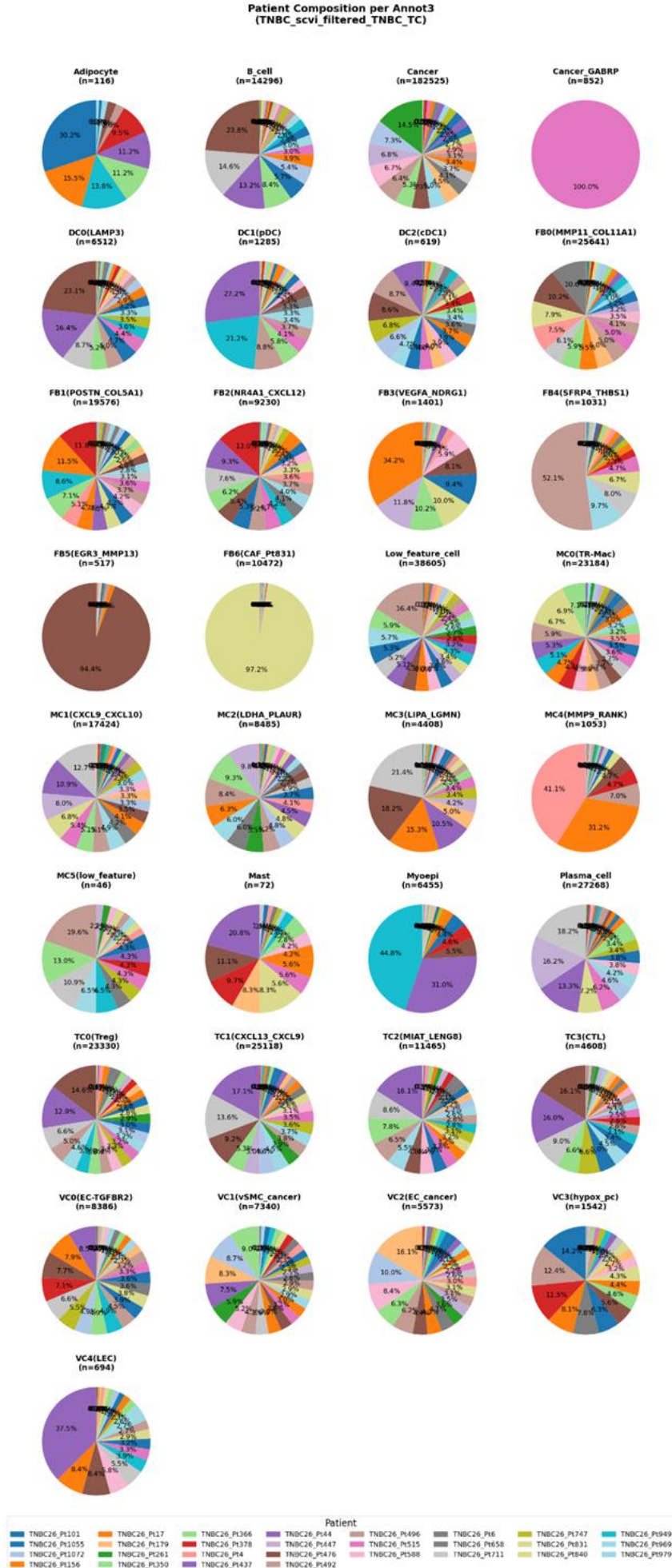
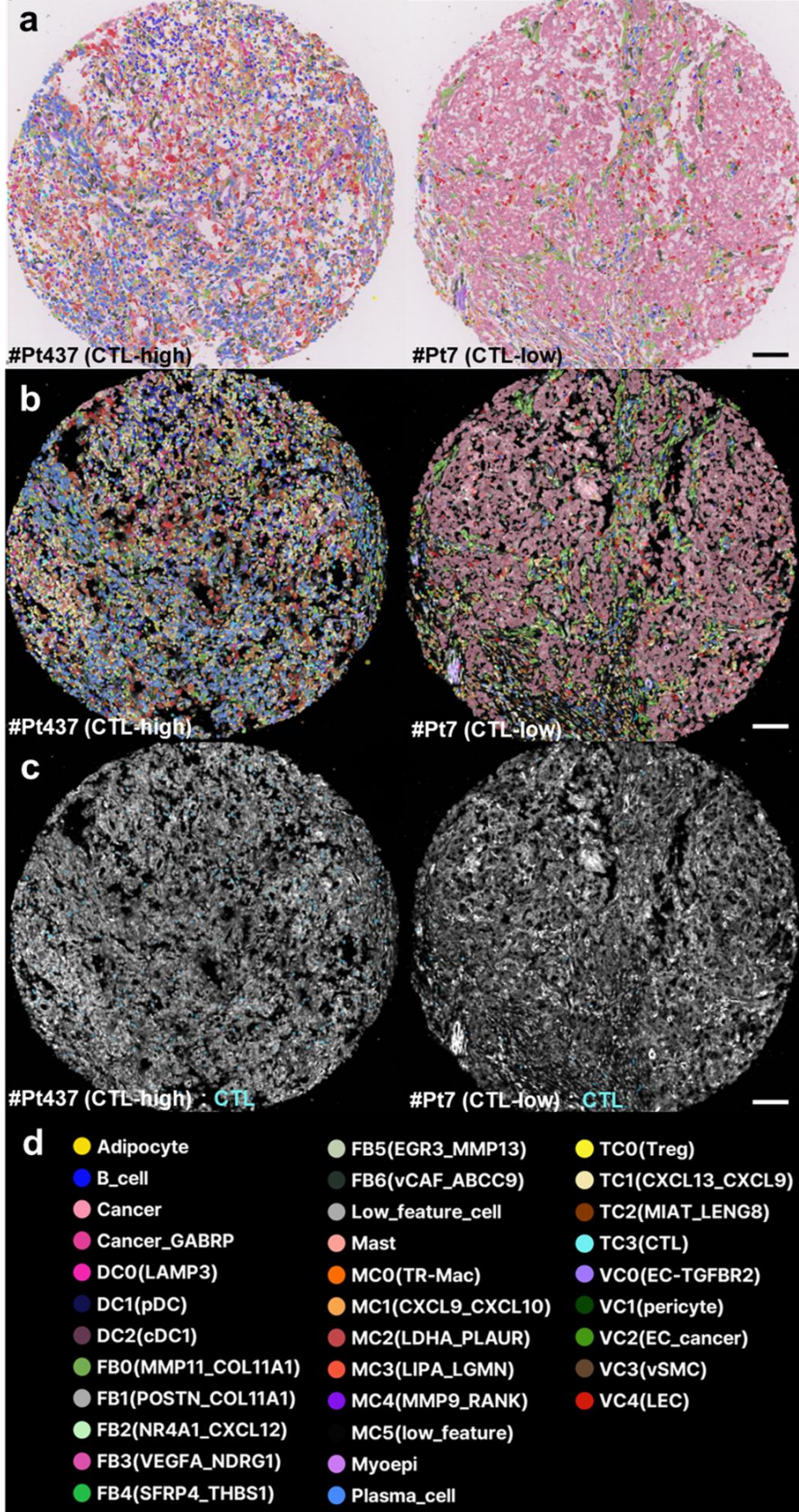


