



2nd BOLOGNA WORKSHOP

on IMMUNOTHERAPY FOR ACUTE MYELOID LEUKEMIA

Bologna, Royal Hotel Carlton
September 29-30, 2026

AML in space: The Spatial Organisation of the AML Bone Marrow

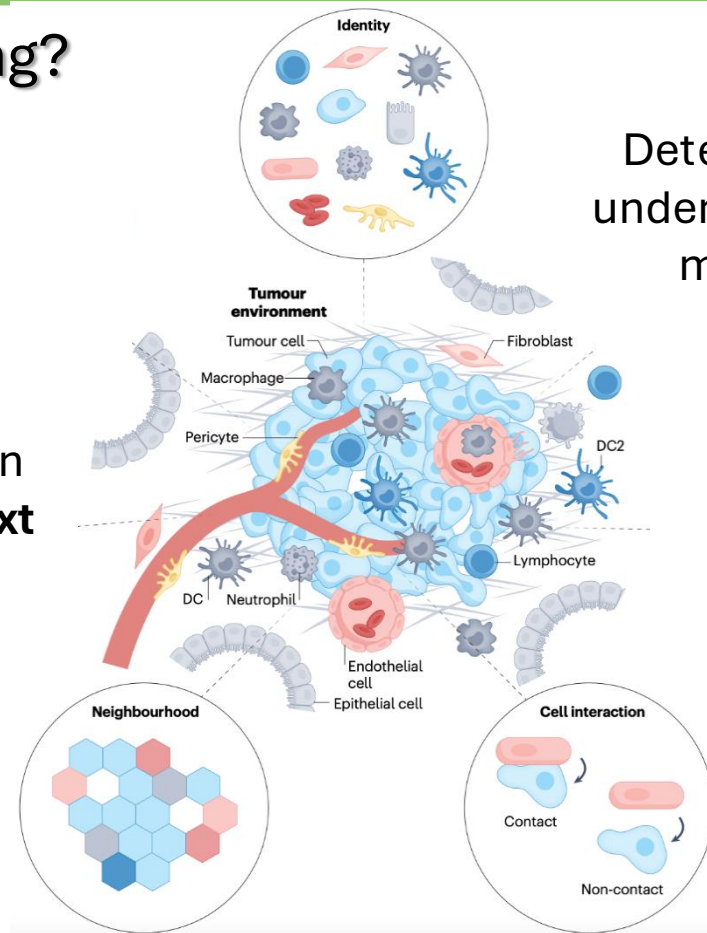
Olaf Heidenreich

Disclosures Olaf Heidenreich

[illegible]

Why Spatial Profiling?

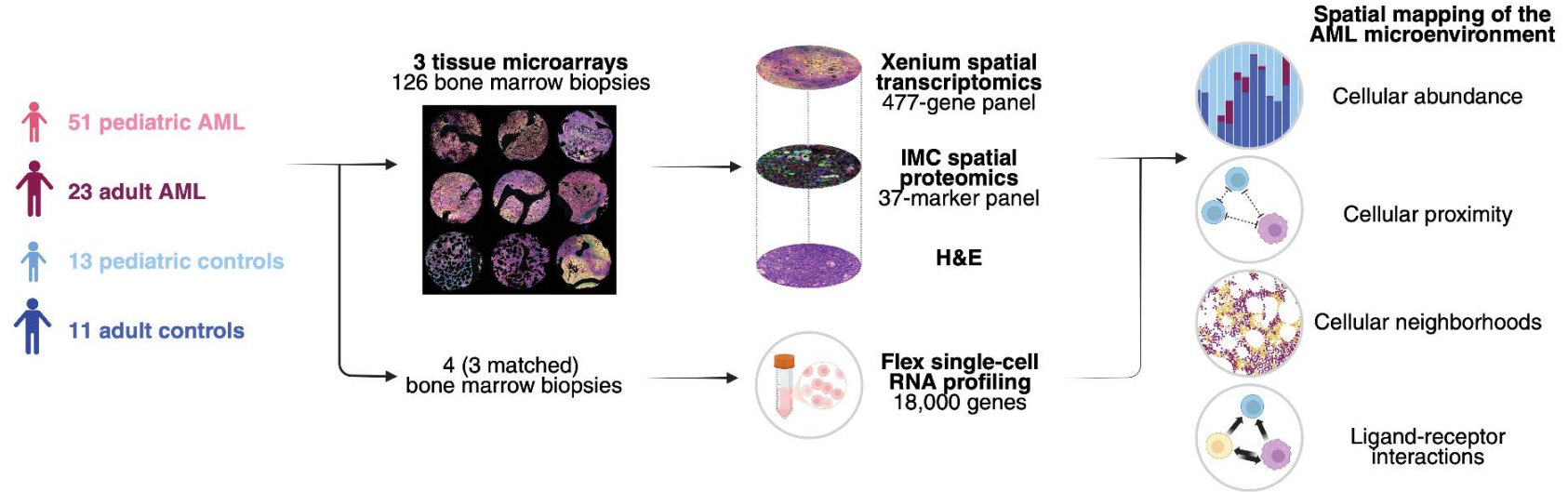
Immune
microenvironment in
native tissue context



