Supplementary Table 1. Cell surface antigen expression in melanoma samples

Patient	CD117	CD133	CD166	CD271	EGFR	CD51/61	CD49f	CD49d	CD90	CD95	CD44	CD47	CD105	CD146	CD56	CD71	CD24	EpCAM	FGF-R	TRAIL-R	CXCR-4	FZD-7
Mel327	H	H	+	H	-	+	H											-				
Mel114	-			H							H			H				-				
Mel525	-	-	-	H			-		-	-	H			H				-				
Mel213	-	-		H							H			H				-			-	-
Mel1119		H		H			H				+			H				-				
Mel826	-	-		H							+			H				-				
Mel210	H	-		H							+	+		H				-				
Mel425	H	H		H		-	H		+			+		H				-			-	-
Mel223	H	-		-	+				+		+	-	+	H				-	H		H	-
Mel43 Xeno P0	-	-	+	H				H	H	+	+	+		H					-	-		
Mel327 Xeno Pi	H	-	-	H	-	+	-		-	+	+	+	H	+	H	+						
Mel 525-Xeno P0	-		H	H	-		-	H	-		+	+	-	H			-					
Mel525- Xeno Pi	-		+	H			H	H	-		+	+		H		H	-	-				
Mel415 Xeno P0	-	-		H			H		-					H								

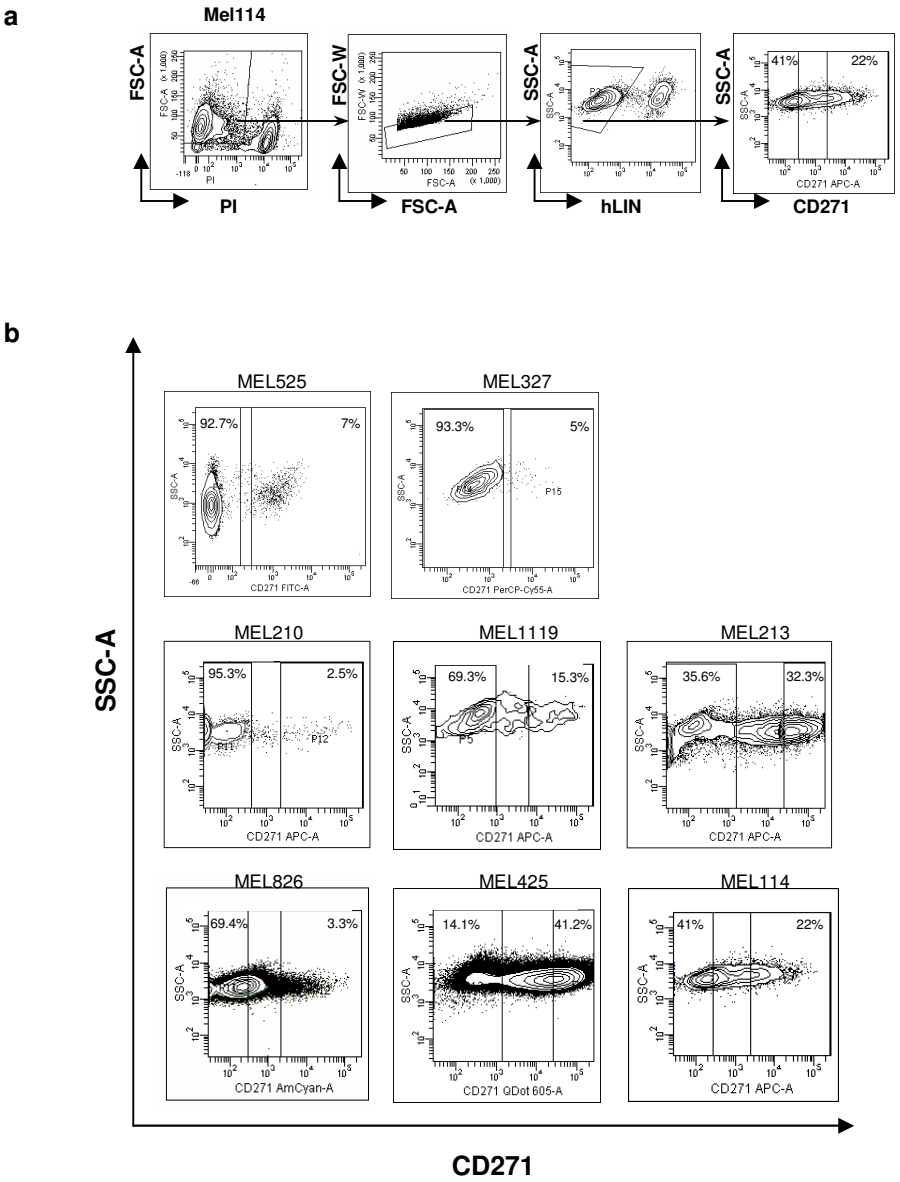
FACS analysis of live, Lin- single cell suspension melanoma cells
(H) Positive and negative cell populations are present in the analyzed single cell suspension
(-) 100% of analyzed cells do not express cell surface antigen
(+) 100% of analyzed cells express cell surface antigen
Xeno (P0) - surgically removed patient's tumor implanted into RagDKO mice;
Xeno (Pi) surgically removed patient's tumor expanded in-vitro and injected into Rag2gDKO mice

Surgical or xenografted melanoma samples were mechanically and enzymatically dissociated to obtain mixture containing single cells that was stained with human (hCD45-/CD2-/CD3-/CD31-) or mouse (H2-Kd/mCD45.2/Ter119) lineage antibody cocktail in combination with Mab to indicated CD antigens
.

Supplementary Table 2. Melanoma – Primary Tumor Characteristics

Patient Sample ID	Diagnosis/Site	Age/ Gender	AJCC Stage at Dx, Breslow Depth, Clark level	Ulceration /Mitotic index (x/mm2)	LV / PN Invasion	Primary Treatment	+SLN/Tot , +CLN/Tot	Sample tissue type, location	Additional Therapy	%CD271+	Follow Up
Mel327	Superficial spreading melanoma, R leg	24, F	IB/T1aN0M0, 1mm, III	N / 2	N / N	WLE, SLNB	0/4 , N/A	LN, Right groin, Stage III	Adjuvant XRT, IFN, IL-2;	5	Alive 9/5/09
Mel43	Spindle cell/desmoplastic melanoma, forehead	80, M	IIB/T4aN0M0, 7mm, V	N / 2	Y / Y	WLE, SLNB	0/3, N/A	Skin (primary), Stage IIB	Palliative CTX	75.3 (xeno only)	Deceased
Mel525	Metastatic melanoma (parotid) L neck , unknown primary	69, M	IIIC/TxN3M0	U / U	U/ U	CLND	N/A, 5/30	LN (parotid/ neck), Stage III	Palliative CTX	7	Deceased
Mel415	Right temple primary melanoma	75, M	IIC/T4bN0M0, >4 mm	Y / U	U / U	WLE	N/A, 5/49	LN (parotid/ neck) Stage III	CLND	11.3 (xeno only)	Deceased
Mel425	Primary dermal melanoma, R back	84, M	IIIB/T4aN2cM0, 3.2mm, IV	N / U	U / U	WLE, SLNB	0/1, N/A	Dermal/ SQ mass (primary) Stage III	None	41.2	Alive, 9/24/09
Mel826	Primary dermal melanoma, L leg	53, F	IIB/T4aN0M0, 10.4mm, V	N / 3	N / N	WLE, SLNB	0/4, N/A	Primary melanoma	Adjuvant HDI	3.3	Alive 9/2/09
Mel1119	Superficial spreading melanoma, L neck	21, M	IB/T1aN0M0, 1.1mm, III	N / 1	N / N	WLE, SLND	0/2 , N/A	Subcutaneous met Stage IV	HDI (Abraxane/ Avastin); XRT	15.3	Deceased
Mel114	Nevoid melanoma, R post shoulder	50, M	IIA/T4bN0M0, 2.2 mm, IV	N / 5	N / N	WLE, SLNB	0/1, N/A	SQ mass, L supraclav c/w satellite/in-transit met Stage III	WLE	22	Alive 1/28/09
Mel210	Superficial spreading melanoma, L leg	52, F	IB/T1bN0M0, 1.4mm, III	N / <1	N / U	WLE, SLNB	0/2, N/A	L knee SQ mass (in-transit met) Stage III	IFN, XRT;	2.5	Alive 9/10/09
Mel213	Regional nodal disease, unknown primary	79, F	IIIC/TxN3M0, xmm, unknown	U / U	U / U	CLND	N/A, 1/9	LN (R axillary) Stage III	Adjuvant XRT	32.3	Alive 4/22/09

Subtypes of melanoma diagnosed, along with anatomic location, patient age, and gender used in this study. The AJCC (2002) Stage at diagnosis is listed. Unknown staging components are marked as "x." Ulceration, mitoses, lymphovascular invasion (LVI) and perineural invasion (PNI) invasion are listed as yes (Y), no (N), or unknown (U). Treatments included wide local excision (WLE), sentinel lymph node biopsy (SLNB), complete lymph node dissection (CLND), adjuvant high-dose interferon (HDI) therapy, cyclophosphamide (CTX) treatment and radiotherapy (XRT). Sentinel and regional node positivity (per CLND) is indicated by number of positive lymph nodes / total # lymph nodes dissected (+SNL/Tot, and +CLN/Tot, respectively). N/A, not applicable/not performed.