a



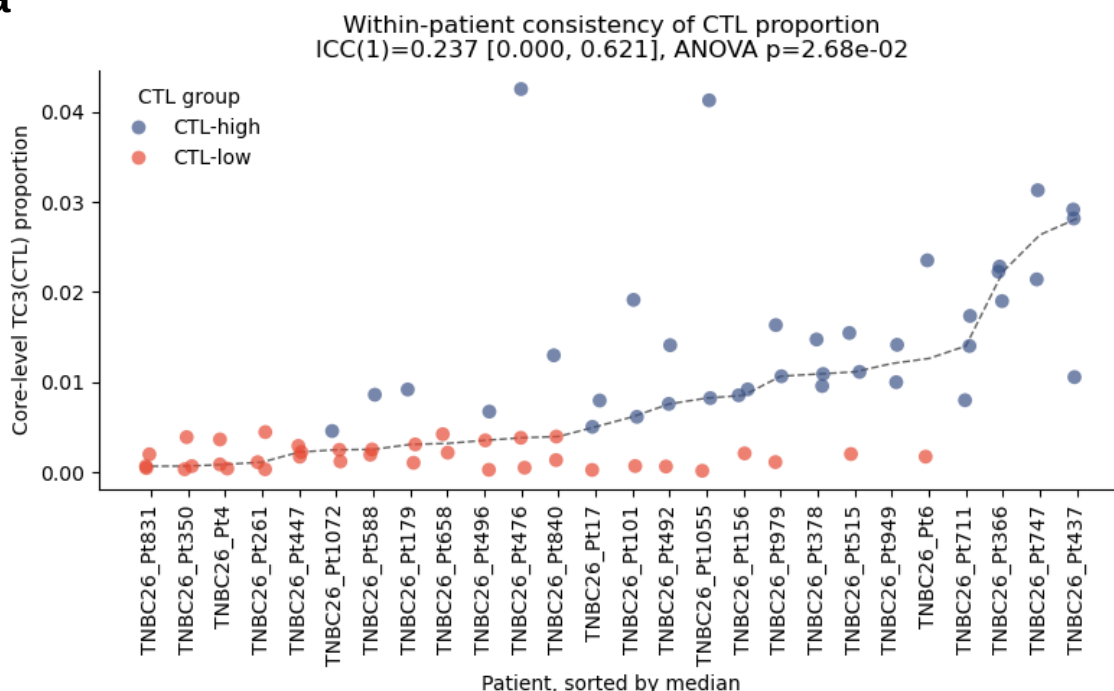


Supplementary Figure 3. Spatial distribution of cell types and CTL infiltration in CTL-high vs CTL-low tumors.

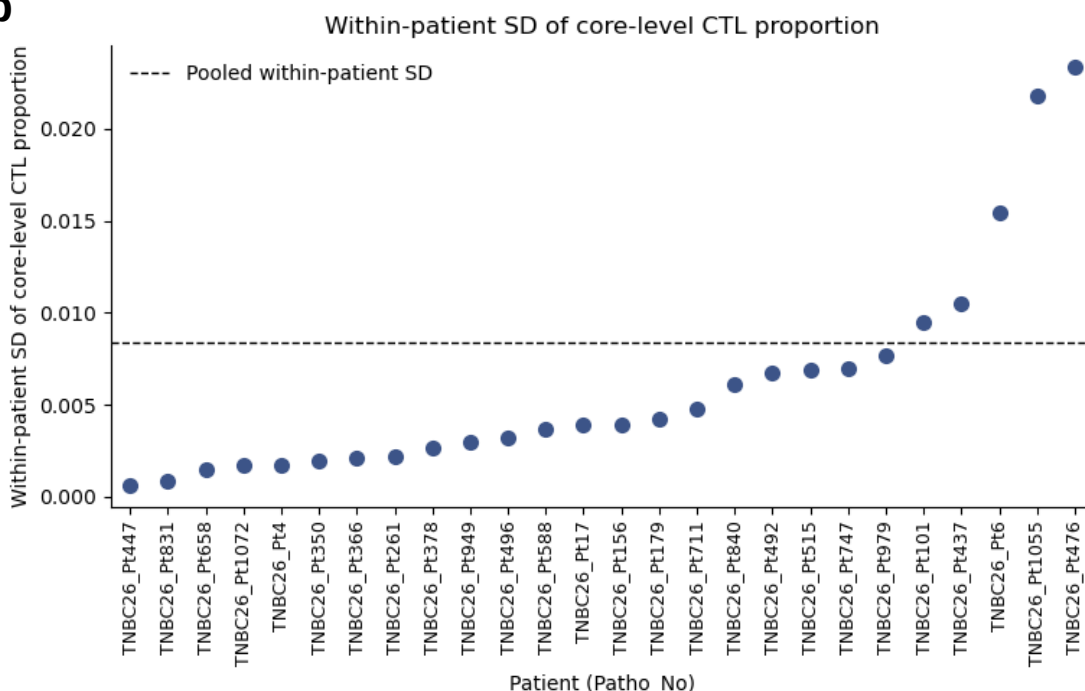
- (a) Representative H&E and Xenium images across CTL groups, comparing a CTL-high core (#Pt437) and a CTL-low core (#Pt7). H&E images overlaid with cell-type annotation colors.
- (b) All cell segmentation channel images overlaid with the same cell-type annotation colors.
- (c) All other cell types shown in grayscale. Same as (b), with only TC3(CTL) cells highlighted (cyan).
- (d) Color legend for cell-type annotations.

Xenium images in (a)-(c) were generated using 10x Genomics Xenium Explorer (v4.1.1). Scale bars, 100 μ m.

a

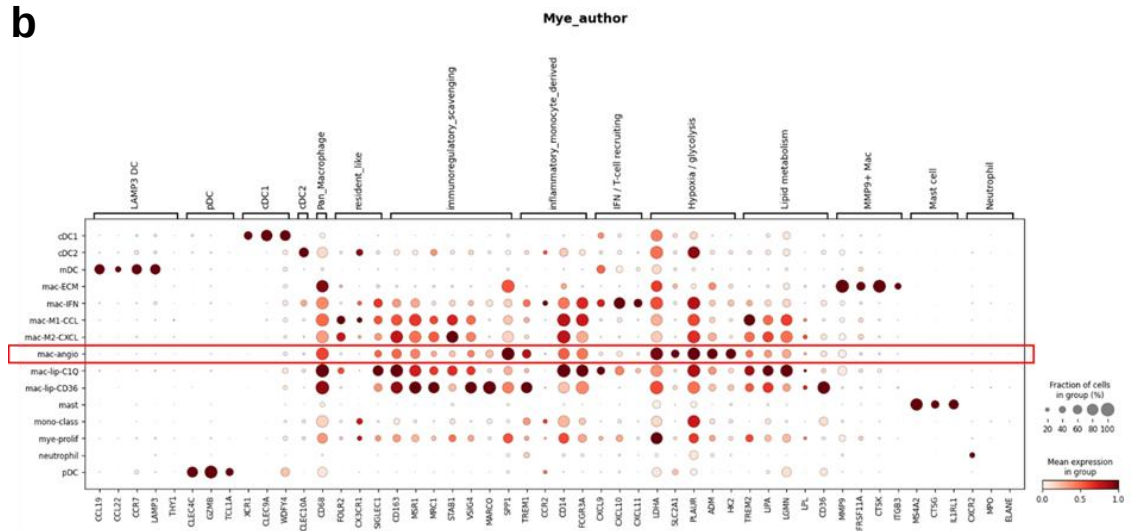
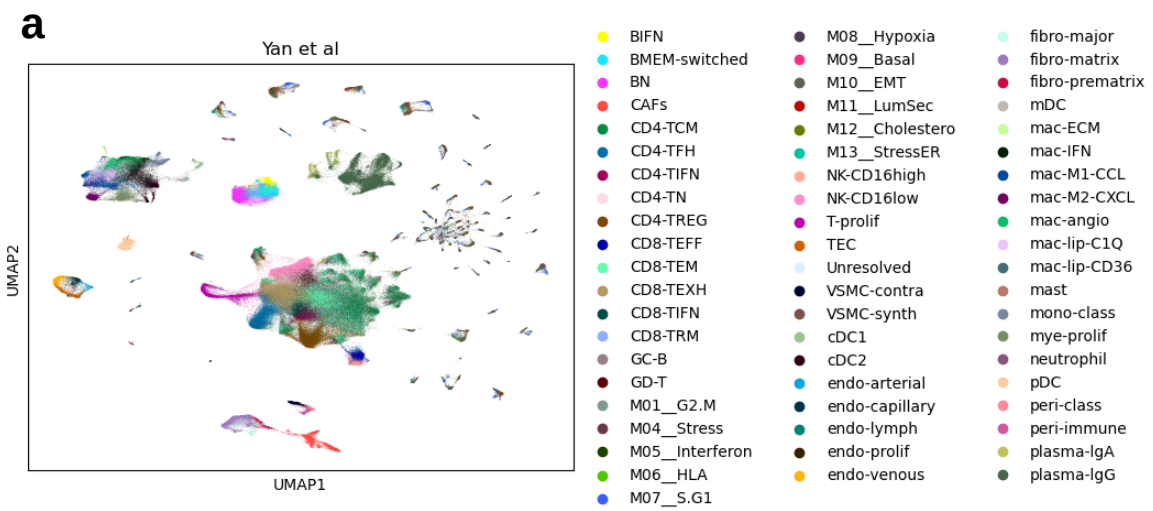