Detection of **cell types**
underrepresented in bone
marrow aspirates

Improved **cellular**
interaction analysis

Study Set-up



Joost Koedijk

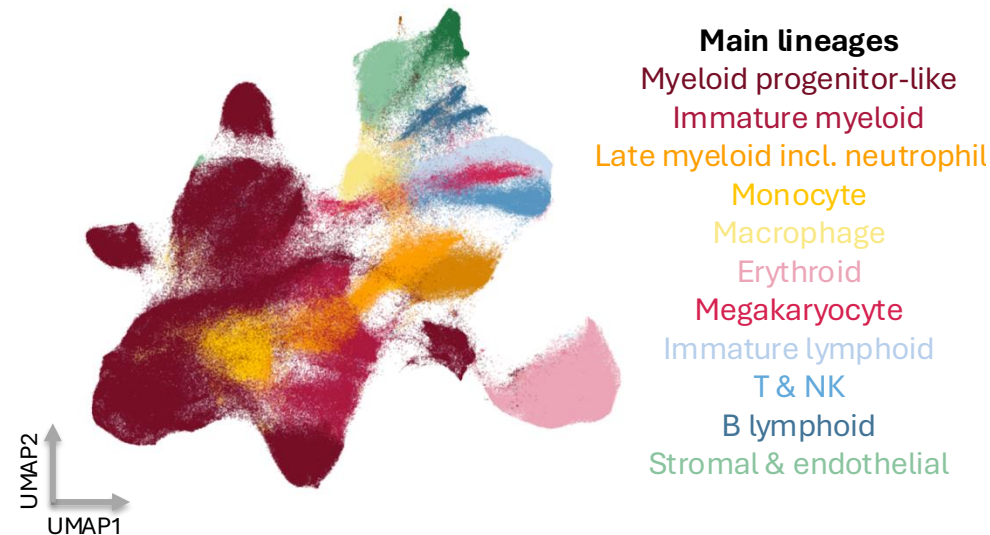


Merel van der Meulen



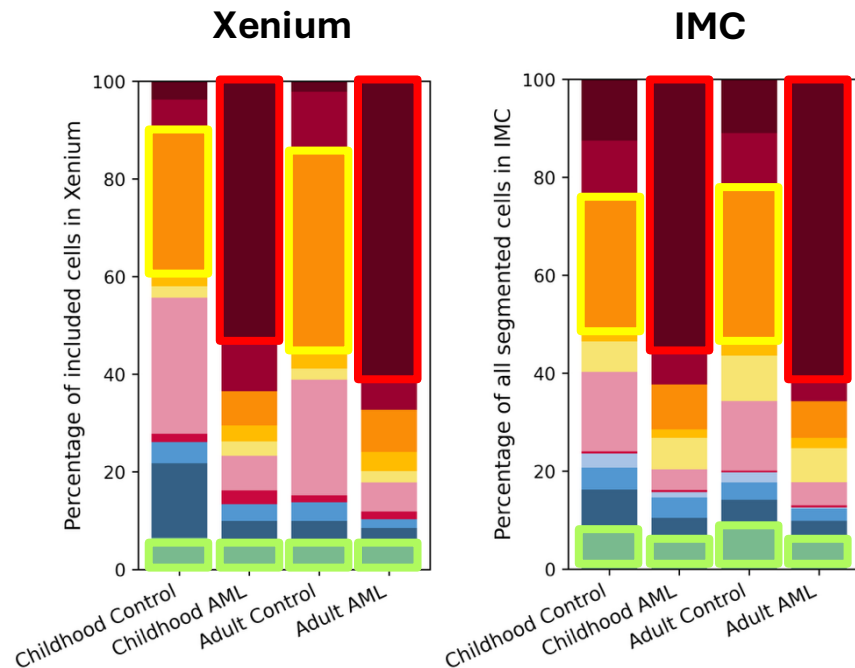
Emma Pool

Distribution of Cell Types Across Modalities

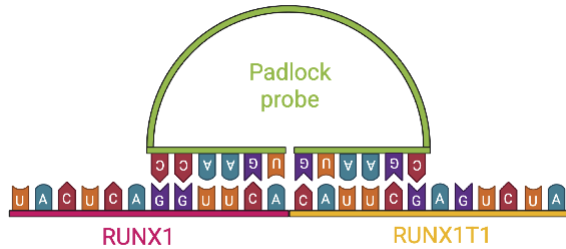


Annotation:

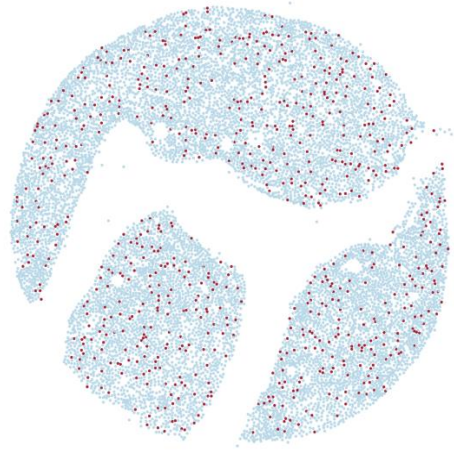
1. Marker genes/proteins
2. scRNA-seq references



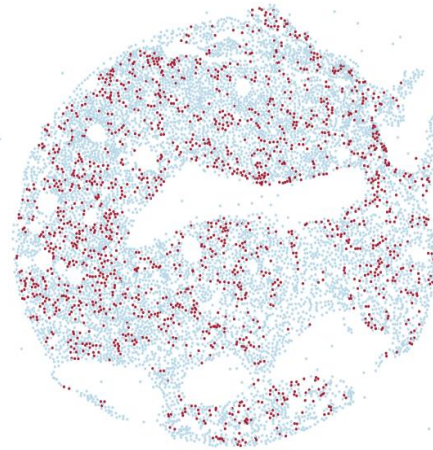
Detection of Fusion Transcript-positive Cells



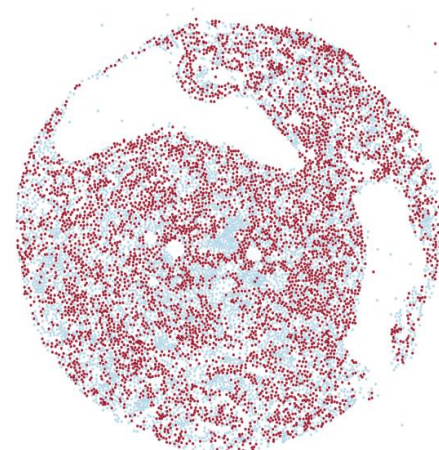
CA28
(15% blasts)



CA49
(35% blasts)