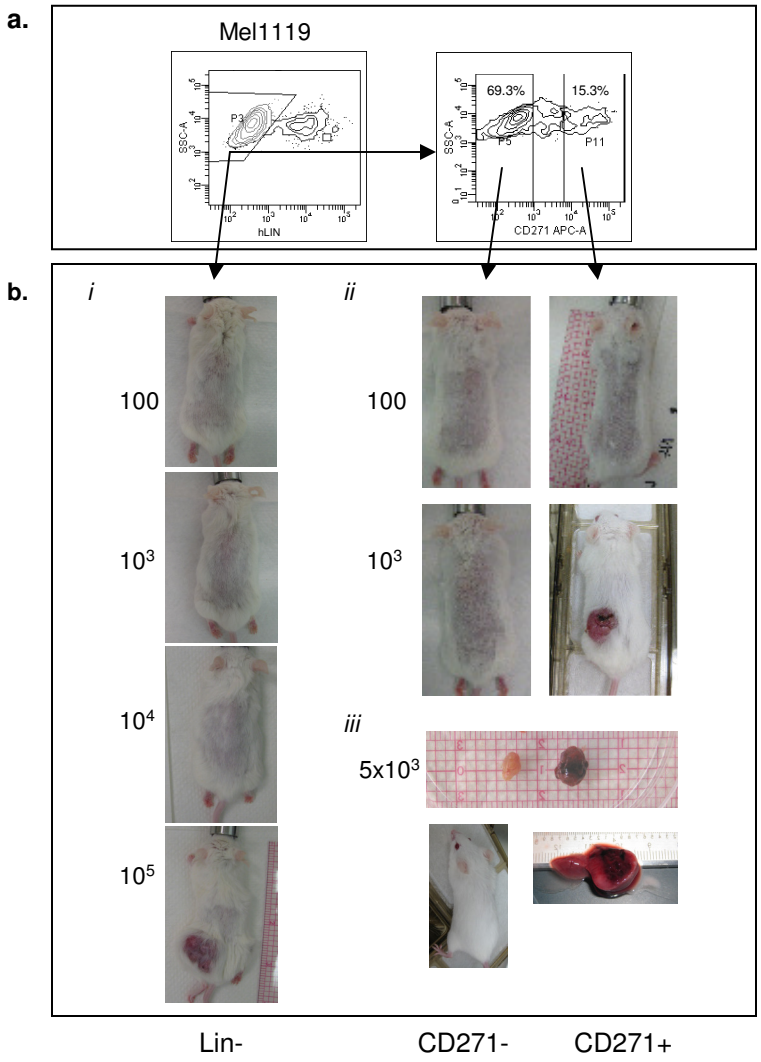
Supplementary Figure 1. Expression of CD271 in melanoma patients.
Single cell suspensions were prepared from surgical samples and live, hLin- (CD45, CD2, CD3, CD31)) cells analyzed on BDFACSAria instrument.
a, Representative contour plot FACS gating sequence of Mel patient sample; **b,** Flow cytometric contour plots demonstrating the variable expression of CD271 in different patients.

Supplementary Table 3. Isolation of melanoma tumor initiating cells expressing CD271$^{P75(NGFR)}$ from melanoma patients

Patient	10			10^2			10^3			$(2\text{-}5)x10^3$			10^4			10^5		
	Lin-	CD271-	CD271+	Lin-	CD271-	CD271+	Lin-	CD271-	CD271+	Lin-	CD271-	CD271+	Lin-	CD271-	CD271+	Lin-	CD271-	CD271+
Mel114	0/2	0/2	0/2	0/2	0/2	1/2	0/2	0/2	2/2	0/2	0/2	1/2	0/2	0/2	2/2	2/2	0/2	
Mel425											1/5	4/5		0/1	1/1			
Mel826											1/4	2/2!		0/1*	1/1*!			
Mel210					0/1	1/1		0/1*	1/1*									
Mel1119	0/2			0/2	0/2	0/2	0/2	0/2	1/2		1/2	1/2!	0/2			1/2		
Mel213	0/2	0/2	0/2	0/2	0/2	2/2	1/2	0/2	2/2	1/2	0/4	4/4!						
Total	**0/6**	**0/4**	**0/4**	**0/6**	**0/7**	**4/7**	**1/6**	**0/7**	**6/7**	**1/4**	**3/17**	**12/15**	**0/4**	**0/4**	**4/4**	**3/4**	**0/2**	

(!) metastatic nodules; (*) humanized mouse system

Table of all injected cell doses and tumor frequency formation induced by CD271+, CD271- and bulk hLin- cell populations isolated directly from melanoma patients. Numbers indicate ratio of tumor incidence relative to the number of injections



c.

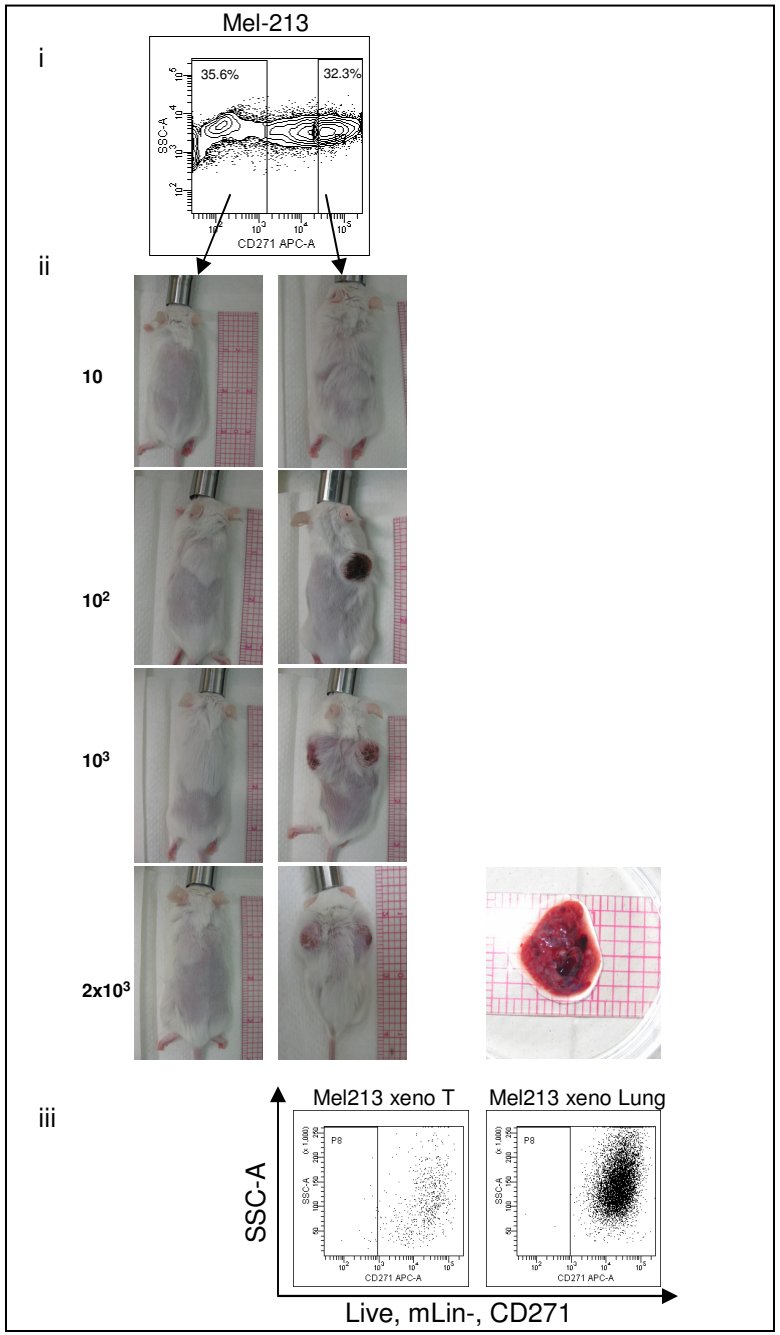
	hLin-	CD271-	CD271+
Mel1119	0/2 (10) 0/2 (100) 0/2 (1,000) 0/2 (10,000) 1/2 (100,000)	0/2 (100) 0/2 (1,000) 1/2 (4,500) small	0/2 (100) 1/2 (1,000) 1/2 (4,500) + liver met
Summary	1/10	1/6	2/6 + liver met

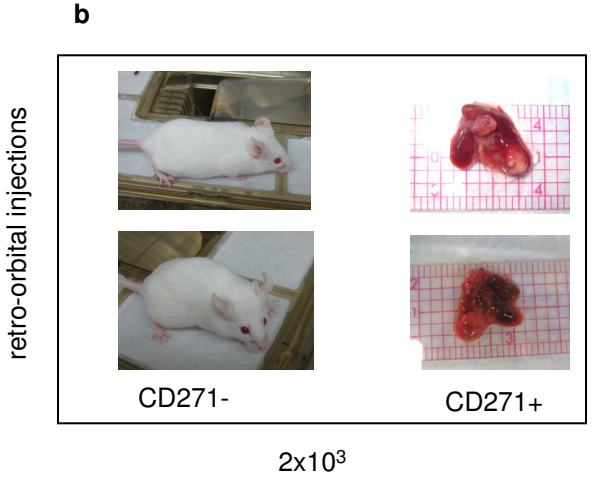
Supplementary Figure 2. Identification of CD271+ melanoma tumor initiating cells in Mel1119, subcutaneous metastatic melanoma patient. **a**, FACS contour plot and sorting strategy of live, single, lineage negative Lin- (CD45, CD2, CD3, CD31), CD271+ and C271- cells isolated from Mel1119 patient sample. **b**, (i) Lin- cells isolated by FACS were mixed with matrigel and injected intradermaly into Rag2gDKO mice at indicated cell doses; tumor growth became notable after 20 weeks. (ii) CD271+ and CD271- cells were isolated at the same time, mixed with matrigel and injected intradermaly into Rag2gDKO mice at indicated cell doses; tumor growth became notable after 20-28 weeks . (iii) tumors that grew at the cell dose 5x103 were surgically removed from anesthetized mice; wound was closed and allowed to heal, 8 weeks post-surgery mouse that was injected with CD271+ cells became morbid and upon examination a large liver met was discovered; mouse injected with CD271- cells is alive after 22 weeks post-surgery **c,** Summary table of all injected cell doses and tumor frequency formation induced by CD271+, CD271- and bulk (Lin-) cell populations isolated from Mel1119 patient.

b

Mel425 Patient	CD271-	CD271+
	1/5 ($2x10^3$)	4/6 ($2x10^3$)
	0/1 (10^4)	1/1 (10^4)
Summary	1/6	5/7

Supplementary Figure 3. Identification of CD271+ melanoma tumor initiating cells in Mel425, subcutaneous primary melanoma patient. **a**, (i) FACS contour plot of live, single, lineage negative (CD45, CD2, CD3, CD31), CD271+ and C271- cells isolated from Mel425 patient sample. (ii) CD271+ and C271- cells isolated by FACS were mixed with matrigel and injected intradermaly into Rag2gDKO mice at indicated cell doses; tumor growth became notable after 24-36 weeks. **b,** Summary table of all injected cell doses and tumor frequency formation induced by CD271+ and CD271- cell populations isolated from Mel425 patient.