b

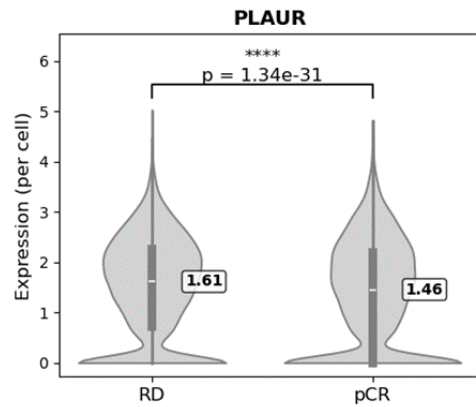


Supplementary Figure 4. Within-patient consistency of core-level TC3(CTL) proportion.

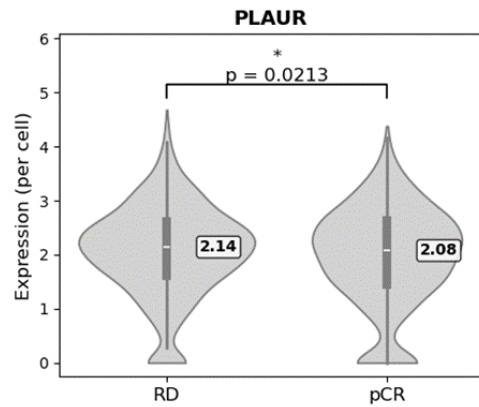
- (a) CTL proportion per core across patients (sorted by median), colored by CTL group (CTL-high/CTL-low); dashed line, per-patient median. ICC(1)=0.237, 95% CI [0.000, 0.621] (patient-level bootstrap, n_boot=1000); ANOVA $p=2.68 \times 10^{-2}$.
- (b) Within-patient SD of core-level CTL proportion per patient, sorted ascending; dashed line, pooled within-patient SD (0.0084).



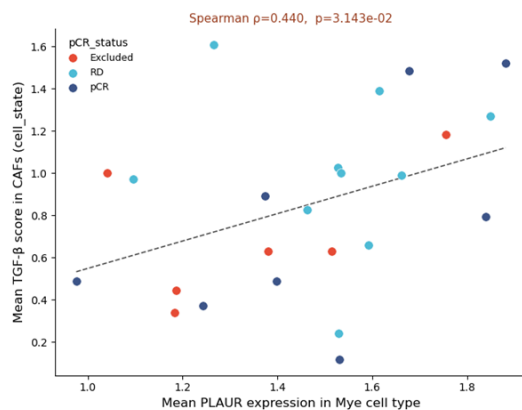
c Mye_author (cell-level)



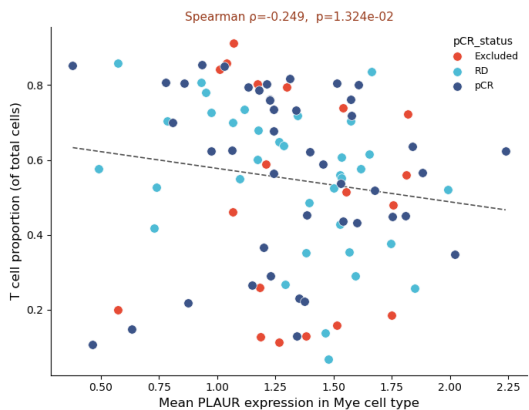
d mac_ang (cell-level)



e Mye PLAUR expression vs CAFs TGF- β score (donor-level)



f Mye PLAUR expression vs T cell proportion (donor-level)



Supplementary Figure 5. PLAUR⁺ myeloid cells associate with residual disease and immune exclusion in an independent cohort.

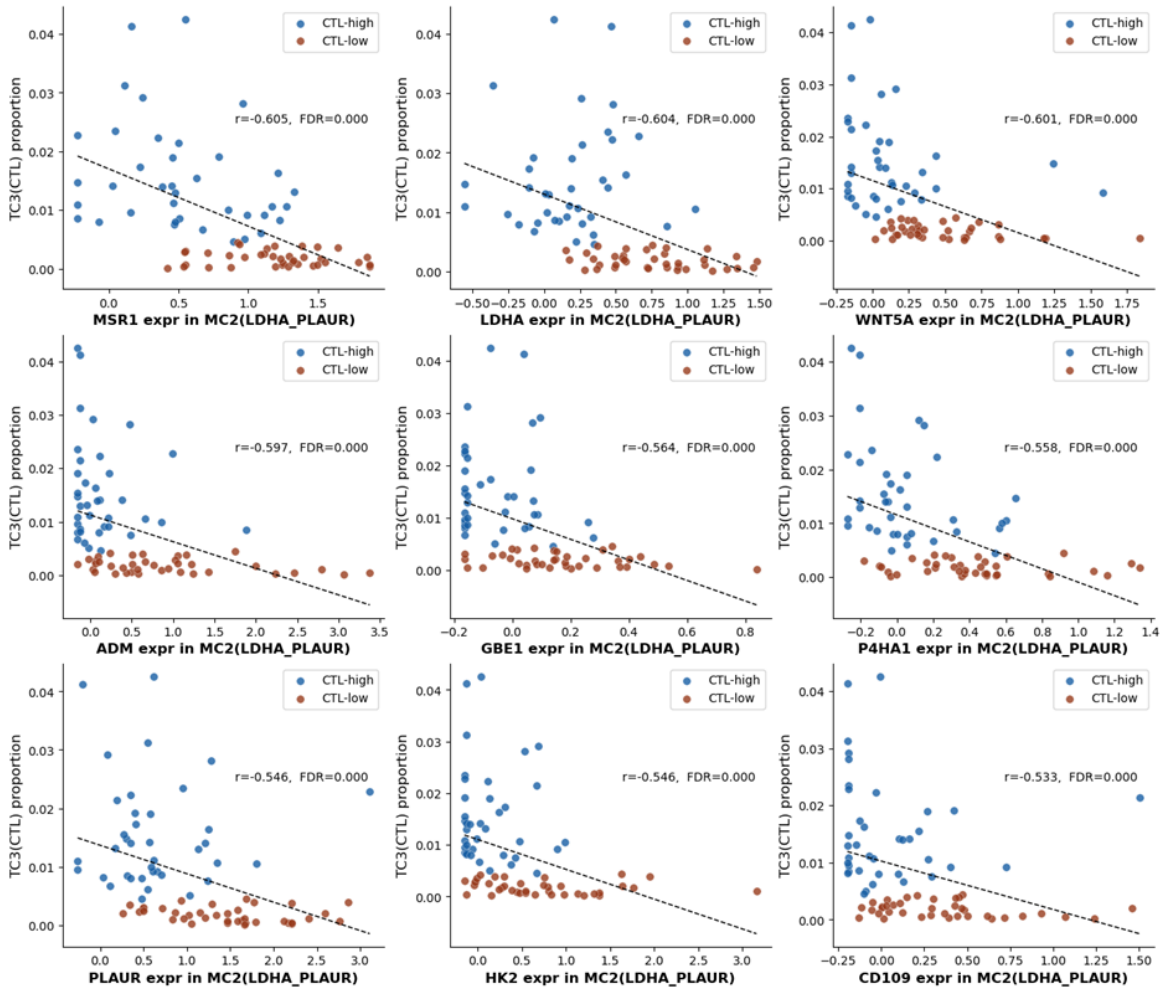
(a, b) UMAP of all cell states (a) and dot plot of hypoxia/glycolysis marker expression across myeloid cell states (b) from an independent scRNA-seq cohort (Yan et al.). Red box, mac-angio, showing marker expression (LDHA, PLAUR, SLC2A1) concordant with MC2(LDHA_PLAUR).