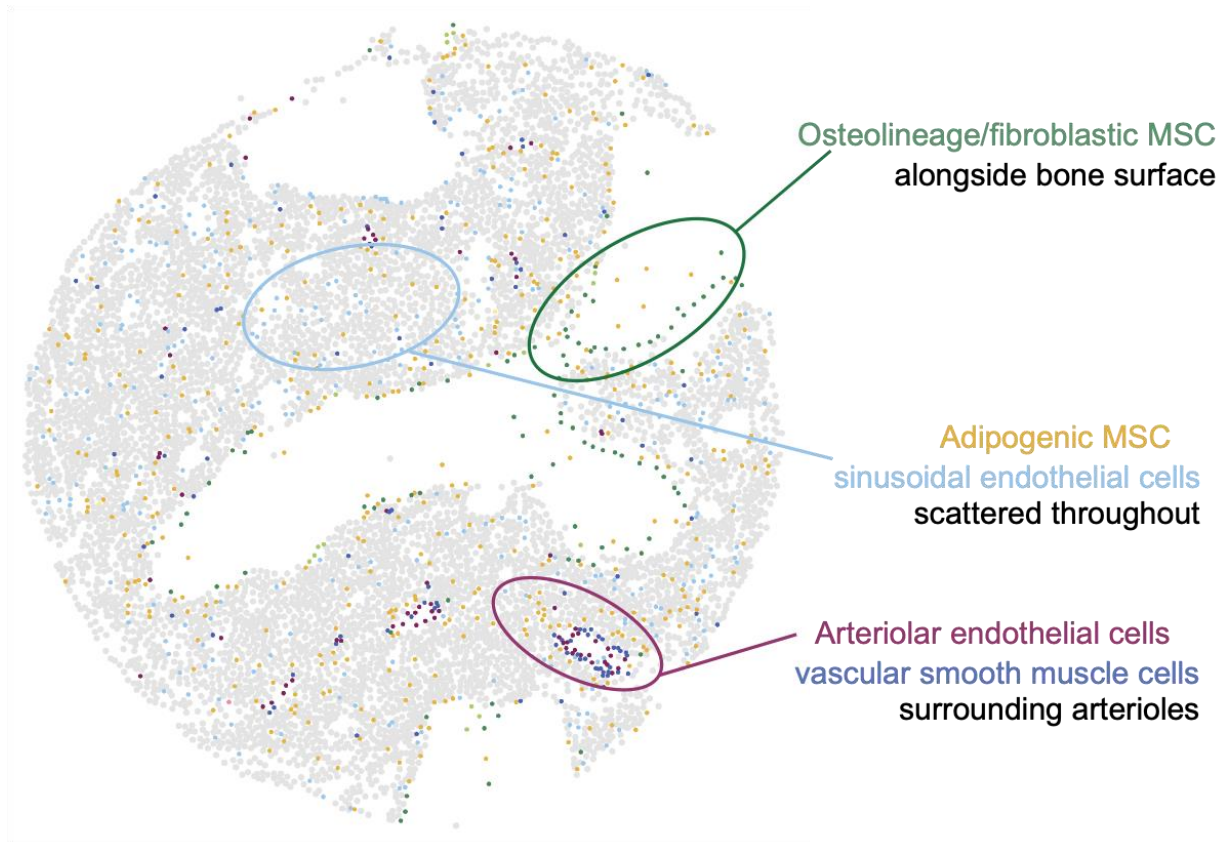


CA51
(96% blasts)



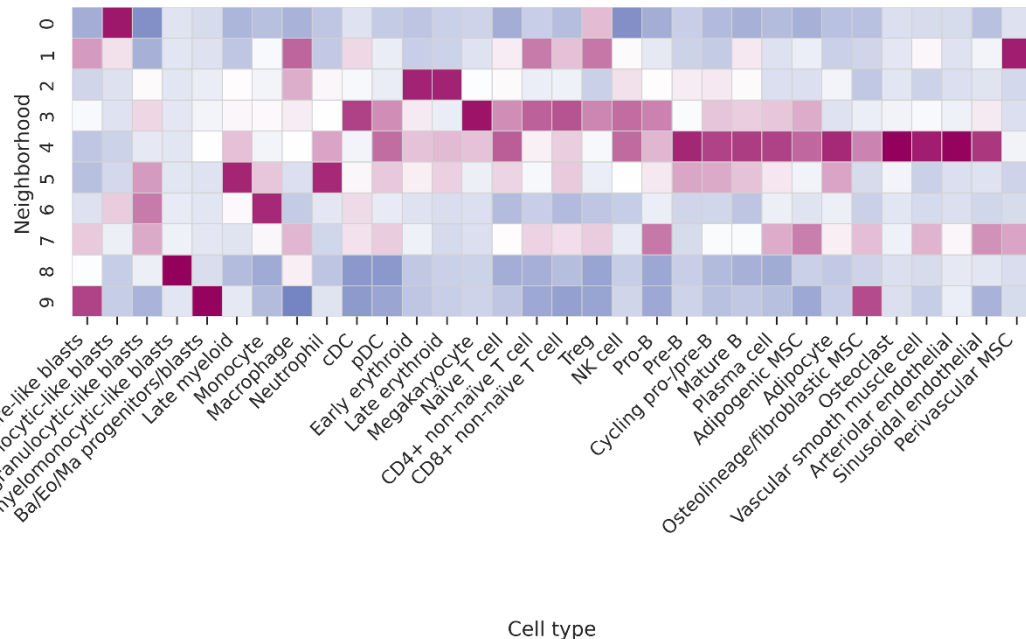
Fusion status (any cell type): Fusion+ / Fusion-