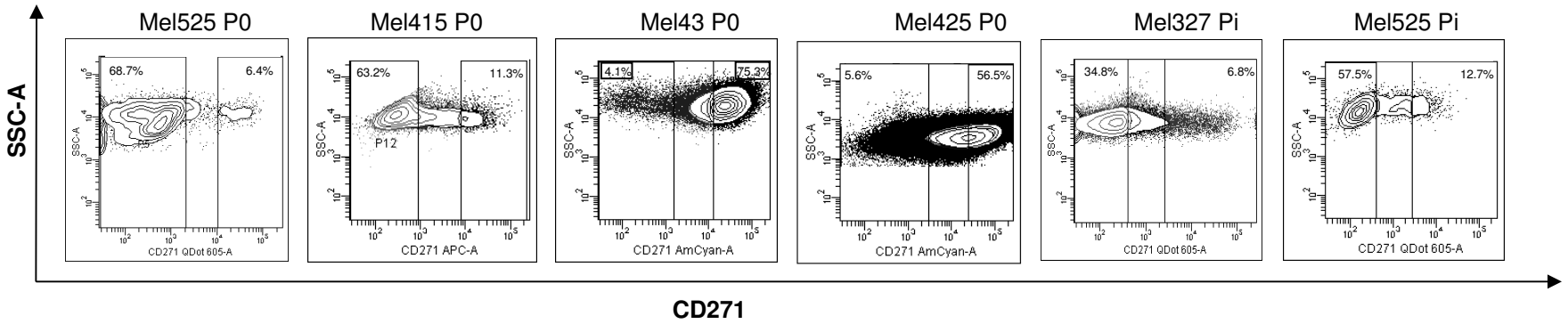


c

	hLin-	intradermal injections	
		CD271-	**CD271+**
Mel 213	0/2 (10) 0/2 (100) 1/2 (1,000) 1/2 (2,000)	0/2 (10) 0/2 (100) 0/2 (1,000) 0/2 (2,000)	0/2 (10) 1/2 (100) 2/2 (1,000) 2/2 (2,000) + Lung Mets
	retro orbital injections		
	0/1 (2,000)	0/1 (2,000) 0/1 (2,000)	1/1 (2,000) Lung Mets 1/1 (2,000) Lung Mets
Summary	2/9	0/10 no Mets	7/10 + Mets

Supplementary Figure 4. Identification of CD271+ melanoma tumor initiating cells in Mel213, lymph node metastatic melanoma patient. **a**, (i) FACS contour

plot of live, single, lineage negative (CD45, CD2, CD3, CD31), CD271+ and C271- cells isolated from Mel213 patient sample. (ii) CD271+ and C271- cells isolated by FACS were mixed with matrigel and injected intradermaly into Rag2gDKO mice at indicated cell doses; tumor growth became notable after 12-16 weeks. (iii) Mouse injected with $2x10^3$CD271+ patient cells developed tumor and lung metastasis after 12 weeks that were re-analalyzed for CD271 expression, FACS contour plot is shown. **b,** CD271+ and C271- cells were isolated by FACS from Mel213 patient and retro-orbital injections at $2x10^3$ cell doses were done into Rag2gDKO mice; after 12 weeks mice injected with CD271+ cells developed morbid phenotype due to lung metastasis. Mice injected with CD271- cells display healthy phenotype with no apparent abnormalities after 28 weeks. **c,** Summary table of all injected cell doses and tumor frequency formation induced by CD271+, CD271- and bulk (Lin-) cell populations isolated from Mel213 patient.



Supplementary Figure 5. Flow cytometric contour plots demonstrating the variable expression of CD271 in melanoma patients whose tumors were analyzed as xenografts. (P0) - surgically removed patient's tumor implanted into RagDKO mice; (Pi) surgically removed patient's tumor expanded in-vitro and injected into Rag2gDKO mice; Single cell suspensions were isolated from xenografted tumors by mechanichal and enzymatic digestion and live, mLin- (H2-Kd-/ mCD45.2-/mTer119-) cells were analyzed with anti-human CD271 Mab on BDFACSAria instrument.

a

Xeno P0	Xeno Pi	10^2		10^3		$2x10^3$		$(3-5)x10^3$		10^4 - $2x10^4$	
		CD271-	CD271+	CD271-	CD271+	CD271-	CD271+	CD271-	CD271+	CD271-	CD271+
Mel525		0/2	0/2	0/2	0/2			0/2	2/2		
	Mel525			0/2	0/2	1/2	2/2	1/2	2/2		
Mel425		0/3	0/3			1/4	4/4			1/4	3/4
Mel43				0/1	6/8		2/4			0/1*	1/1*
	Mel327			0/2	2/2	1/2	2/2				

Summary

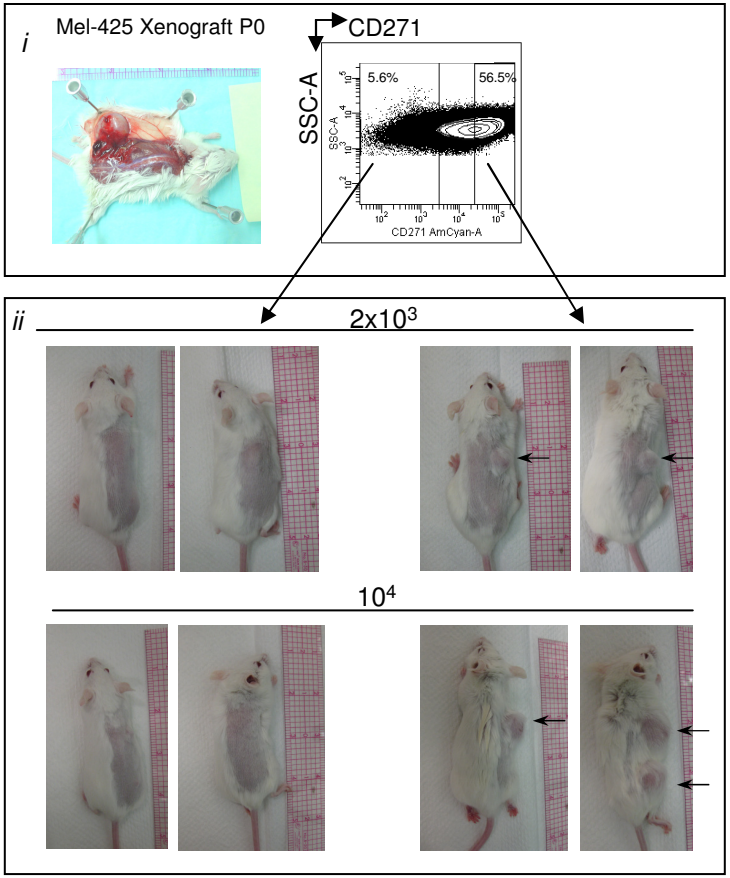
Xenografted Samples (P0, Pi)	10^2		$10^3 – 5x10^3$		10^4 - $2x10^4$	
	CD271-	CD271+	CD271 -	CD271+	CD271-	CD271+
	0/5	0/5	3/11	18/22	1/5	4/5

b

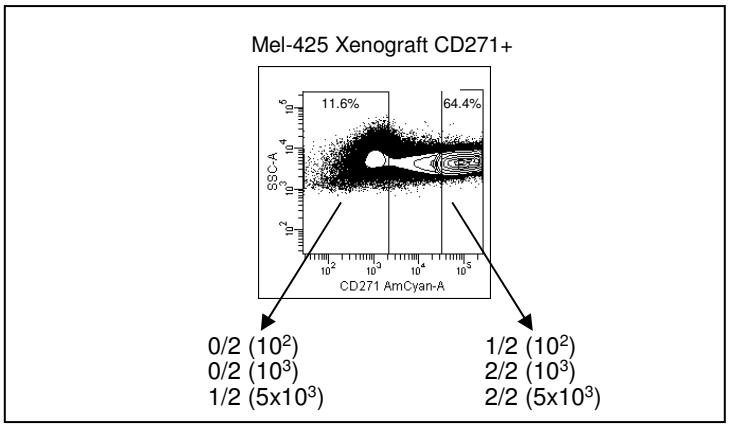
Xenografted Samples (Ps)	10-10^2		$10^3 – 5x10^3$	
	CD271-	CD271+	CD271 -	CD271+
	4/14	9/14	1/4	4/4

Supplementary Figure 6. Melanoma cell engraftment from xenografts
CD271limiting dilution analysis of the human melanoma tumor initiating cell from xenografted patient tumors**.** Rag2gDKO mice were injected with live, Lin- (H2-Kd-/mCD45-/mTer119-), CD271+ and CD271- melanoma cells isolated by FACS and mixed with matrigel. Numbers indicate ratio of tumor incidence relative to the number of injections. **a,** xenograft obtained from surgically removed patient's tumor that was implanted into RG mice (Xeno P0), or expanded in-vitro before being implanted into RG mice (Xeno Pi); **b,** xenograft obtained from surgically removed patient's tumor serially passaged in RG mice

a.



b.