(c, d) Violin plots of mean PLAUR expression per cell between residual disease (RD) and pathological complete response (pCR) groups, in all myeloid cells (c; $p=1.34 \times 10^{-31}$) and in the mac-angio subtype specifically (d; $p=0.0213$).

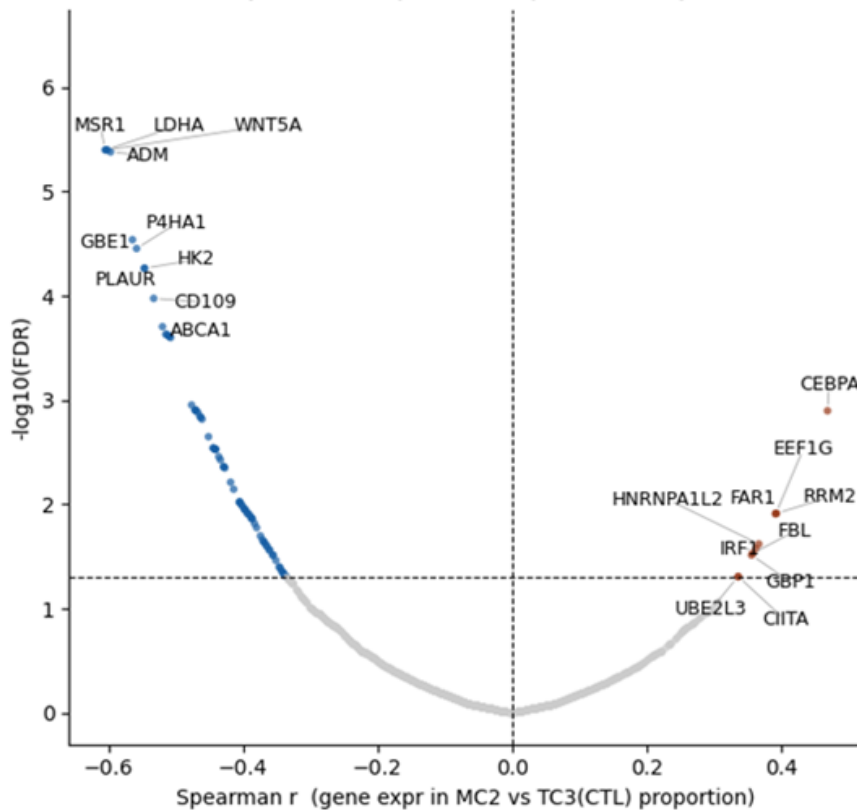
(e, f) Donor-level Spearman correlation between mean PLAUR expression in myeloid cells and TGF- β downstream signaling score in CAFs (e; $\rho=0.440$, $p=0.031$) or T-cell proportion (f; $\rho=-0.249$, $p=0.013$).

a

Top genes in MC2(LDHA_PLAUR) vs TC3(CTL) infiltration
(Whole niche, Core-level)

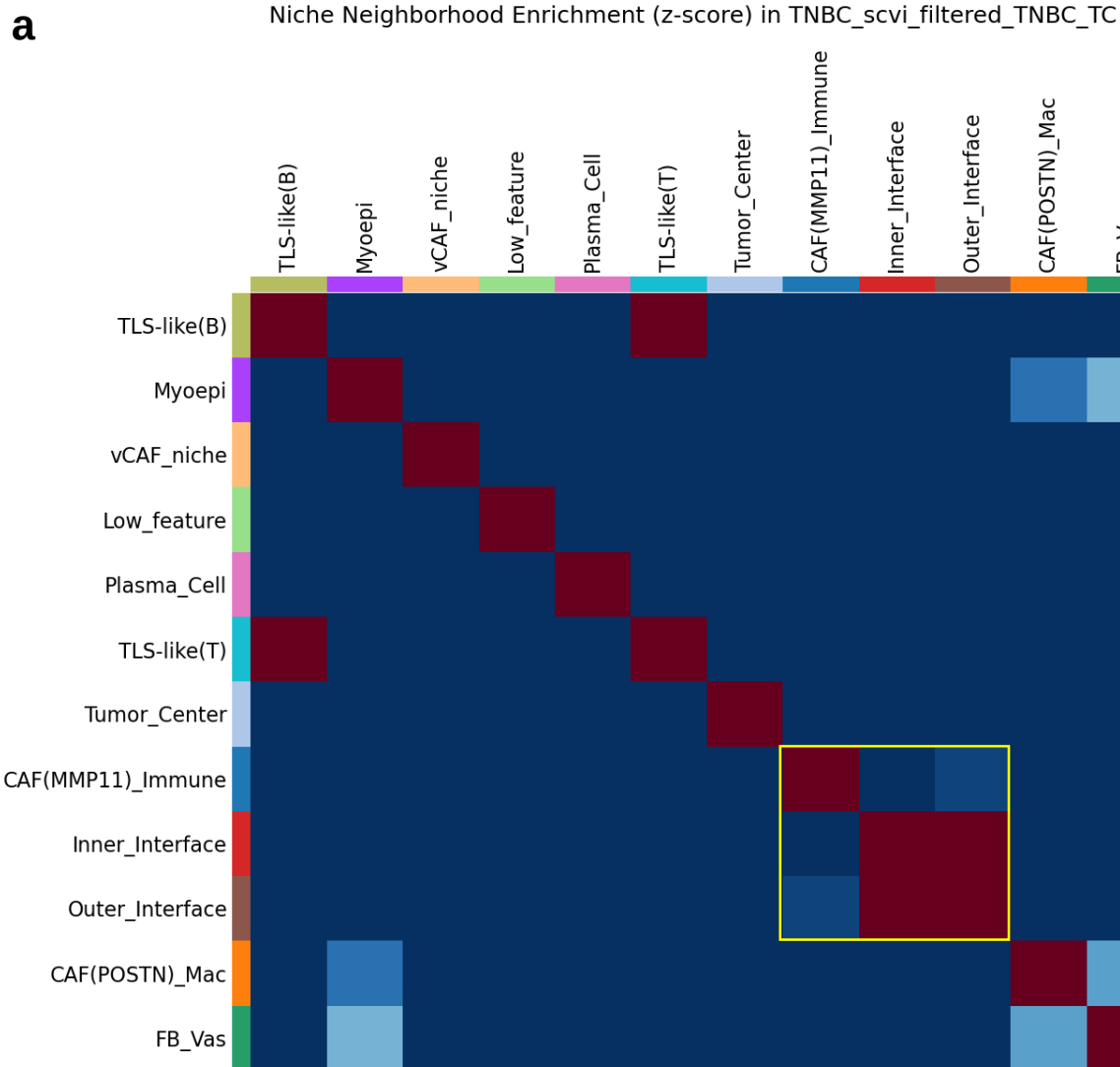
**b**

Genome-wide correlation: MC2(LDHA_PLAUR) expression vs TC3(CTL) infiltration
(Whole niche, Core-level, $n=76$ cores)