Localisation of Stromal Cells



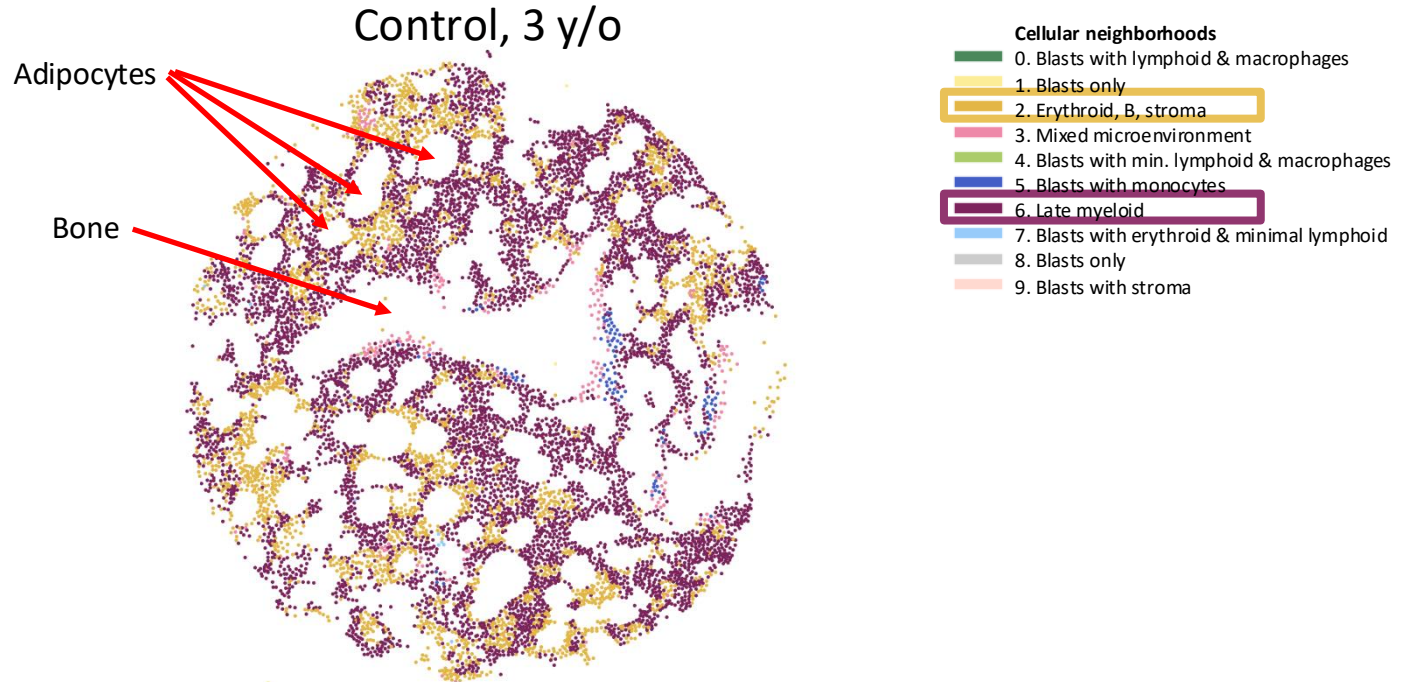
Defining Cellular Neighbourhoods

Z-score of cell type enrichment per neighborhood

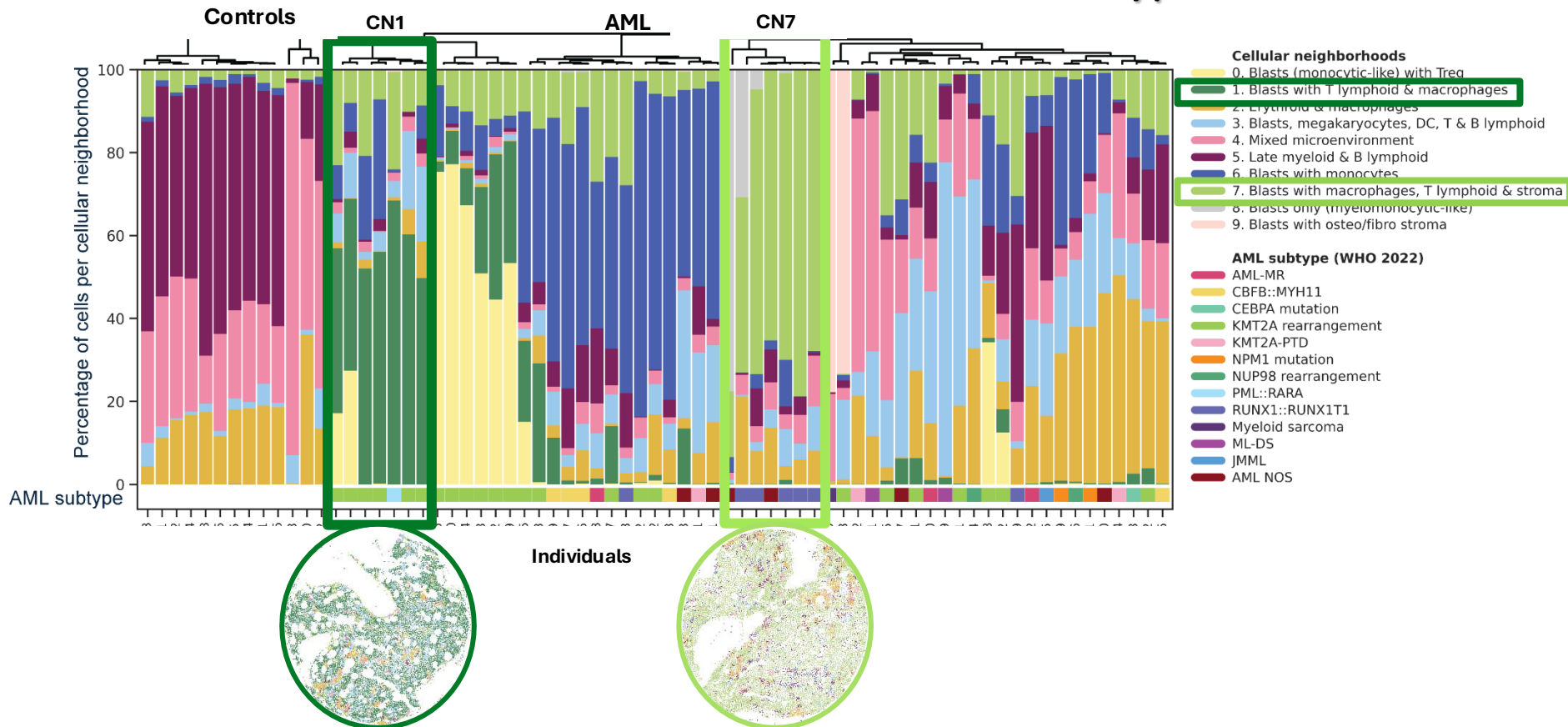


- CN0. Blasts with lymphoid & macrophages
- CN1. Blasts only
- CN2. Erythroid, B, stroma
- CN3. Mixed microenvironment
- CN4. Blasts with with minimal lymphoid & macrophages
- CN5. Blasts with monocytes
- CN6. Late myeloid
- CN7. Blasts with erythroid & minimal lymphoid
- CN8. Blasts only
- CN9. Blasts with stroma