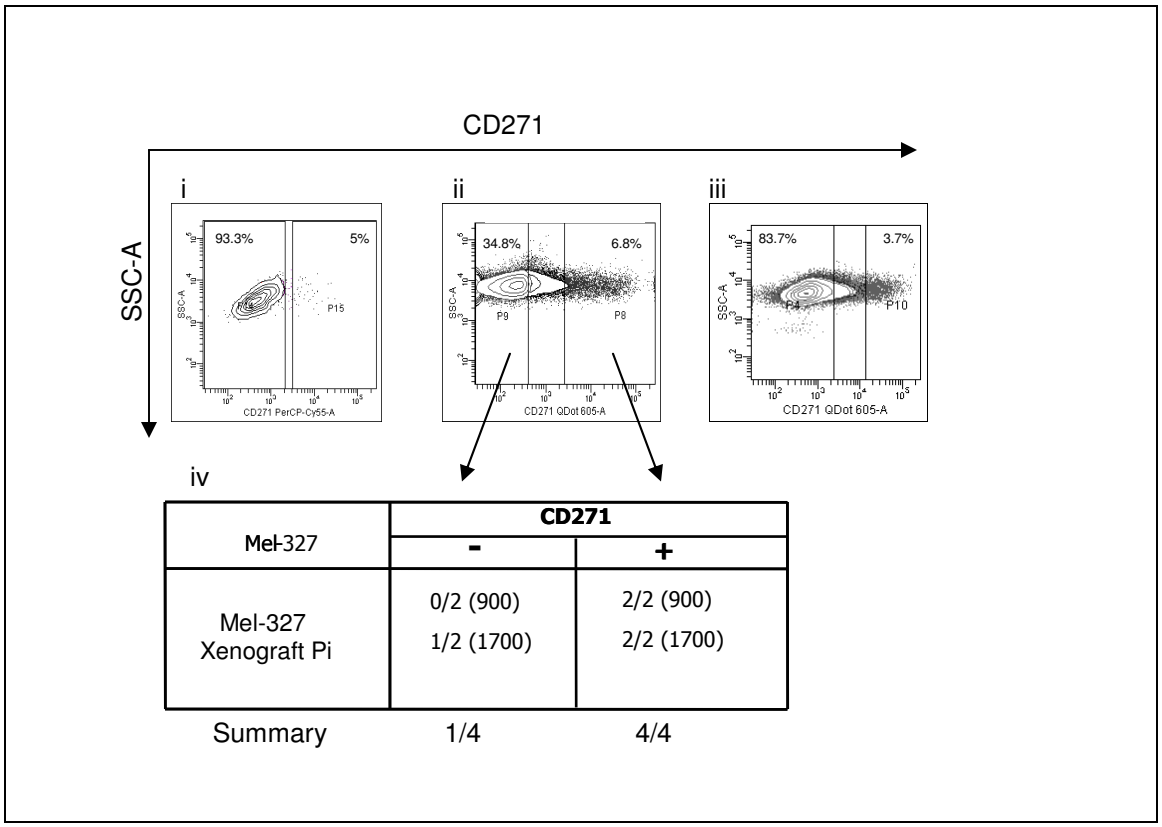


c.

	CD271	
	-	+
Mel425 Xeno P0	1/4 ($2x10^3$) 1/4 (10^4)	4/4 ($2x10^3$) 3/4 (10^4)
Mel425 Xeno P1 CD271+ 10^4	0/2 (100) 0/2 (10^3) 1/2 ($5x10^3$)	1/2 (100) 2/2 (10^3) 2/2 ($5x10^3$)
Summary	3/14	12/14

Supplementary Figure 7. Identification of CD271+ melanoma tumor initiating cells in Mel425 Xenograft P0, subcutaneous primary melanoma

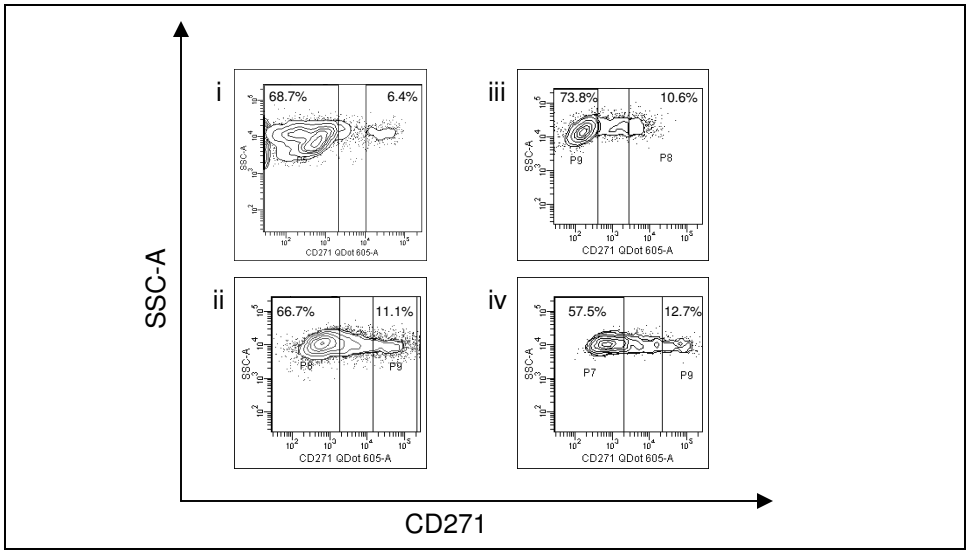
patient. **a**, (i) Surgically removed tumor sample was implanted into the back of Rag2gDKO mouse and after 11 weeks resulting tumor was analyzed for CD271 expression; FACS contour plot of live, single, lineage negative (H2-Kd-/ mCD45.2-/mTer119-), CD271+ and C271- cells isolated from Mel425 Xenograft P0. (ii) CD271+ and C271- cells isolated by FACS were mixed with matrigel and injected intradermaly into Rag2gDKO mice at indicated cell doses; tumor growth became notable after 21-24 weeks. **b,** Tumor induced by injection of 104 CD271+ cells was re-analyzed for CD271 expression; FACS contour plot of live, single, lineage negative (H2-Kd-/ mCD45.2-/mTer119-) cells based on CD271 expression is shown. CD271+ and C271- cells isolated by FACS were mixed with matrigel and injected intradermaly into Rag2gDKO mice at indicated cell doses; tumor growth became notable after 18-23 weeks; frequencies of tumor formation are indicated. **c,** Summary table of all injected cell doses and tumor frequency formation induced by CD271+ and CD271- cell populations isolated from Mel425 Xenograft P0.



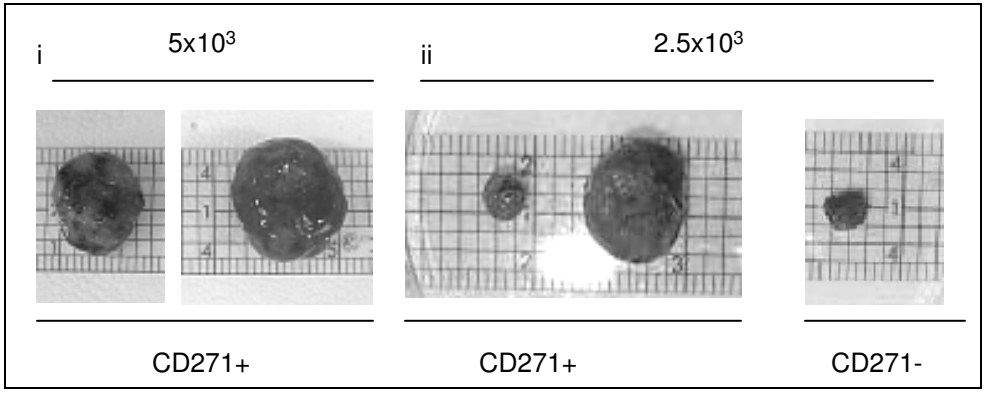
Mel-327	CD271 -	CD271 +
Mel-327 Xenograft Pi	0/2 (900) 1/2 (1700)	2/2 (900) 2/2 (1700)
Summary	1/4	4/4

Supplementary Figure 8. Identification of CD271+ melanoma tumor initiating cells in Mel 327 groin lymph node metastatic melanoma patient. Flow cytometric contour plots demonstrating the variable expression of CD271. (i) Surgical sample (ii) Xenograft Pi (surgically removed patient's tumor expanded in-vitro and injected into Rag2gDKO mice); (iii) Xenograft Pi CD271+ (tumor induced after injection of 900 CD271+ cells into Rag2gDKO mice). (iv) table summarizing tumor transplantation assay: single cell suspension was obtained from xenograft Pi tumor by mechanical and enzymatic digestion and live, mLin- (H2-Kd-/ mCD45.2-/mTer119-), CD271+ and CD271- cells were isolated by FACS. Purified cells were mixed with matrigel and injected intradermaly into Rag2gDKO mice at indicated cell doses;