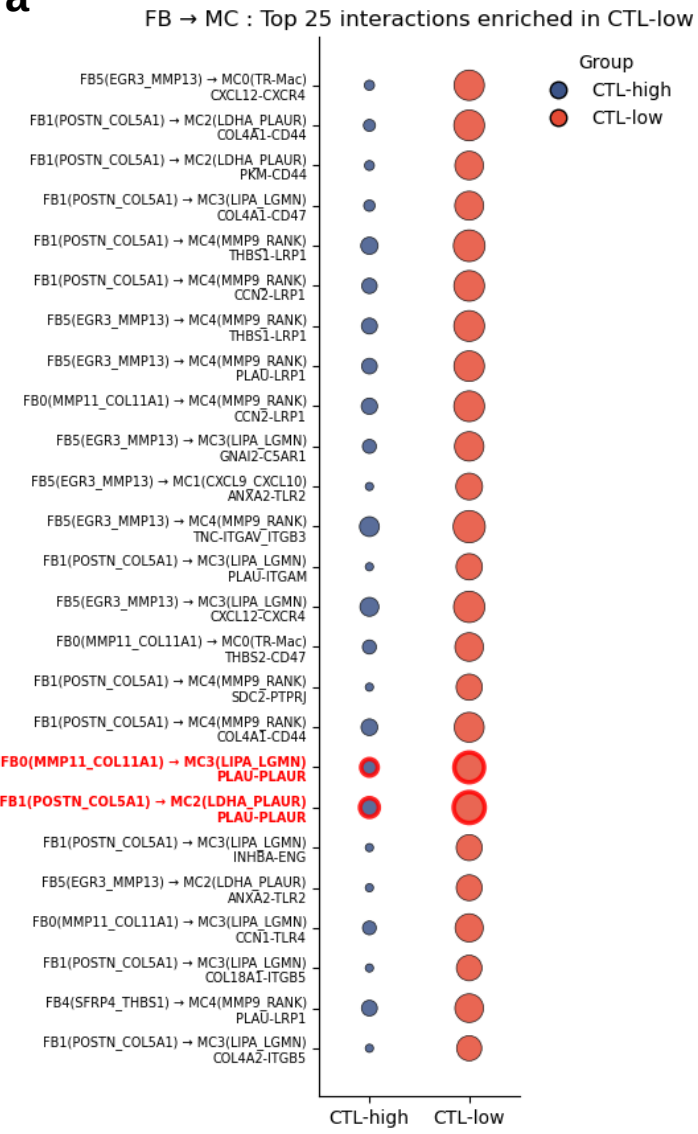
Supplementary Figure 6. Panel-wide correlation confirms PLAUR among top genes linked to CTL exclusion from MC2(LDHA_PLAUR)

- (a) Top nine genes negatively correlated with TC3(CTL) proportion in panel-wide (Xenium 5K panel) screen. Scatter plots show gene expression in MC2(LDHA_PLAUR) vs. TC3(CTL) proportion per core (n=76 cores).
- (b) Volcano plot of gene expression in MC2(LDHA_PLAUR) and TC3(CTL) proportion. Blue/red, FDR<0.05 negative/positive correlations. PLAUR ranks 7th among negatively correlated genes.

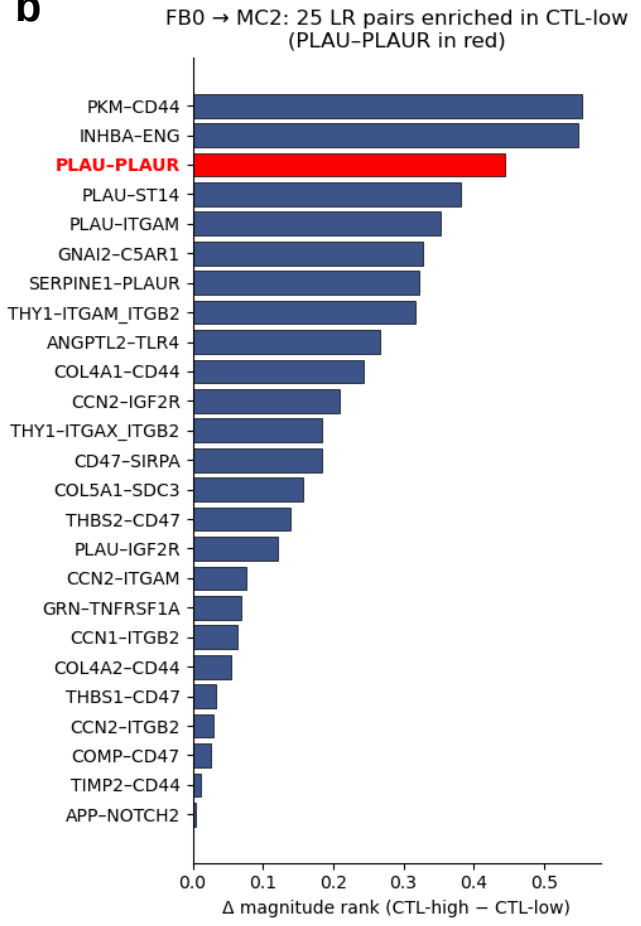


Supplementary Figure 7. (a) Heatmap of pairwise neighborhood enrichment z-scores between spatial niches, computed via permutation testing (squidpy (sq.gr.nhood_enrichment), 1,000 permutations, 50 μ m radius). Yellow box indicates that CAF(MMP11)_Immune niche is enriched with tumoral niche (Outer_Interface).

a



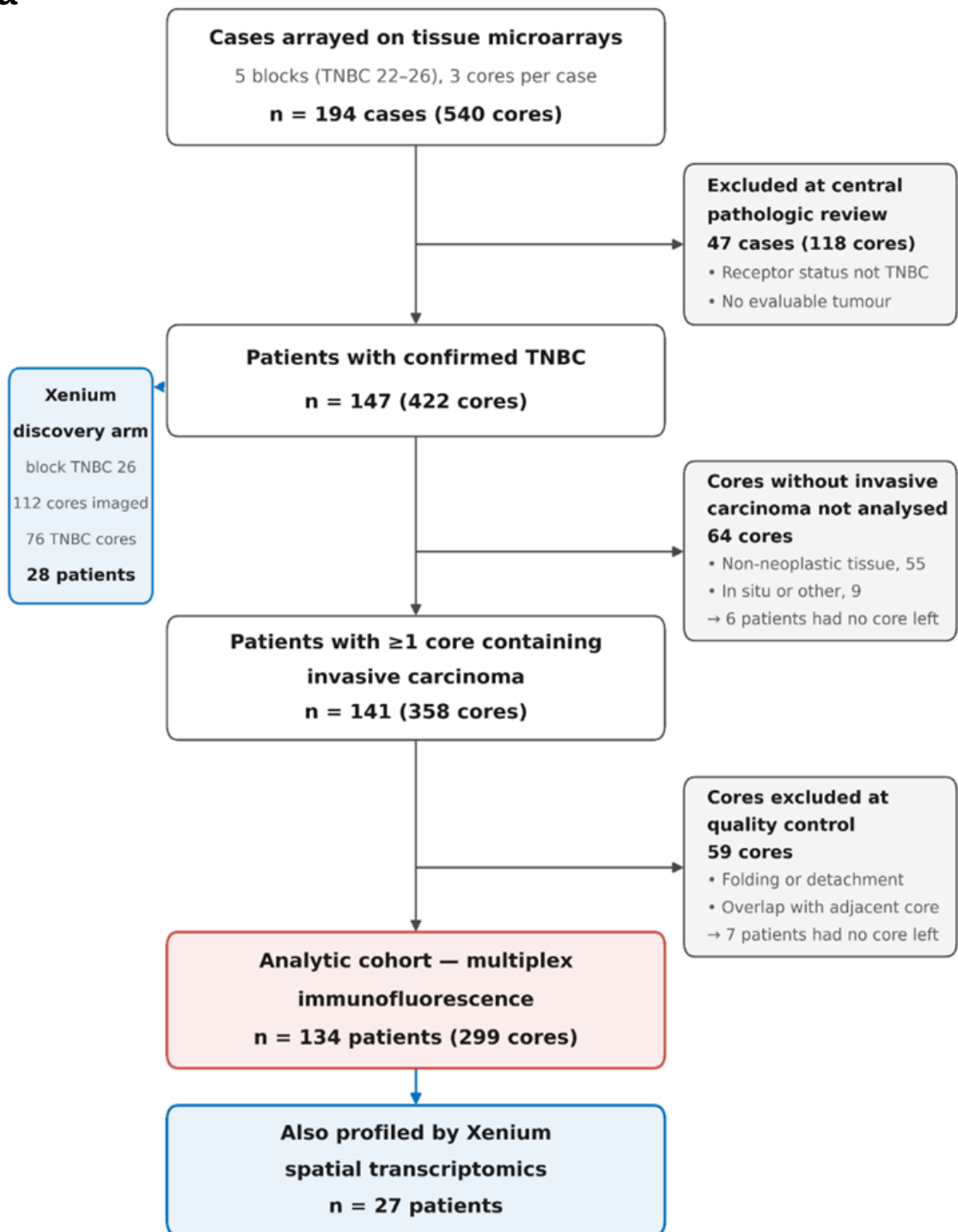
b



Supplementary Figure 8. *PLAU–PLAUR* ligand–receptor signaling is enriched in CTL-low tumors.

