Recurrence-Associated Spatial Niches and Tumor–Stroma Interface Architecture in Colorectal Cancer

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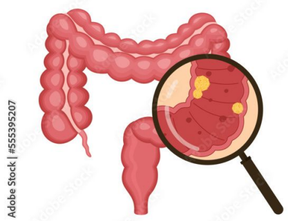
Introduction

Problem:

- CRC: 4th most common cancer, 2nd leading cause of cancer death worldwide
- Up to 30% of Stage II patients recur despite complete resection
- TNM staging & Immunoscore rely on bulk biomarker expression/broad T-cell counts

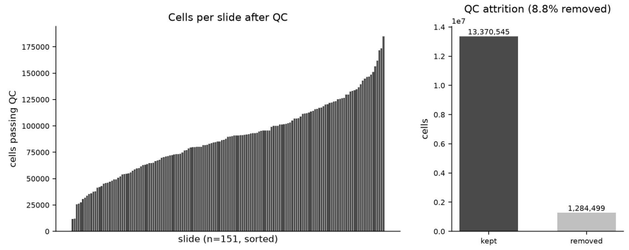


Project Goals

1. Identify recurrence-associated immune-exclusion niches using spatial transcriptomics. (Eesha)
2. Determine whether recurrence-associated niches preferentially occur in collagen-remodeled microenvironments. (Further work with partner)

Hypothesis: Recurrent CRC samples contain spatial cellular neighborhoods that differ systematically from non-recurrent CRC samples.

Cohort & QC



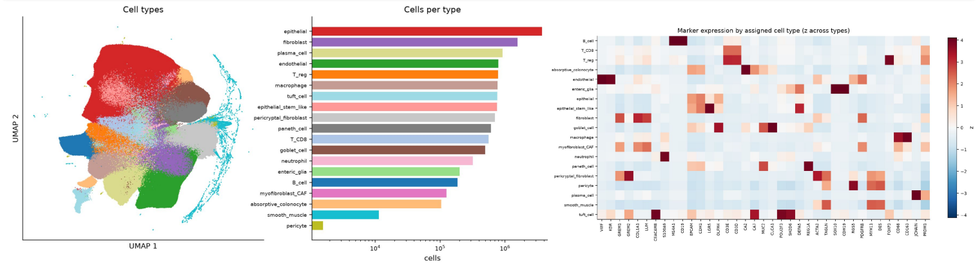
126 patients, 151 slides, 13,370,545 cells
318 targets & 46 controls, 24 recurrences (19.0%)
8.8% of cells removed during QC; no slides excluded

Workflow

QC	EMBED	TYPE	SPATIAL	NICHES
metadata join noise floor + QC	22 PCs Harmony on source	Leiden 1.0 19 cell types	tumor boundary 3 primary endpoints	CellCharter exploratory K

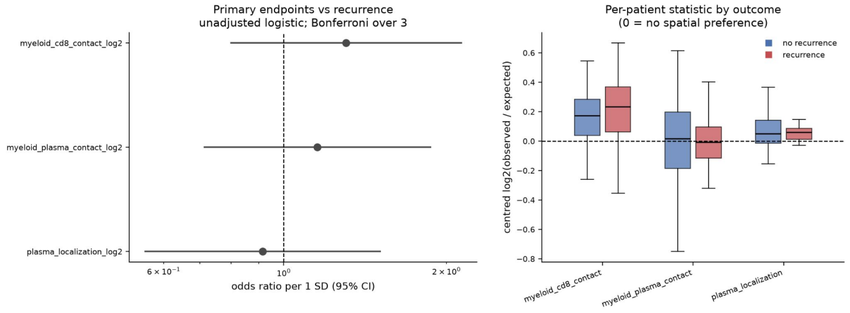
Patient-level unadjusted logistic regression; Bonferroni for the 3 prespecified endpoints, BH within exploratory families.

Cell Typing



19 cell types across 38 Leiden clusters
Marker-set competition included an expression floor plus guards for epithelial, NK/CD8, Treg/CD4, CAF/smooth muscle, and neutrophil/monocyte overlap.

Primary Endpoints

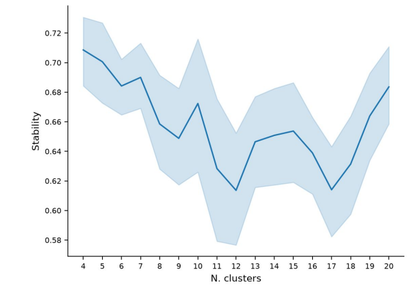


Myeloid–CD8 contact	Myeloid–plasma contact	IFN-high plasma localization
OR 1.305	**OR 1.155**	**OR 0.914**
95% CI 0.799–2.130 Bonf. p = 0.862	95% CI 0.714–1.867 Bonf. p = 1.000	95% CI 0.555–1.507 Bonf. p = 1.000

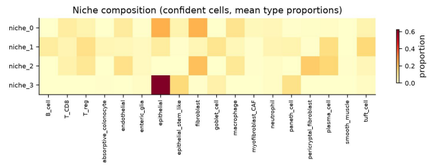
All confidence intervals crossed 1.0.

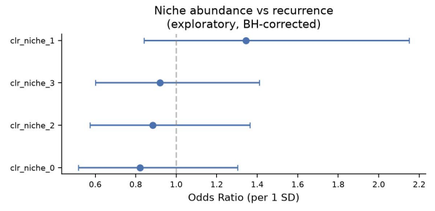
Exploratory niche discovery

CellCharter reused the Harmony embedding, built a per-slide Delaunay graph, aggregated 3 neighbor layers (~31 µm), and selected K on a stratified 500k-cell subsample.



K = 4 was the stability maximum (~0.71); K = 10 was a secondary peak (~0.67).





No niche-recurrence association survived BH correction

Key Findings and Limitations

- myeloid–CD8 contact and IFN-high plasma localization were enriched across the cohort, but not differentially associated with recurrence after the prespecified correction.
- tumor-proximal myeloid–CD8 contact was nominally associated (OR 1.726, p=0.012); this was not prespecified and is descriptive.

Limitations:

unadjusted models, classifier transfer to Cedars not independently validated, fragmented interface band, and no spillover correction.

edit.