a



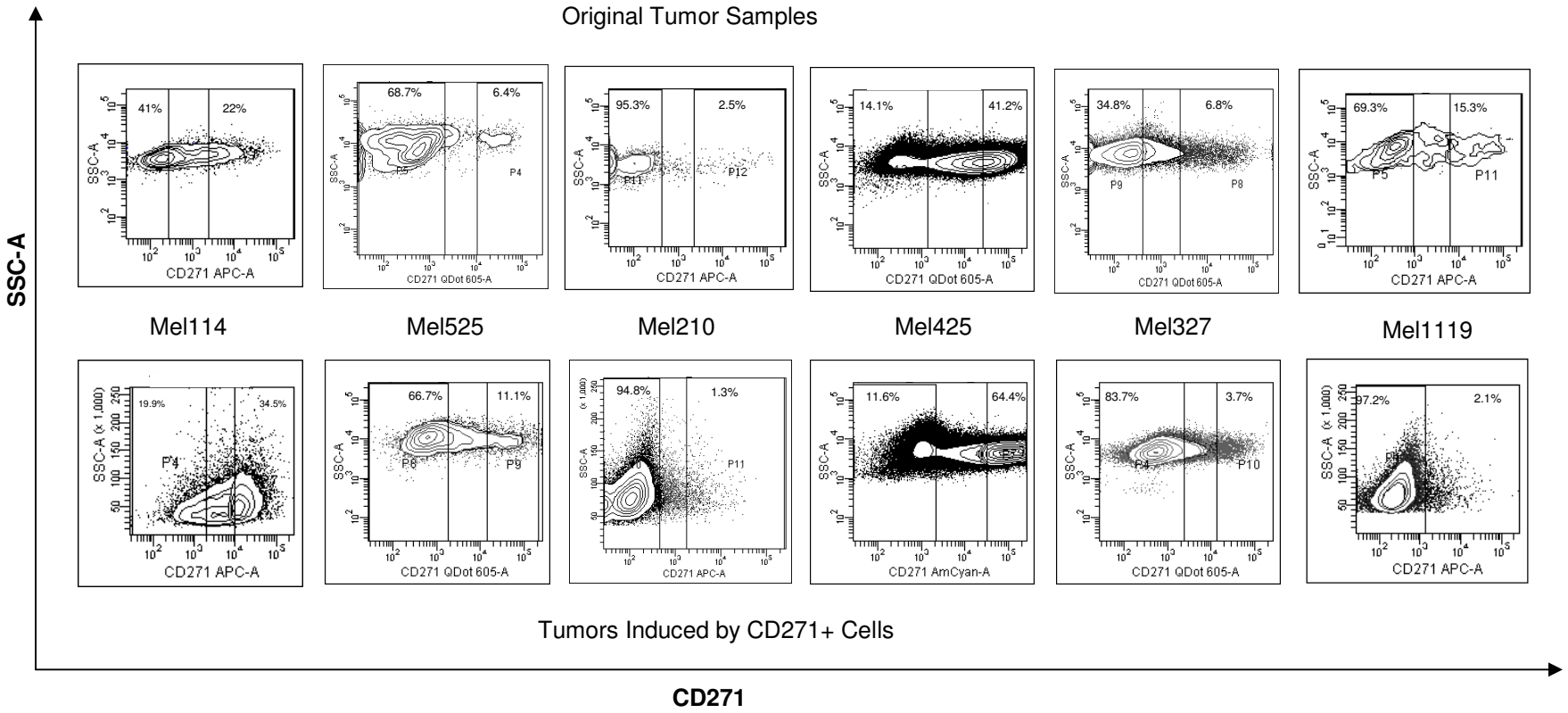
b



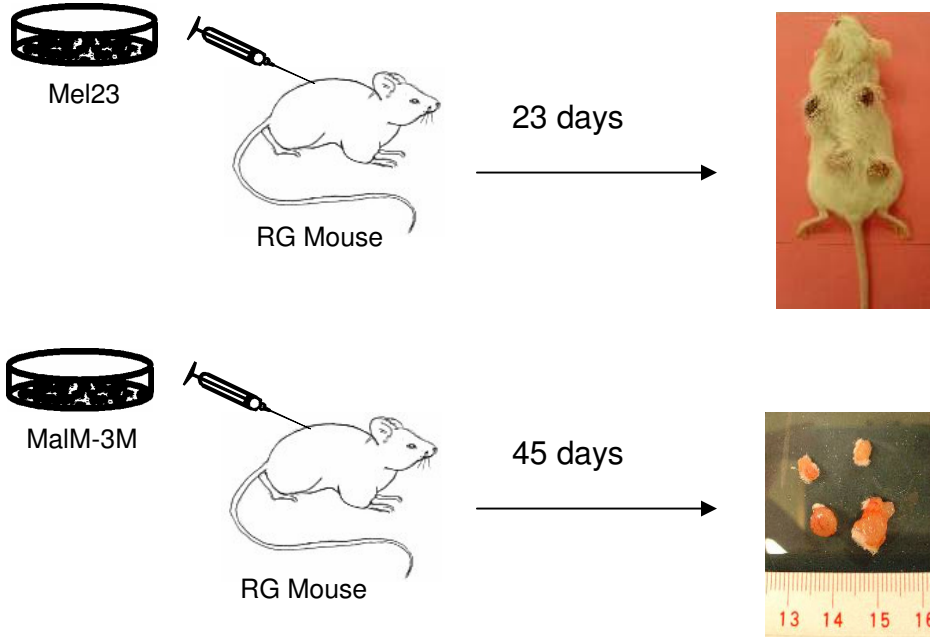
c

Mel-525	CD271 -	CD271 +
m525-Xenograft (P0)	0/2 (10^2) 0/2 (10^3) 0/2 ($5x10^3$)	0/2 (10^2) 0/2 (10^3) 2/2 ($5x10^3$)
M525-Xenograft (Pi)	0/2 (10^3) 1/2 ($2x10^3$) 1/2 ($5x10^3$)	0/2 (10^3) 2/2 ($2x10^3$) 2/2 ($5x10^3$)
Summary	2/12	6/12

Supplementary Figure 9. a, Flow cytometric contour plots demonstrating the variable expression of CD271 in Mel 525 neck lymph node metastatic melanoma patient. (i) Xenograft P0 (surgically removed patient's tumor implanted into Rag2gDKO mice) (ii) Xenograft P1 CD271+ (tumor induced after injection of $5x10^3$CD271+ cells into Rag2gDKO mice) (iii) Xenograft Pi (surgically removed patient's tumor expanded in-vitro and injected into Rag2gDKO mice); (iv) Xenograft Pi CD271+ (tumor induced after injection of $2.5x10^3$ CD271+ cells into Rag2gDKO mice). **b,** (i) Single cell suspension was isolated from xenograft P0 tumor by mechanical and enzymatic digestion and live, mLin- (H2-Kd-/ mCD45.2-/mTer119-), CD271+ and CD271- cells were isolated by FACS. $5x10^3$ cells were mixed with matrigel and injected intradermaly into Rag2gDKO mice; only CD271+ cells gave rise to tumors after 14 weeks. (ii) Single cell suspension was isolated from xenograft Pi tumor by mechanical and enzymatic digestion and live, mLin- (H2-Kd-/ mCD45.2-/mTer119-), CD271+ and CD271- cells were isolated by FACS. 2.5x103 cells were mixed with matrigel and injected intradermaly into Rag2gDKO mice; tumors became notable after 8 weeks. **c,** Summary table of all injected cell doses and tumor frequency formation induced by CD271+ and CD271- cell populations isolated from Mel525.



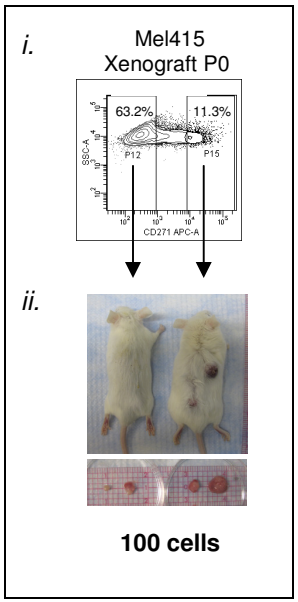
Supplementary Figure 10. Differentiation of CD271+ cells during tumor formation in Rag2gKO mice. Flow cytometric contour plots demonstrating the variable expression of CD271 in tumors re-established by injection of C271+ cells. Upper panel: original melanoma sample from which CD271+ cells were isolated; lower pane:l tumors induced after injection of CD271+ cells. Single cell suspensions were prepared from surgical or xenografted samples and live, hLin- (CD45, CD2, CD3, CD31) or mLin- (H2-Kd-/ mCD45.2-/mTer119-) cells analyzed on BDFACSAria instrument.



	Engraftment Frequency					
Cell Dose	10	50	$5x10^2$	$5x10^3$	$5x10^4$	$5x10^5$
Mel23	3/3	4/4	4/4	4/4	4/4	4/4
MalM-3M	2/3	3/4	4/4	4/4	4/4	4/4

Supplementary Figure 11. Tumorigenic engraftment frequencies of bulk tumor cells representing two established melanoma cell lines Mel23 and Malm-3M. Cells were cultured in RPMI 1640 Media supplemented with 10% fetal bovine serum, 0.5% gentamicin and 1% L-glutamine in a humidified atmosphere Incubator with 5% CO2 at 37℃ and were passaged twice each week. Cells were trypsinized, mixed with Trypan blue Stain (0.4%) and live cells were counted in hemocytometer. Indicated number of cells were mixed at 2:1 ratio with Matrigel in 100 µl volume and injected intradermaly on the flank of 6-8 weeks old RG mice anesthetized with isoflurane-O2.

a



b

Mel-415	CD271	
	-	+
M415 -Xeno (P0)	2/2 (100) small nodules 2/2 (1,000) 2/2 (5,000)	2/2 (100) 2/2 (1,000) 2/2 (5,000)
M415 -Xeno (P1)	0/4 (1) 1/8 (10) small nodule 3/4 (100) small tumors	0/4 (1) 5/8 (10) 3/4 (100)
Summary	13/26	17/26

Supplementary Figure 12. Identification of CD271+ melanoma tumor initiating cells in Mel 415 H&N melanoma patient. **a,** (i) Flow cytometric contour plots demonstrating the variable expression of CD271 in Mel415 xenograft P0 (surgically removed patient's tumor was implanted subcutaneously on the back of Rag2gDKO mice prior to cell isolation; (ii) single cell suspension was obtained by mechanical and enzymatic digestion and live, mLin- (H2-Kd-/ mCD45.2-/mTer119-), CD271+ and CD271- cells were isolated by FACS. Purified cells were mixed with matrigel and injected intradermaly into Rag2gDKO mice; representative cell dose 100 is shown. **b,** Summary table of all injected cell doses and tumor frequency formation induced by CD271+, and CD271- cell populations isolated from Mel415 Xeno P0.