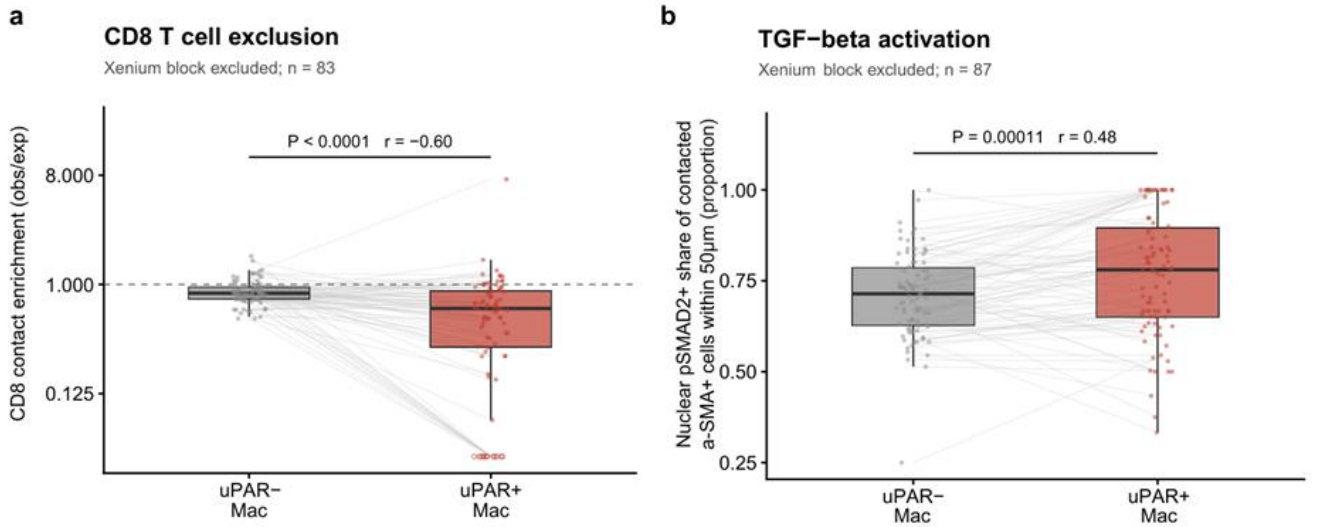
- (a) Dot plot of the top 25 fibroblast (FB)→macrophage (MC) ligand–receptor (LR) pairs ranked by enrichment in CTL-low cores, identified using LIANA (rank_aggregate, core-level). Dot size indicates interaction strength (1 – magnitude_rank) within each group (CTL-high, blue; CTL-low, red). *PLAU*(FB0)–*PLAUR*(MC3) and *PLAU*(FB1)–*PLAUR*(MC2), highlighted in red, rank among the top 25 CTL-low-enriched pairs.
- (b) All 25 ligand–receptor pairs from FB0(MMP11_COL11A1) to MC2(LDHA_PLAUR) enriched in CTL-low cores, ranked by the difference in LIANA magnitude rank between groups (CTL-high – CTL-low). Positive values indicate stronger predicted interactions in CTL-low cores. *PLAU–PLAUR* (red) ranks third.

a



Supplementary Figure 9. Patient selection.

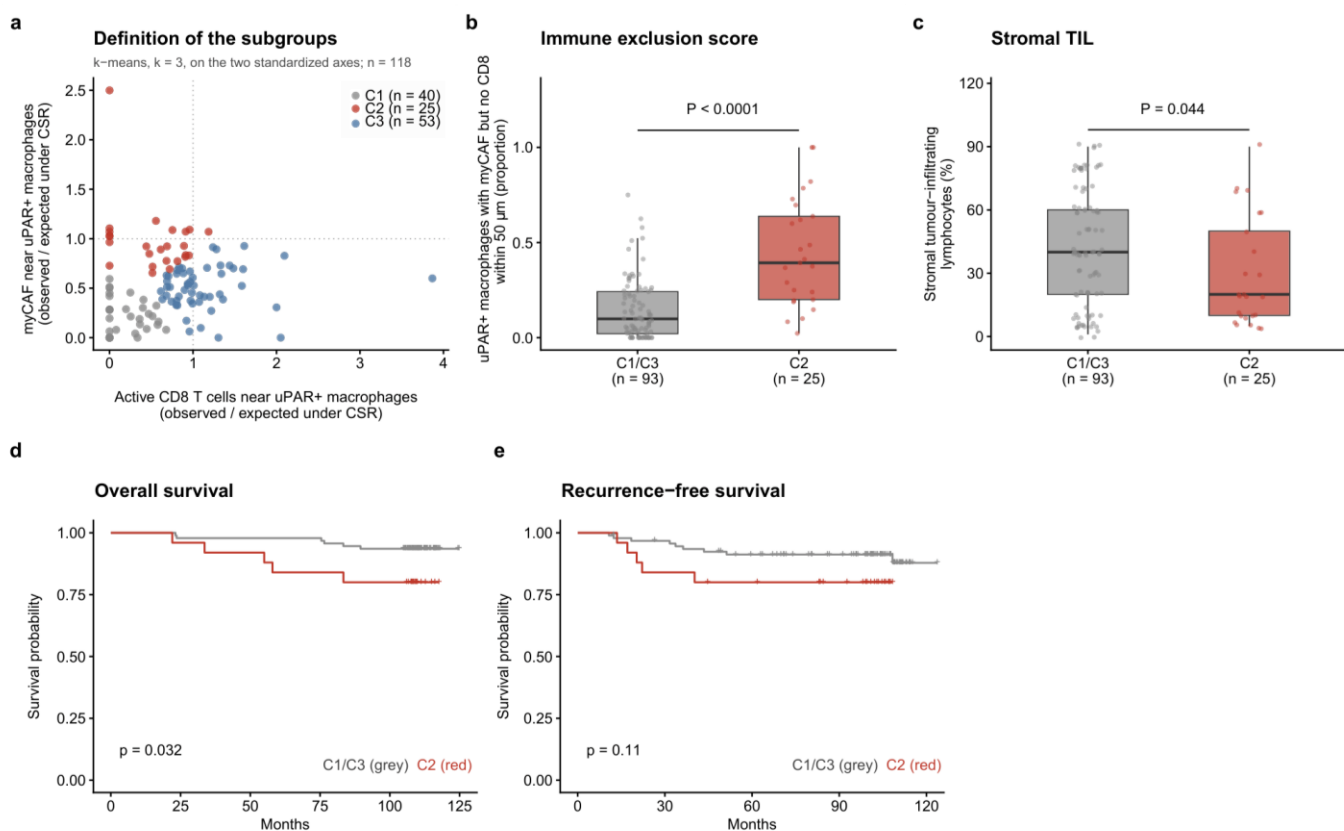
(a) Cases arrayed on five tissue microarray blocks were reviewed centrally, and cases that did not fulfil the definition of TNBC or that contained no evaluable tumor were excluded. Analyses were restricted to cores containing invasive carcinoma.