Cellular Neighbourhoods



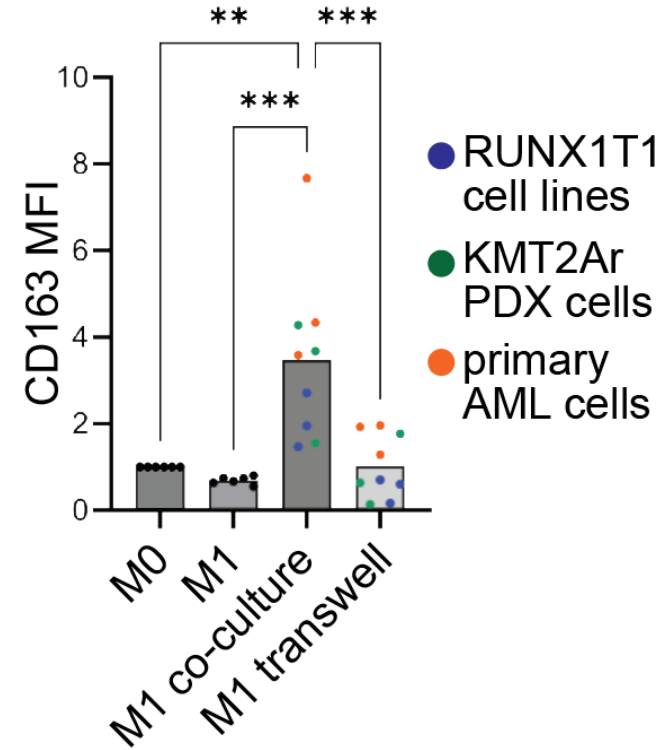
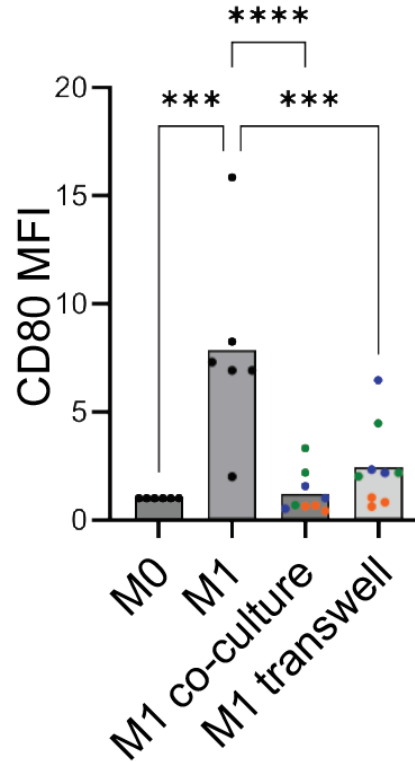
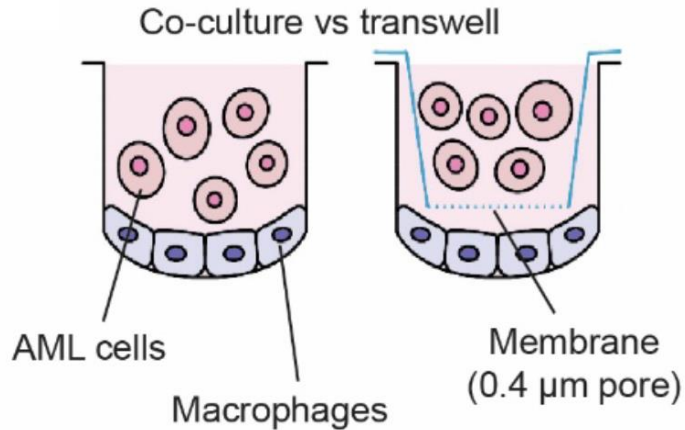
Cellular Neighbourhoods Correlate with AML subtype



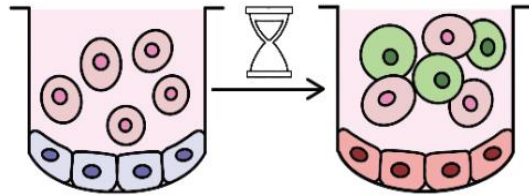
AML Cells Induce Immunosuppressive Macrophages