a

Cell Dose	10^3		10^4		$2x10^4$ - $7x10^4$	
Mouse Strain	RG	NOG	RG	NOG	RG	NOG
Melanoma tumor cells	3/3	3/3	3/3	3/3	2/2	2/2
Bladder tumor cells	6/6	6/6	3/3	3/3	3/3	3/3

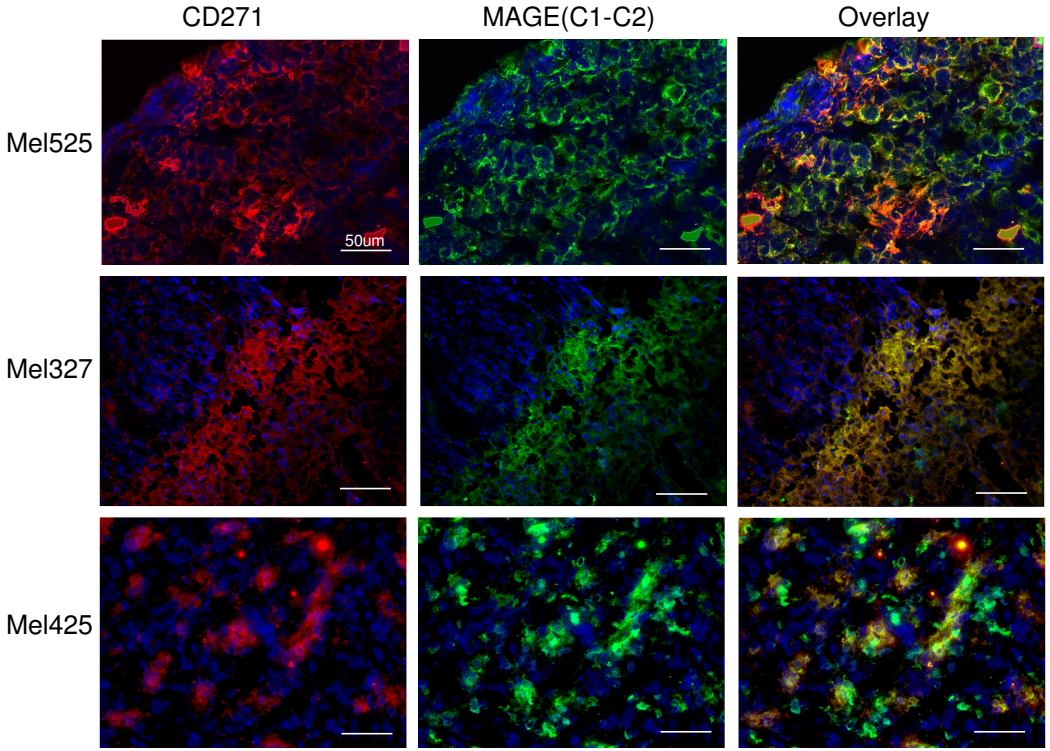
Total: RG mice: 16/16 (melanoma tumors); 24/24 (bladder tumors)
NOG mice: 16/16 (melanoma tumors); 24/24 (bladder tumors)

b

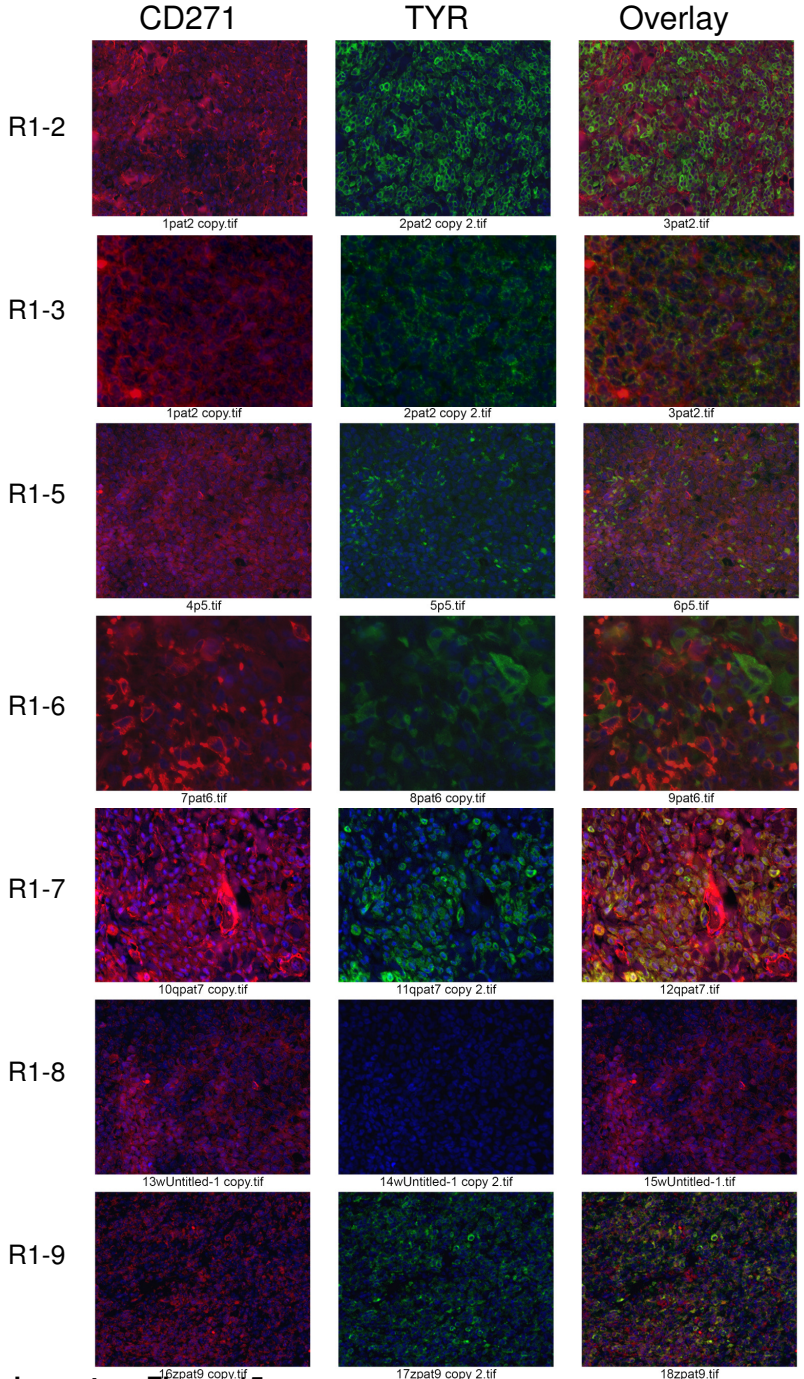
Cell Dose	10		100		10^3		$2X10^3$		$5X10^3$	
Mouse Strain	RG	NOG	RG	NOG	RG	NOG	RG	NOG	RG	NOG
CD271 Sorted Melanoma tumor cells	2/3	1/3	2/3	3/3	3/3	3/3	3/3	3/3	2/2	1/1

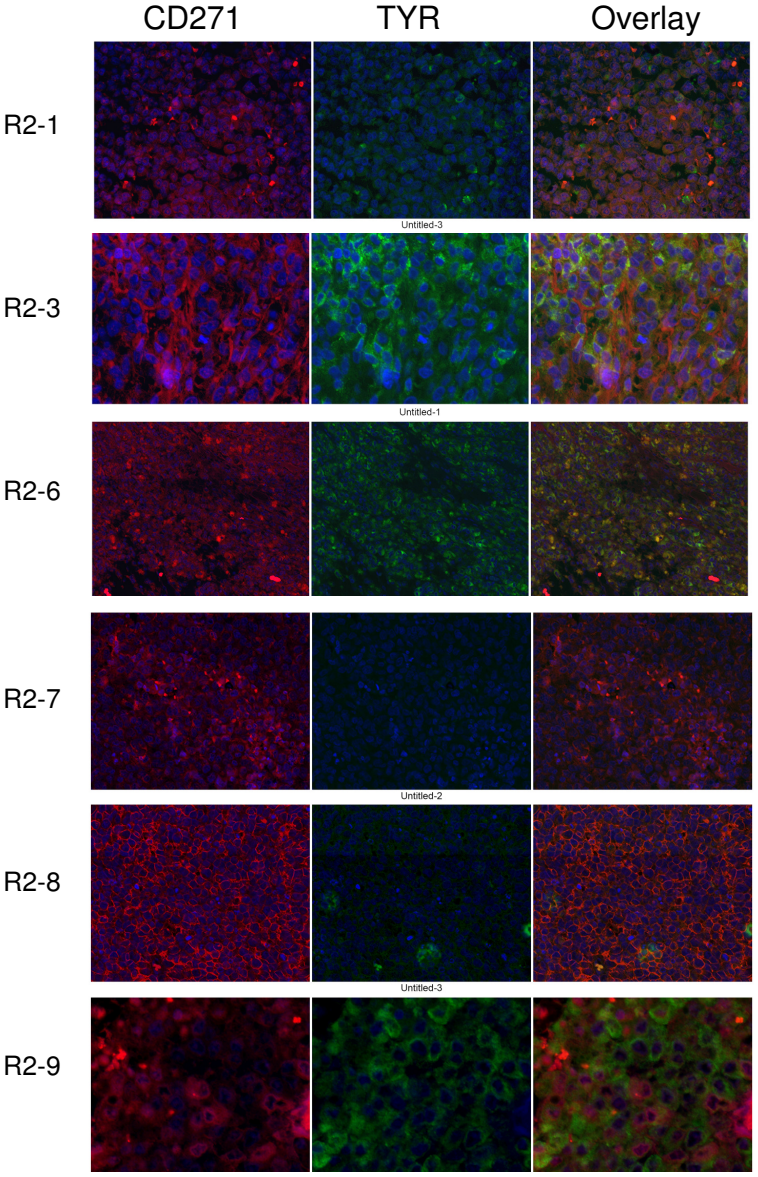
Total RG mice: 12/14
NOG mice: 11/13

Supplementary Figure 13. RG and NOG mice engraft tumor cells at equal efficiency during transplantation assays.
a, RG and NOG mice were injected with live, Lin- (H2-Kd-/mCD45-/mTer119-) melanoma or bladder cells isolated by FACS and mixed with matrigel; numbers indicate ratio of tumor incidence relative to the number of injections. **b,** CD271+ melanoma cells isolated by FACS (live, Lin- (H2-Kd-/mCD45-/mTer119-) and mixed with matrigel; numbers indicate ratio of tumor incidence relative to the number of injections