Supplementary Figure 10. CD8 exclusion and CAF pSMAD2 activation by PLAUR⁺ macrophages persist after excluding the Xenium-profiled patients.

- (a) CD8⁺ T-cell contact enrichment around uPAR⁺ versus uPAR⁻ macrophages (50 μ m), as in Fig. 6e, after excluding patients also profiled by Xenium (TMA block TNBC26); validation cohort n=83. Dashed line, enrichment = 1. Paired Wilcoxon signed-rank $P < 0.0001$, $r = -0.60$.
- (b) pSMAD2⁺ fraction of neighboring α SMA⁺ CAFs around uPAR⁺ versus uPAR⁻ macrophages (50 μ m), as in Fig. 6f; n=87. Paired Wilcoxon signed-rank $p = 0.00011$, $r = 0.48$.

Boxes, median and IQR; grey lines connect paired per-patient values.



Supplementary Figure 11. The immune-excluded C2 subgroup.

- (a) Definition of the subgroups. Unsupervised k-means clustering ($k = 3$, random seed 42) of patients on two standardized, CSR-normalized enrichment features computed over the whole core: enrichment of nuclear-pSMAD2⁺ (activated) CD8⁺ T cells near uPAR⁺ macrophages (x) and of α SMA⁺ myCAFs near uPAR⁺ macrophages (y). Of 134 patients, 128 had at least one uPAR⁺ macrophage in an evaluable core, and a further 10 had no activated CD8⁺ T cell in any such core and could not be assigned, leaving $n = 118$. C1 (grey, $n = 40$), low engagement with either population; C3 (blue, $n = 53$), CD8-enriched; C2 (red, $n = 25$), CD8-low and myCAF-high, defining the immune-excluded subgroup. Dotted lines, enrichment = 1.
- (b) The immune exclusion score — the proportion of uPAR⁺ macrophages with a myCAF but no CD8⁺ T cell within 50 μ m — is highest in C2 relative to the combined C1/C3 subgroups (Wilcoxon rank-sum $P < 0.0001$).
- (c) Stromal TILs are lower in C2 than C1/C3 (Wilcoxon $P = 0.044$).
- (d,e) Kaplan–Meier overall survival (d) and recurrence-free survival (e) for C2 versus C1/C3.

Overall survival: 11 events; unadjusted Cox HR 3.39 (95% CI 1.03–11.10), log-rank $P = 0.032$. Recurrence-free survival: 14 events; unadjusted HR 2.41 (95% CI 0.80–7.29), log-rank $P = 0.11$.

Boxes in (b, c), median and IQR.

Order	Antibody	Vendor	Cat. no.	Dilution	Astra-dye	Dye dilution
1	CD8	Bio-Rad	MCA1817	1:50	570 (C0110)	1:150
2	α SMA	Abcam	ab5694	1:100	520 (C0109)	1:150
3	pSmad2	Abcam	ab53100	1:50	690 (C0114)	1:150
4	uPAR (PLAUR)	Abcam	ab218106	1:500	620 (C0113)	1:150
5	CD68	Abcam	ab192847	1:300	480 (C0115)	1:150
6	Pan-cytokeratin	Novus	NBP2-29429	1:300	TSA-DIG (C0116); 780	1:150; 1:75

Supplementary Table S1. Antibodies and fluorophores used for multiplex immunofluorescence.

Characteristic	Overall (n = 118)	C1/C3 (n = 93)	C2 (n = 25)
Age, years	47 (40, 52)	47 (40, 51)	49 (40, 54)
Tumor size, cm	2.10 (1.73, 2.50)	2.00 (1.70, 2.50)	2.40 (2.10, 2.50)
Ki-67, %	70 (50, 90)	70 (40, 90)	70 (60, 80)
Stromal TIL, %	40 (10, 60)	40 (20, 60)	20 (10, 50)
pT category			
T1	56 (47%)	50 (54%)	6 (24%)
T2	62 (53%)	43 (46%)	19 (76%)
pN category			
N0	87 (74%)	70 (75%)	17 (68%)
N1-N3	31 (26%)	23 (25%)	8 (32%)
pStage			
I	48 (41%)	43 (46%)	5 (20%)
II-III	70 (59%)	50 (54%)	20 (80%)
Lymphovascular invasion			
Absent	99 (84%)	85 (91%)	14 (56%)
Present	19 (16%)	8 (9%)	11 (44%)

Continuous variables are median (Q1, Q3) and were compared with the Wilcoxon rank-sum test; categorical variables are n (%) and were compared with the chi-square test (pT) or Fisher exact test (pN, pStage, LVI).
CI, confidence interval; LVI, lymphovascular invasion; myCAF, myofibroblastic cancer-associated fibroblast; STIL, stromal tumour-infiltrating lymphocytes.

Supplementary Table S2. Clinicopathologic characteristics according to spatial k-means subgroup

Patients were assigned to one of three subgroups by k-means clustering (k = 3) on patient-level uPAR⁺ macrophage–nuclear pSMAD2⁺ CD8⁺ T cells (activated CD8) and uPAR⁺ macrophage–a-SMA⁺ myCAFs neighbourhood enrichment. C2 denotes the immune-excluded subgroup (activated CD8-low, myCAF-high); C1 and C3 are pooled for comparison. Of the 134 patients in the multiplex immunofluorescence cohort, 118 had evaluable enrichment values on both axes and could be assigned to a subgroup.

Variable	n	Overall survival (11 events)		Recurrence-free survival (14 events)	
		HR (95% CI)	P	HR (95% CI)	P
Age, per year	118	0.96 (0.90–1.03)	0.241	0.98 (0.93–1.04)	0.557
Tumor size, per cm	118	2.36 (1.25–4.45)	0.008	1.96 (1.04–3.70)	0.036
Ki-67, per 10%	100	1.01 (0.74–1.38)	0.957	1.13 (0.85–1.50)	0.398
sTIL, per 10%	118	0.98 (0.79–1.21)	0.818	0.87 (0.71–1.07)	0.193
pT category, T2 vs T1	118	4.33 (0.93–20.03)	0.061	2.49 (0.78–7.94)	0.124
pN category, N1-3 vs N0	118	3.51 (1.07–11.50)	0.038	3.07 (1.08–8.76)	0.036
pStage, II-III vs I	118	3.25 (0.70–15.04)	0.132	2.79 (0.78–10.00)	0.116
Lymphovascular invasion, present vs absent	118	11.17 (3.26–38.23)	<0.001	3.47 (1.16–10.42)	0.026
k-means subgroup, C2 vs C1/C3	118	3.39 (1.03–11.10)	0.044	2.41 (0.80–7.30)	0.120

HR, hazard ratio; CI, confidence interval; LVI, lymphovascular invasion; sTIL, stromal tumour-infiltrating lymphocytes.

Supplementary Table S3. Univariate Cox regression for overall and recurrence-free survival

Univariate Cox proportional-hazards models in the 118 patients assignable to a spatial k-means subgroup (median follow-up 109.9 months). Each variable was entered alone. The k-means subgroup term contrasts the immune-excluded C2 subgroup (n = 25) with C1/C3 (n = 93).