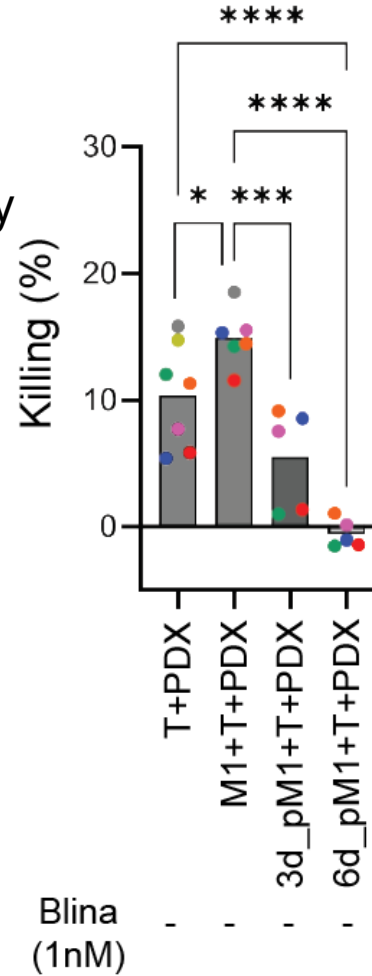
Alicia Perzoli



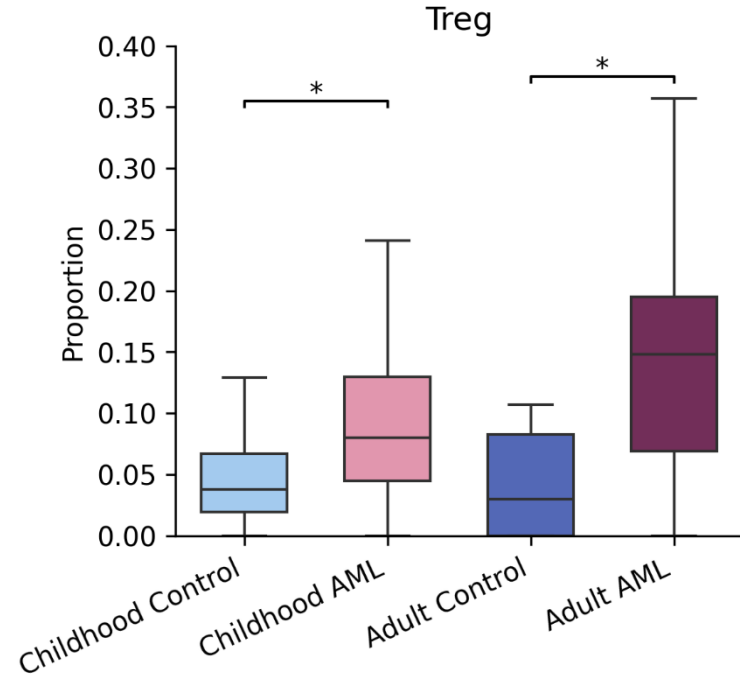
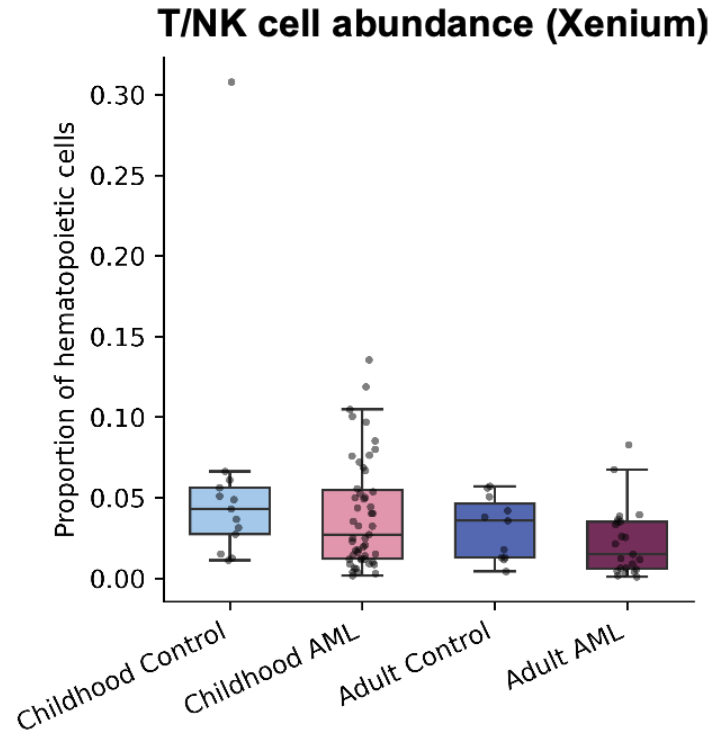
Primed macrophage killing assay



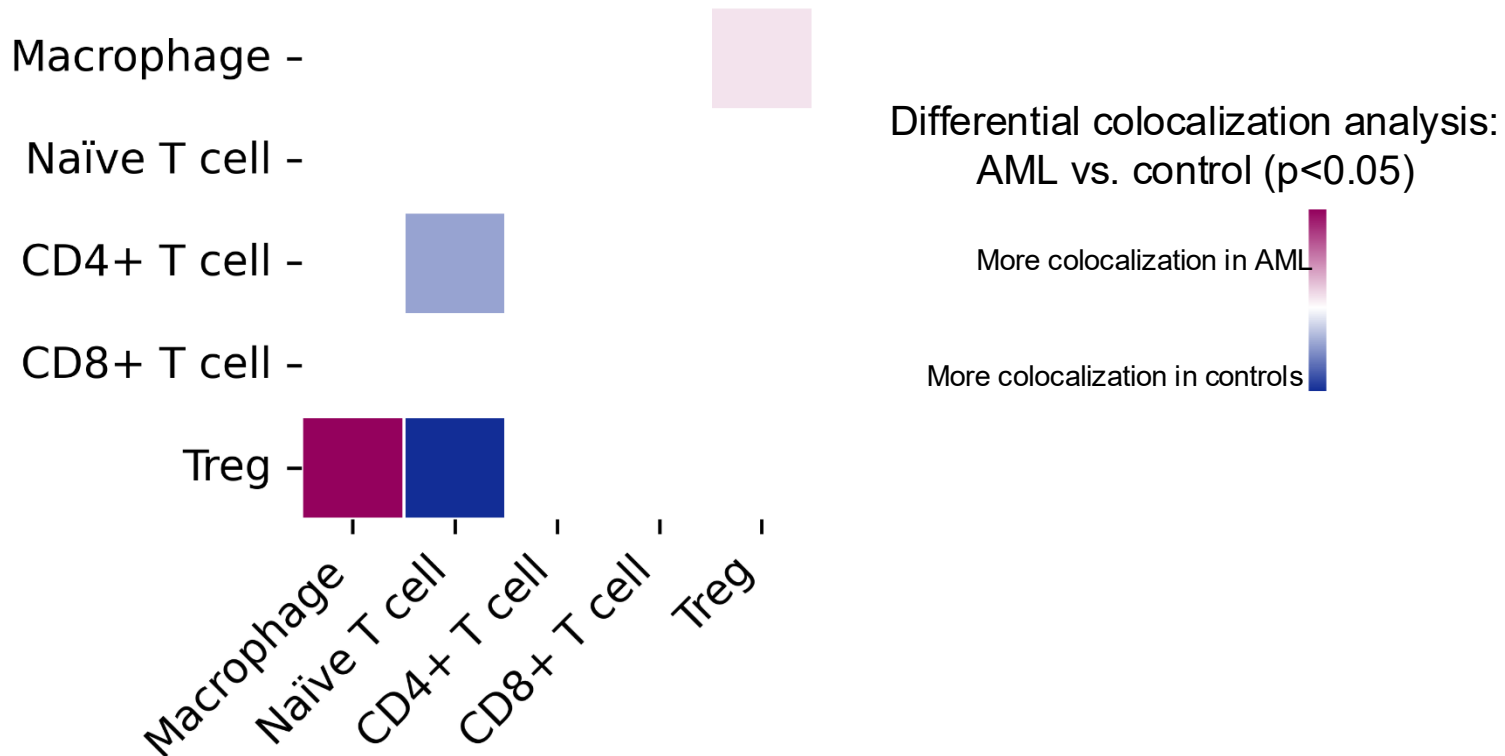
- AML cells
- T cells
- Macrophages
- Primed macrophages