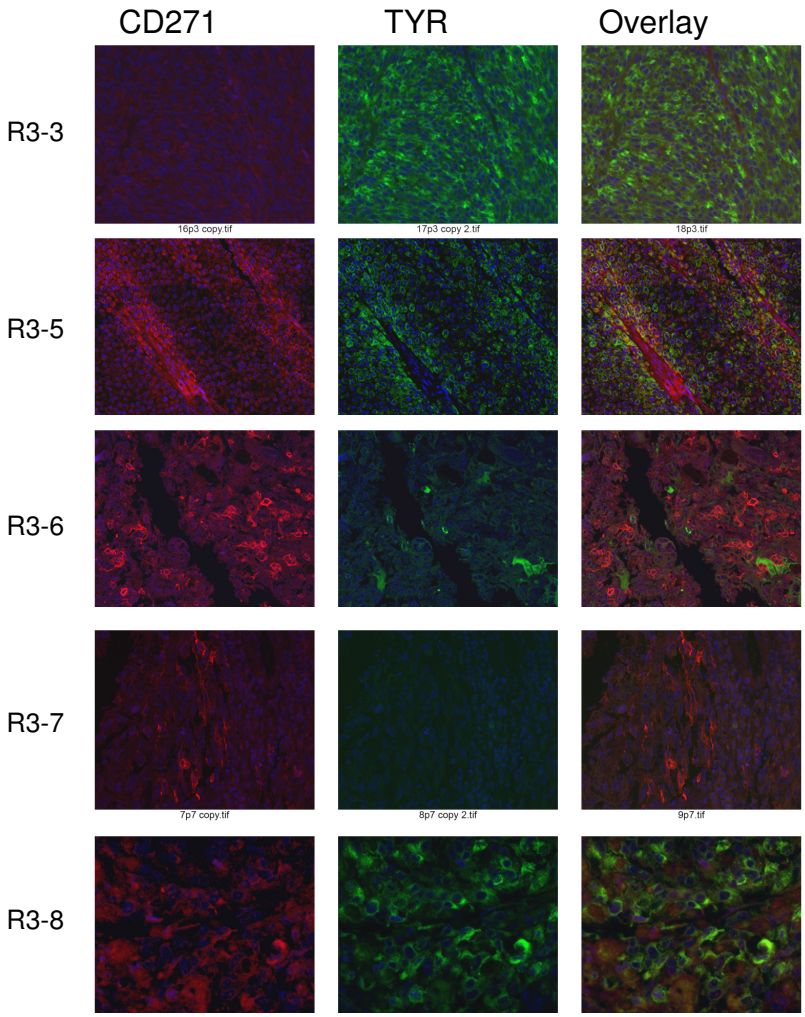


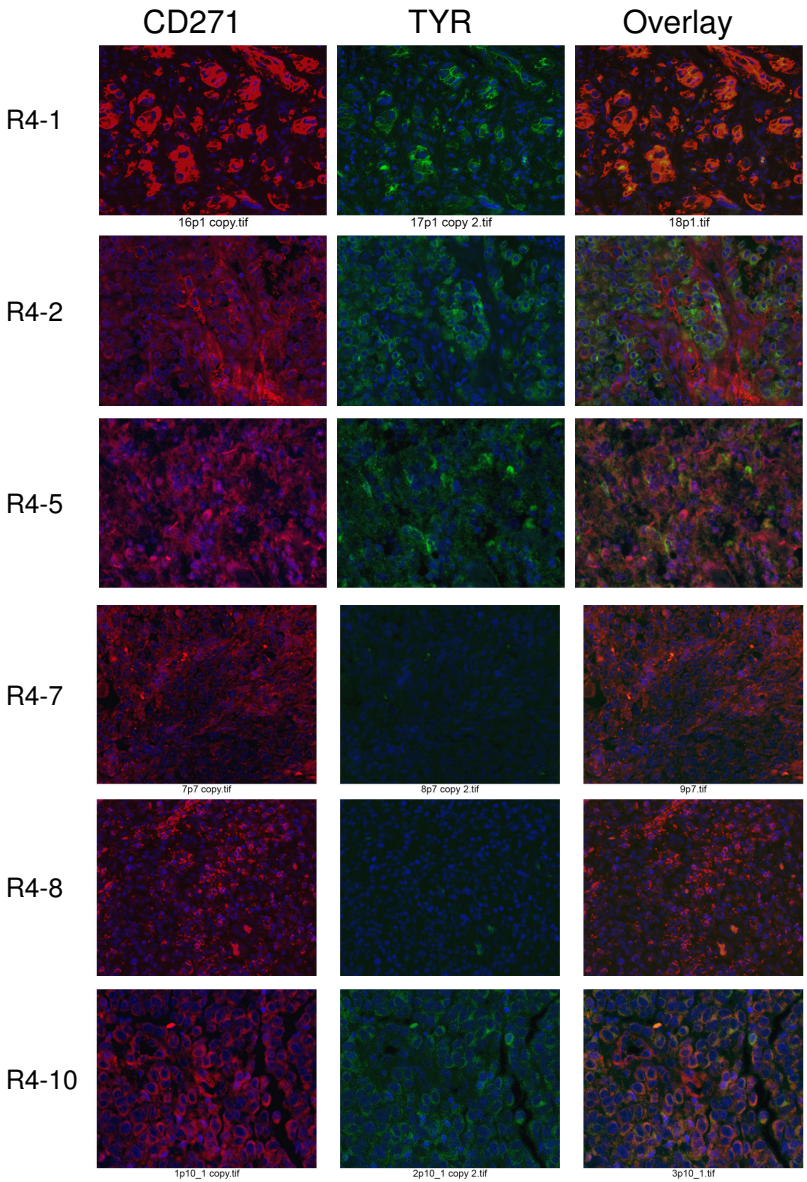
Supplementary Figure 14. Immunofluorescent analysis of CD271and MAGE(C1-C2) expression in tissue sections of melanoma patients . Tissue sections were stained with antibodies recognizing CD271 and MAGE(C1-C2) followed by AlexaFluor 594/488 secondary antibodies. Nuclei were visualized with Hoechst 33342. Pictures were taken under 40x objective and scale bars are equal to 50um.





Supplementary Figure 15.