Variable	Univariate		Model 1: C2 + pT category		Model 2: C2 + lymphovascular invasion	
	HR (95% CI)	P	HR (95% CI)	P	HR (95% CI)	P
Overall survival						
Immune-excluded subgroup (C2 vs C1/C3)	3.39 (1.03–11.10)	0.044	2.56 (0.76–8.62)	0.129	1.23 (0.33–4.60)	0.762
pT2 vs pT1	4.33 (0.93–20.03)	0.061	3.51 (0.73–16.80)	0.116	—	—
Lymphovascular invasion, present vs absent	11.17 (3.26–38.23)	<0.001	—	—	10.20 (2.59–40.13)	<0.001
Recurrence-free survival						
Immune-excluded subgroup (C2 vs C1/C3)	2.41 (0.80–7.29)	0.120	1.97 (0.63–6.15)	0.243	1.61 (0.47–5.55)	0.452
pT2 vs pT1	2.49 (0.78–7.94)	0.124	2.15 (0.65–7.11)	0.208	—	—
Lymphovascular invasion, present vs absent	3.47 (1.16–10.41)	0.026	—	—	2.85 (0.84–9.71)	0.094

¹ HR, hazard ratio; CI, confidence interval; LVI, lymphovascular invasion. Em dash, variable not included in that model. The proportional-hazards assumption was assessed using scaled Schoenfeld residuals and was not violated in any model (all P>0.05).

Supplementary Table S4. Cox proportional-hazards analysis of overall and recurrence-free survival in relation to the immune-excluded subgroup (C2).

Annot3	n_cells	proportion
Cancer	182525	0.37316
Low_feature_cell	38605	0.07893
Plasma_cell	27268	0.05575
FB0(MMP11_COL11A1)	25641	0.05242
TC1(CXCL13_CXCL9)	25118	0.05135
TC0(Treg)	23330	0.04770
MC0(TR-Mac)	23184	0.04740
FB1(POSTN_COL5A1)	19576	0.04002
MC1(CXCL9_CXCL10)	17424	0.03562
B_cell	14296	0.02923
TC2(MIAT LENG8)	11465	0.02344
FB6(vCAF_ABCC9)	10472	0.02141
FB2(NR4A1_CXCL12)	9230	0.01887
MC2(LDHA_PLAUR)	8485	0.01735
VC0(EC-TGFBR2)	8386	0.01714
VC1(pericyte)	7340	0.01501
DC0(LAMP3)	6512	0.01331
Myoepi	6455	0.01320
VC2(EC_cancer)	5573	0.01139
TC3(CTL)	4608	0.00942
MC3(LIPA_LGMN)	4408	0.00901
VC3(vSMC)	1542	0.00315
FB3(VEGFA_NDRG1)	1401	0.00286
DC1(pDC)	1285	0.00263
MC4(MMP9_RANK)	1053	0.00215
FB4(SFRP4_THBS1)	1031	0.00211
Cancer_GABRP	852	0.00174
VC4(LEC)	694	0.00142
DC2(cDC1)	619	0.00127
FB5(EGR3_MMP13)	517	0.00106
Adipocyte	116	0.00024
Mast	72	0.00015
MC5(low_feature)	46	0.00009

Supplementary Table S5. Cell count per subcluster.

Core	Patient	n_total	n_CTL	CTL_prop	CTL_group
Core49	Pt476	16528	703	0.04253388	CTL-high
Core96	Pt1055	5087	210	0.0412817	CTL-high
Core72	Pt747	5782	181	0.03130405	CTL-high
Core42	Pt437	12174	355	0.02916051	CTL-high
Core43	Pt437	9940	280	0.02816901	CTL-high
Core5	Pt6	2891	68	0.02352127	CTL-high
Core31	Pt366	1796	41	0.02282851	CTL-high
Core30	Pt366	6154	137	0.02226194	CTL-high
Core74	Pt747	5652	121	0.02140835	CTL-high
Core12	Pt101	6689	128	0.01913589	CTL-high
Core32	Pt366	6685	127	0.01899776	CTL-high
Core110	Pt711	11180	194	0.01735242	CTL-high
Core93	Pt979	7040	115	0.01633523	CTL-high
Core58	Pt515	1811	28	0.01546107	CTL-high
Core33	Pt378	3257	48	0.01473749	CTL-high
Core85	Pt949	5514	78	0.01414581	CTL-high
Core53	Pt492	5957	84	0.01410106	CTL-high
Core109	Pt711	10631	149	0.01401561	CTL-high
Core87	Pt969	8734	115	0.01316693	CTL-high
Core81	Pt840	2080	27	0.01298077	CTL-high
Core57	Pt515	6911	77	0.01114166	CTL-high
Core35	Pt378	3209	35	0.01090682	CTL-high
Core95	Pt979	8165	87	0.01065524	CTL-high
Core44	Pt437	9753	103	0.01056085	CTL-high
Core84	Pt949	6301	63	0.00999841	CTL-high
Core34	Pt378	4073	39	0.00957525	CTL-high
Core14	Pt156	5436	50	0.00919794	CTL-high
Core18	Pt179	5769	53	0.00918703	CTL-high
Core63	Pt588	7667	66	0.00860832	CTL-high
Core16	Pt156	1639	14	0.00854179	CTL-high
Core97	Pt1055	2429	20	0.00823384	CTL-high
Core108	Pt711	9257	74	0.00799395	CTL-high
Core9	Pt17	4646	37	0.00796384	CTL-high
Core52	Pt492	5929	45	0.00758981	CTL-high
Core55	Pt496	11433	77	0.00673489	CTL-high
Core11	Pt101	4552	28	0.00615114	CTL-high
Core7	Pt17	7137	36	0.00504414	CTL-high
Core102	Pt1072	8739	40	0.00457718	CTL-high
Core21	Pt261	9421	42	0.00445813	CTL-low

Core70	Pt658	7101	30	0.00422476	CTL-low
Core83	Pt840	8827	35	0.00396511	CTL-low
Core26	Pt350	6149	24	0.00390307	CTL-low
Core50	Pt476	9670	37	0.00382627	CTL-low
Core3	Pt4	5213	19	0.00364473	CTL-low
Core56	Pt496	8757	31	0.00354003	CTL-low
Core17	Pt179	4529	14	0.00309119	CTL-low
Core46	Pt447	8205	24	0.00292505	CTL-low
Core10	Pt44	7441	20	0.00268781	CTL-low
Core65	Pt588	5130	13	0.00253411	CTL-low
Core104	Pt1072	6815	17	0.0024945	CTL-low
Core45	Pt447	8467	19	0.00224401	CTL-low
Core69	Pt658	7799	17	0.00217977	CTL-low
Core15	Pt156	4299	9	0.00209351	CTL-low
Core59	Pt515	4939	10	0.0020247	CTL-low
Core78	Pt831	2508	5	0.00199362	CTL-low
Core64	Pt588	9265	18	0.0019428	CTL-low
Core6	Pt6	3458	6	0.00173511	CTL-low
Core47	Pt447	8100	14	0.0017284	CTL-low
Core82	Pt840	5913	8	0.00135295	CTL-low
Core103	Pt1072	6676	8	0.00119832	CTL-low
Core94	Pt979	5312	6	0.00112952	CTL-low
Core22	Pt261	9812	11	0.00112108	CTL-low
Core19	Pt179	5712	6	0.00105042	CTL-low
Core1	Pt4	4595	4	0.00087051	CTL-low
Core13	Pt101	1448	1	0.00069061	CTL-low
Core25	Pt350	5840	4	0.00068493	CTL-low
Core80	Pt831	5995	4	0.00066722	CTL-low
Core51	Pt492	3128	2	0.00063939	CTL-low
Core48	Pt476	5935	3	0.00050548	CTL-low
Core79	Pt831	6475	3	0.00046332	CTL-low
Core2	Pt4	2396	1	0.00041736	CTL-low
Core24	Pt350	5948	2	0.00033625	CTL-low
Core20	Pt261	12010	4	0.00033306	CTL-low
Core54	Pt496	7268	2	0.00027518	CTL-low
Core8	Pt17	3857	1	0.00025927	CTL-low
Core98	Pt1055	6089	1	0.00016423	CTL-low

Supplementary Table S6. CTL proportion, patient ID, and CTL group per core.

Supplementary Data S1. Differentially expressed gene list for each cell type

(subclusters), including expression scores, log fold-changes, adjusted p-values, and the percentage of cells expressing each gene within (pct.in) and outside (pct.out) the group.