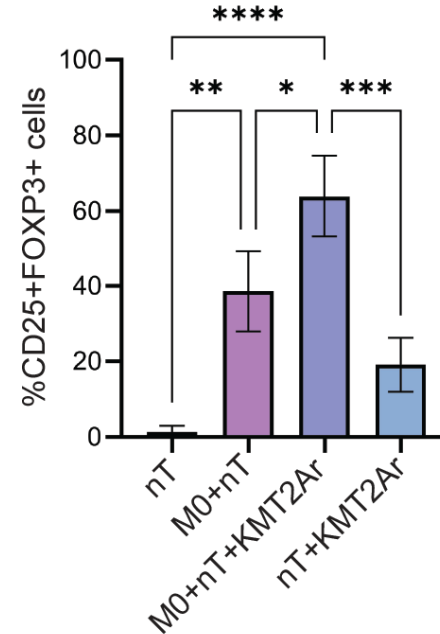
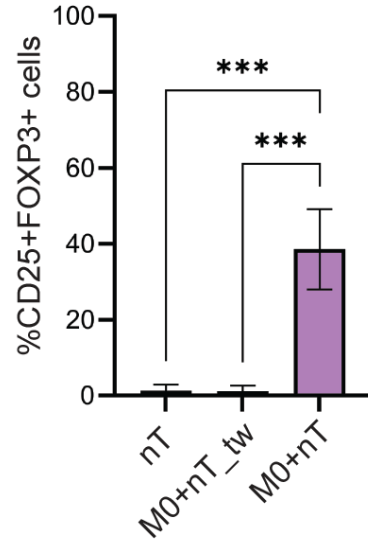
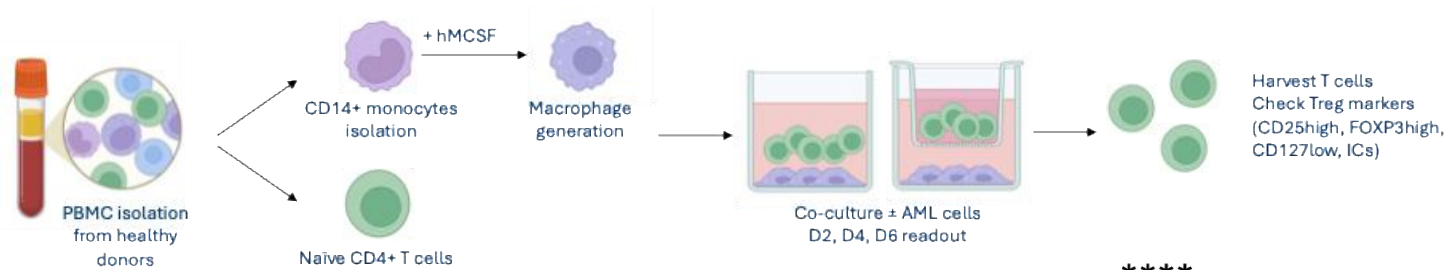
Treg Cells Are More Abundant in AML Bone Marrow



Treg Cells Interact with Macrophages in the AML BM

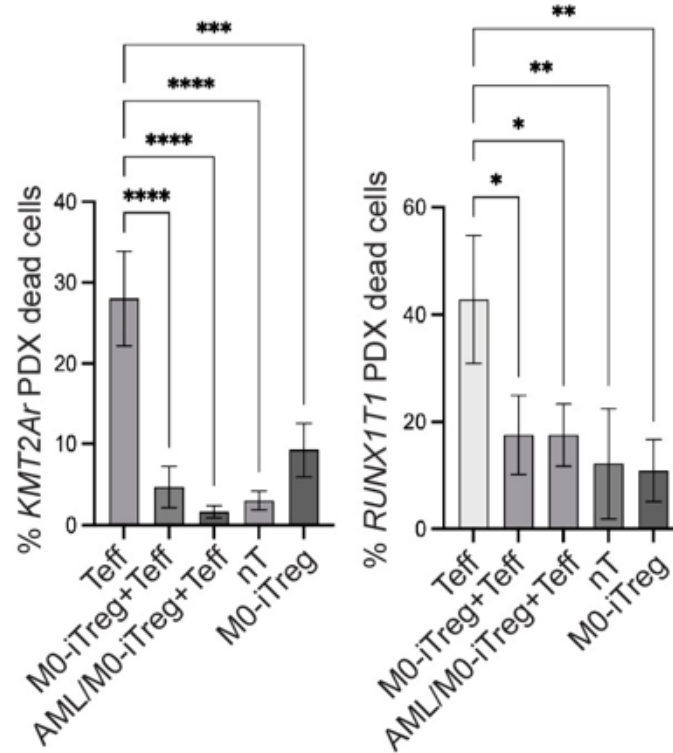
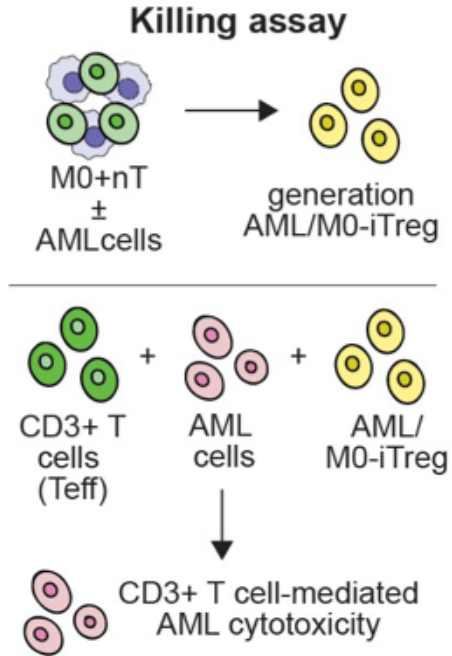


