical cells and is evaluated on small volumes of resolution $55\times1{,}024\times1{,}024$. By contrast, our approach scales to much larger data and is able to account for diverse cell shapes.

In summary, our approach makes the following **contributions**: **(1)** a method to detect protein-expressing regions and identify spatially co-located marker sets in the tissue. **(2)** A scalable overview-detail visualization, combining multi-scale volume and surface rendering, to guide users to regions of interest, and **(3)** integrated contextual querying, providing semantic explanations of displayed patterns. We analyzed the data using our tool in joint sessions with a biomedical expert (co-author) from *Northwell Health, New York*; the findings reported in the abstract and the supplemental materials stem from these sessions, aligning with manual analyses by experts [11].

2 Approach

Identification of Marker Sets For **T1**, we use a set-based method to detect regions of marker co-expression. We use the iso-value selection method of Pekar *et al.* [7] to find out which voxels capture the markers expressed in each channel. Since the detected iso-value τ is more accurate in a local context than globally, we process the volume in $194\times1377\times909$ tiles at the finest resolution level. We then threshold each tile per channel at $I \geq \tau$ to obtain masks that approximate marker-expressing voxels (Fig. 2 **(i)**).

Subsequently, we compute the intersections between channels. Voxels that are marked as 'ON' in the binary mask at the same spatial location across tile masks corresponding to each channel are being co-expressed. Lastly, we perform these calculations for finer-grained tiles of resolution $194\times459\times303$ and use the results to visualize the *hotspots* in Fig. 4 and in the drill-down view visible in Fig. 1 **(A)**.

Hybrid Multi-Volume and Surface Rendering To provide overview, we visualize the data globally (**T2**, Fig 1 **(A)**). Given the

*corresponding (first) author: Chahat Kalsi, chahat.kalsi@nyu.edu — Second authors Shen, Gaupp, and Reichmann contributed equally.

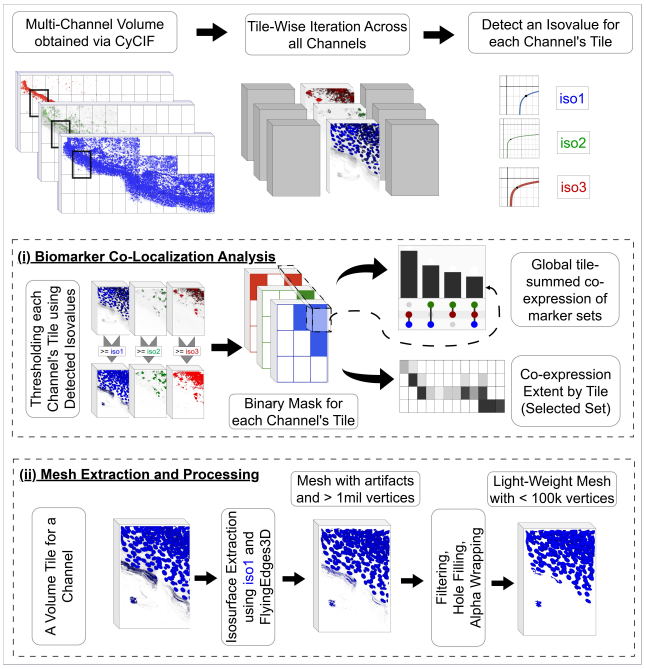

Figure 2: **Processing and Analysis Pipeline.** We iterate the multi-channel volume tile-by-tile and estimate a per-channel iso-value for each tile. We use this iso-value for **(i)** Biomarker Co-Location Analysis and **(ii)** Mesh Extraction and Processing.

6 resolution levels in the dataset, we built a web-based multi-scale volume renderer based on VTK.js [8], capable of switching between resolutions based on camera distance from the volume center. In practice, it can render resolution levels 4-6 and support up to seven channels simultaneously. To explore the data interactively, users can search and select channels of interest from a marker list (Fig. 1 **(D)**), assign them colors and adjust their intensity ranges. Additionally, a preselected interaction section below allows selection of marker groups whose spatial overlap indicates a specific biological context (e.g., immune cells).

While volume rendering is sufficient for global views, it can be difficult to demarcate the boundaries of expressed markers on a cellular level (**T3**). This is due to the often diffuse protein marker distribution. To achieve a clearer view of regions with expressed markers, we employ surface rendering. To derive the surfaces (Fig. 2 **(ii)**), we first extract per-channel meshes for each of the $194\times1377\times909$ tiles using the *FlyingEdges3D* [9] algorithm with the previously detected iso-values τ. We then perform a series of processing steps (Fig. 2 **(ii)**) on the raw meshes obtained through this. We, **(i)** drop connected components below a face-count threshold, **(ii)** fill small holes using mean component diameter and face count statistics [2], **(iii)** apply *alpha-wrapping* with parameters derived from the mean component diameter [1], and **(iv)** remove residual noisy components by face count. The result is a clean, low-polygon mesh per tile and channel for interactive, cell-level inspection.

Guidance to Regions of Interest To visualize which sets of markers frequently spatially overlap, we integrate two interactive UpSet plots [4], as shown in Fig. 1 **(B)**. In Fig. 1 **(B)**, the global plot displays the overall marker abundances and co-localizations across the whole dataset, while the filtered plot is updated dynamically based on the current marker selection. The UpSet plots (Fig. 1 **(B)**) and the viewer **(A)** are linked: selecting markers updates the view, while current channel selections in the viewer highlight the corresponding sets in the filtered UpSet plot. As the plot can get cluttered as the number of channels grows, we provide an additional filtering feature to only include markers of interest in the plot.

To guide users to regions with prominent interactions (**T2**), we overlay the volume with a tile-level co-expression heatmap: each $194\times1137\times909$ tile is colored by the count of co-expressing voxels for the selected group (white = high; black = low with reduced opacity), revealing global ROIs. We use grayscale to avoid interfering with channel colors. Selecting a tile opens a drill-down with combined volume+mesh rendering and a sub-tile heatmap at $194\times459\times303$, guiding users to cell-scale structures (**T3**). As a coarse heatmap can miss small interaction *hotspots*, we also compute ROIs at the finer $194\times459\times303$ tile-size and visualize them as circles scaled by co-expression extent in the main view (Fig. 4).

Contextual Queries for Biomedical Explanations To aid the intepretation process, we integrate semantic textual explanations using Biomni [3]. Biomni is an AI agent designed to perform domain-specific reasoning on biomedical data. Explanation buttons send targeted queries to Biomni together with their relevant context. The output (Fig. 1 **(C)**) summarizes what is visible, explains its biological relevance, suggests ROIs, and offers an assessment of possible clinical relevance. This is valuable for cases where the significance of markers and their combinations is not immediately apparent, particularly to non-specialists.

3 Discussion and Future Work

Our solution draws on a hierarchical subdivision (tiling) of the multi-volume data to identify co-located protein markers and a combined volume and surface rendering from overview to detail. Current limitations thus lie in the artificial spatial binning and the level of detail. In the future, we aim to leverage cell segmentation to identify spatial patterns on a cellular basis and to integrate steerable deep-learning for scalable feature-based ROI classification and search.

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