Macrophages induce Treg phenotype *in vitro*



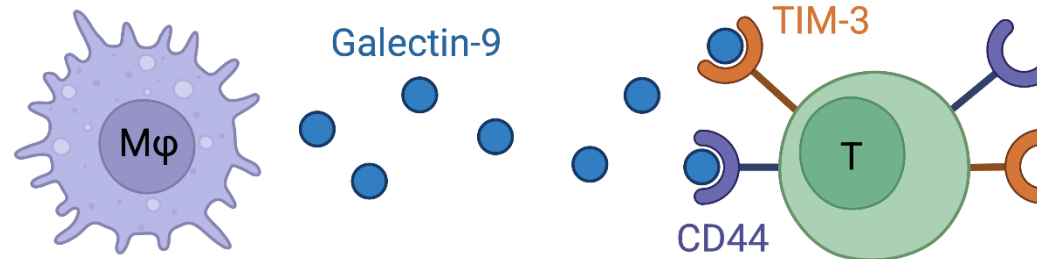
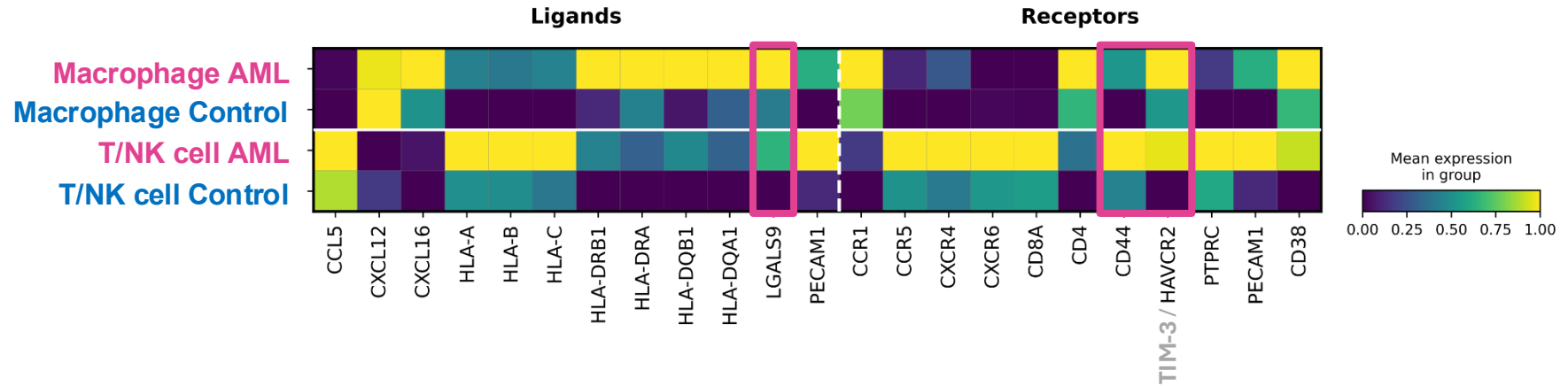
Legend: nT = naïve T cells; M0_tw = co-culture with naïve macrophages in transwell setting; KMT2Ar = KMT2A-rearranged patient-derived xenograft

Induced Treg Cells inhibit T Cell-mediated AML Killing

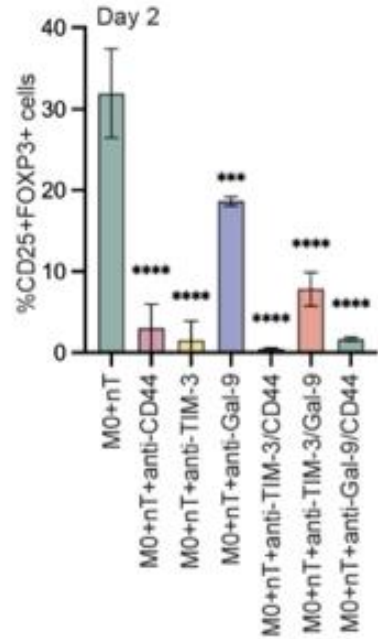


Legend: nT = naïve T cells; M0_{tw} = co-culture with naïve macrophages in transwell setting; KMT2Ar = KMT2A-rearranged patient-derived xenograft

Ligand-Receptor Interactions between Macrophages and T cells



Blocking TIM-3, CD44 and GALS9 Prevents Treg Induction

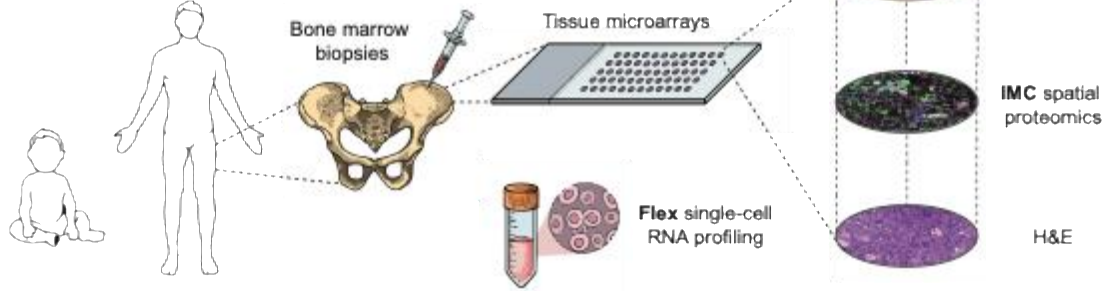


Summary

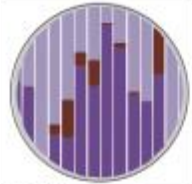
- Single cell spatial proteo-transcriptomic atlas of AML at diagnosis.
- Detection of fusion transcripts permits analysis of AML cells and their descendants.
- AML cells generate an immunosuppressive microenvironment by repolarizing macrophages and inducing Treg cells.
- Blocking TIM3, CD44 or GALS9 impairs Treg induction.

1. Data collection

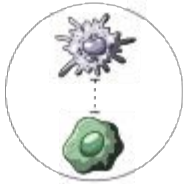
Adult/pediatric AML and non-leukemic controls



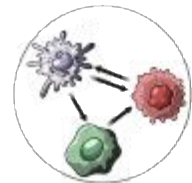
2. Spatial mapping of AML microenvironment: colocalization between macrophages and Tregs



Cellular abundance



Cellular proximity

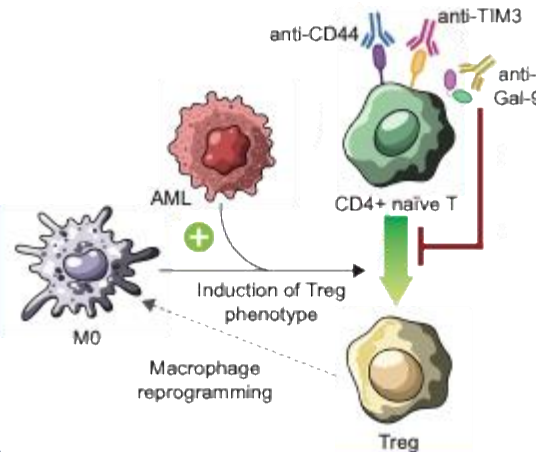


Cellular interaction



Cellular neighborhoods

3. Co-culture system: macrophages and AML induce Tregs



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All patients and their families