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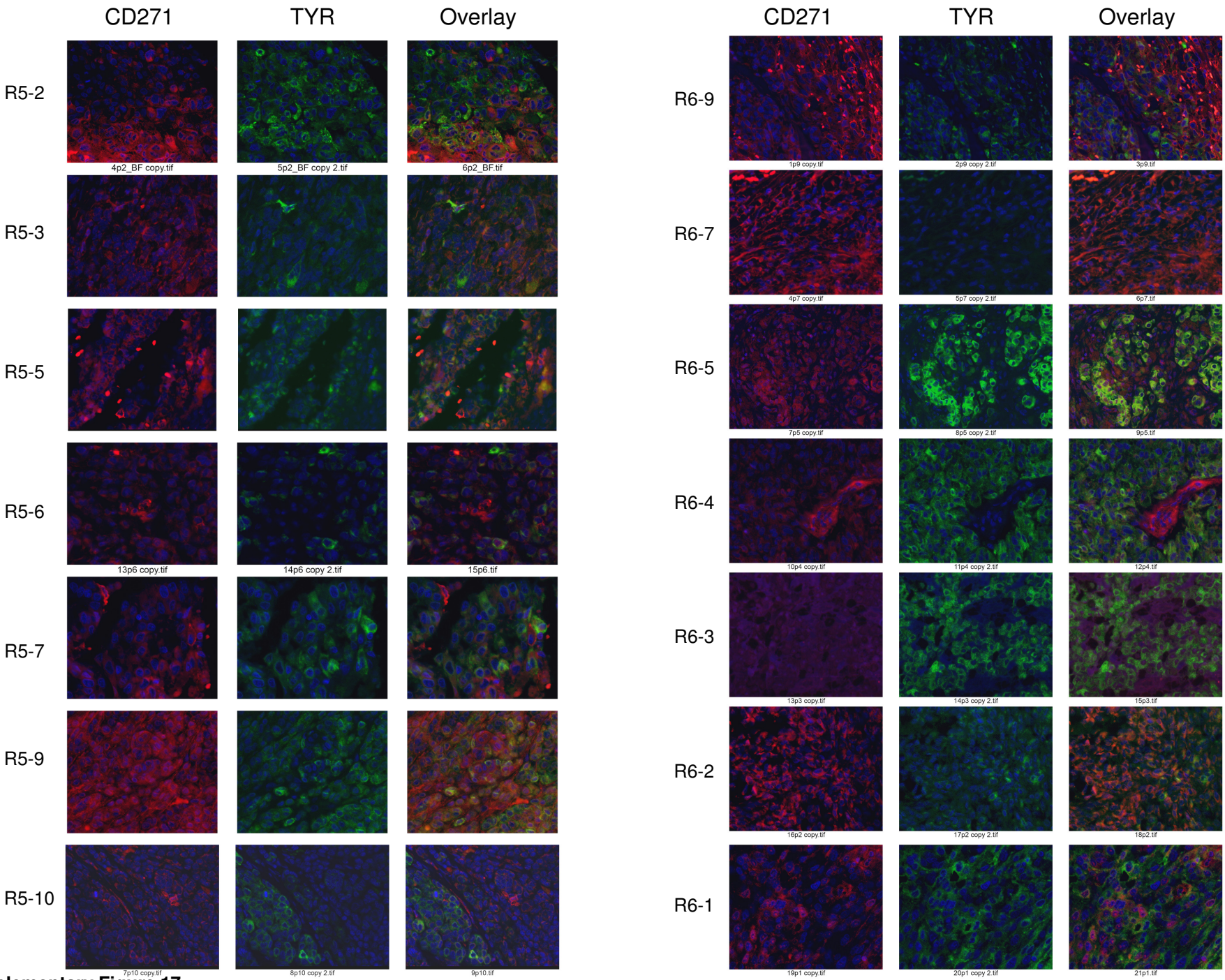
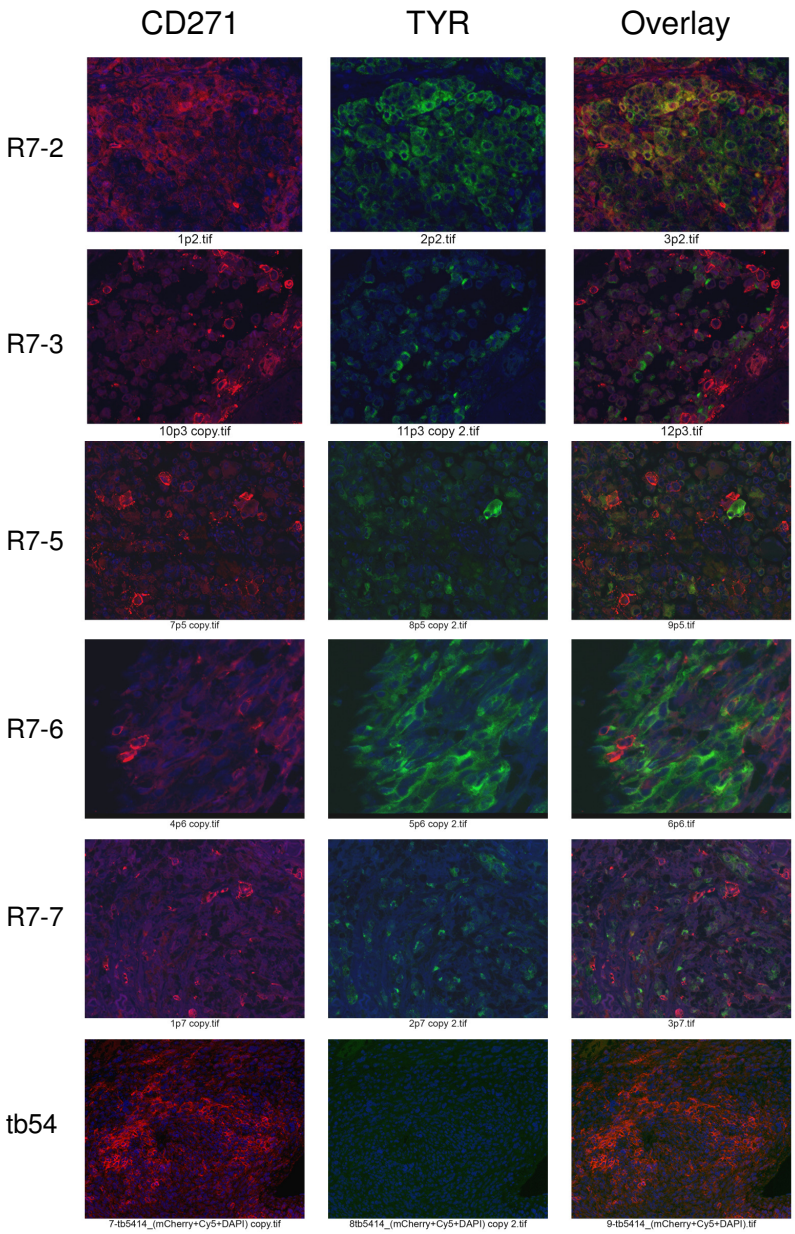
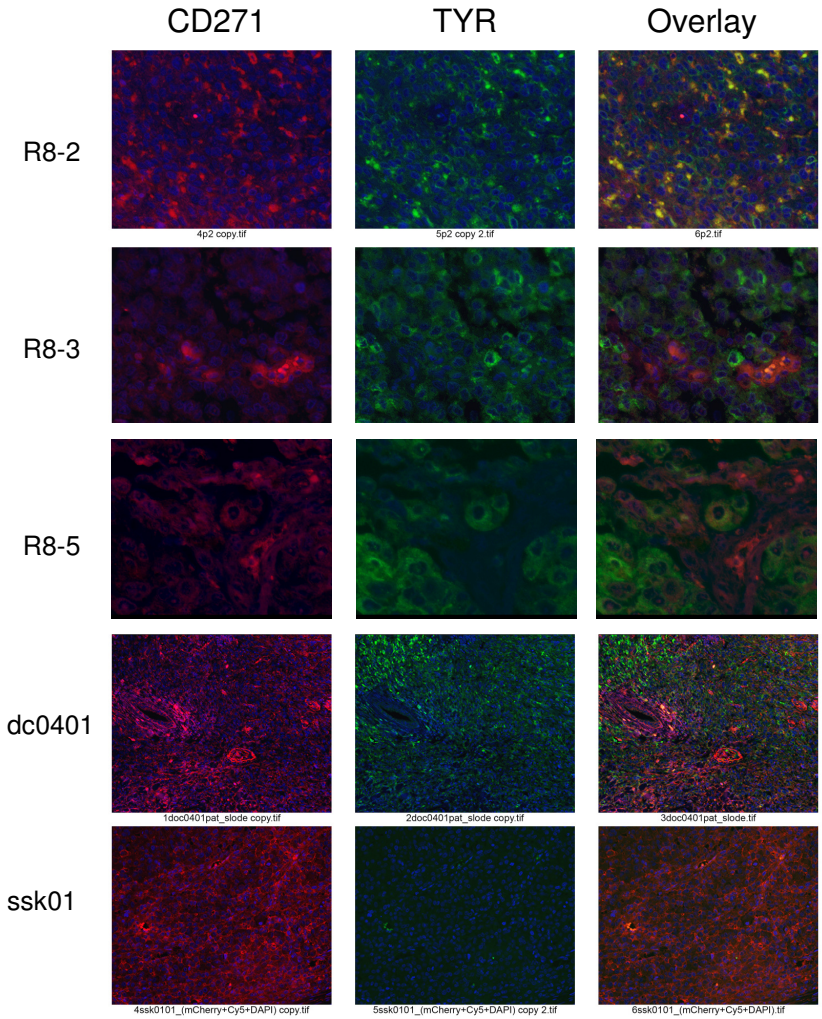
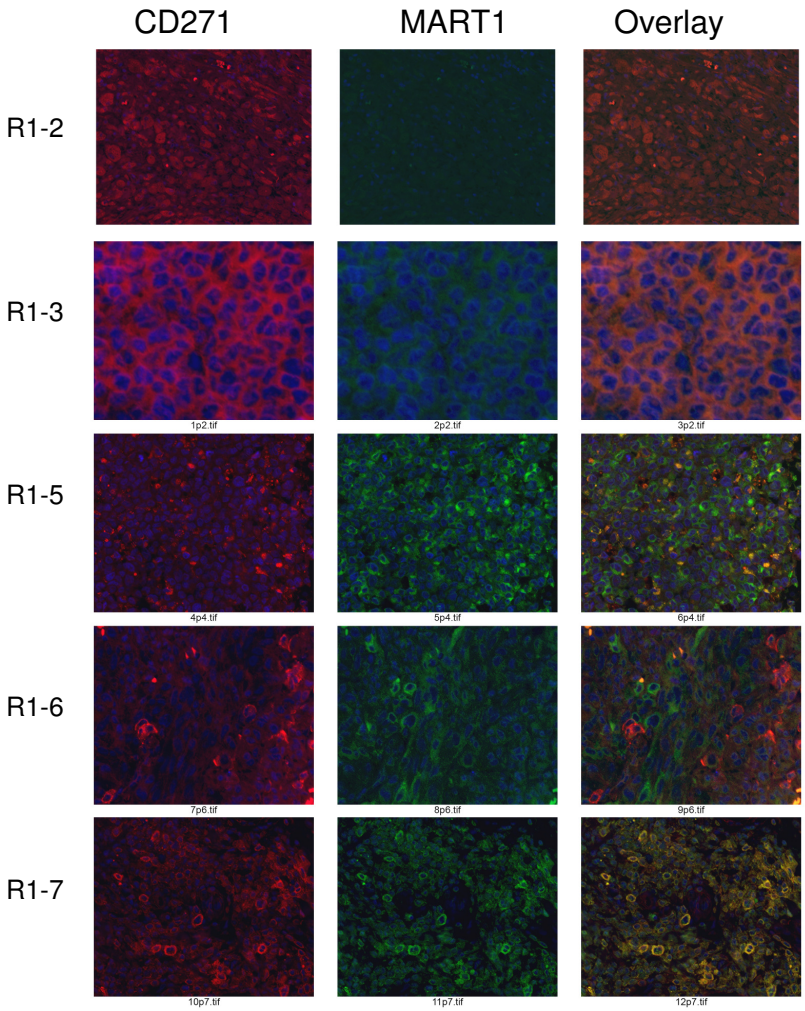


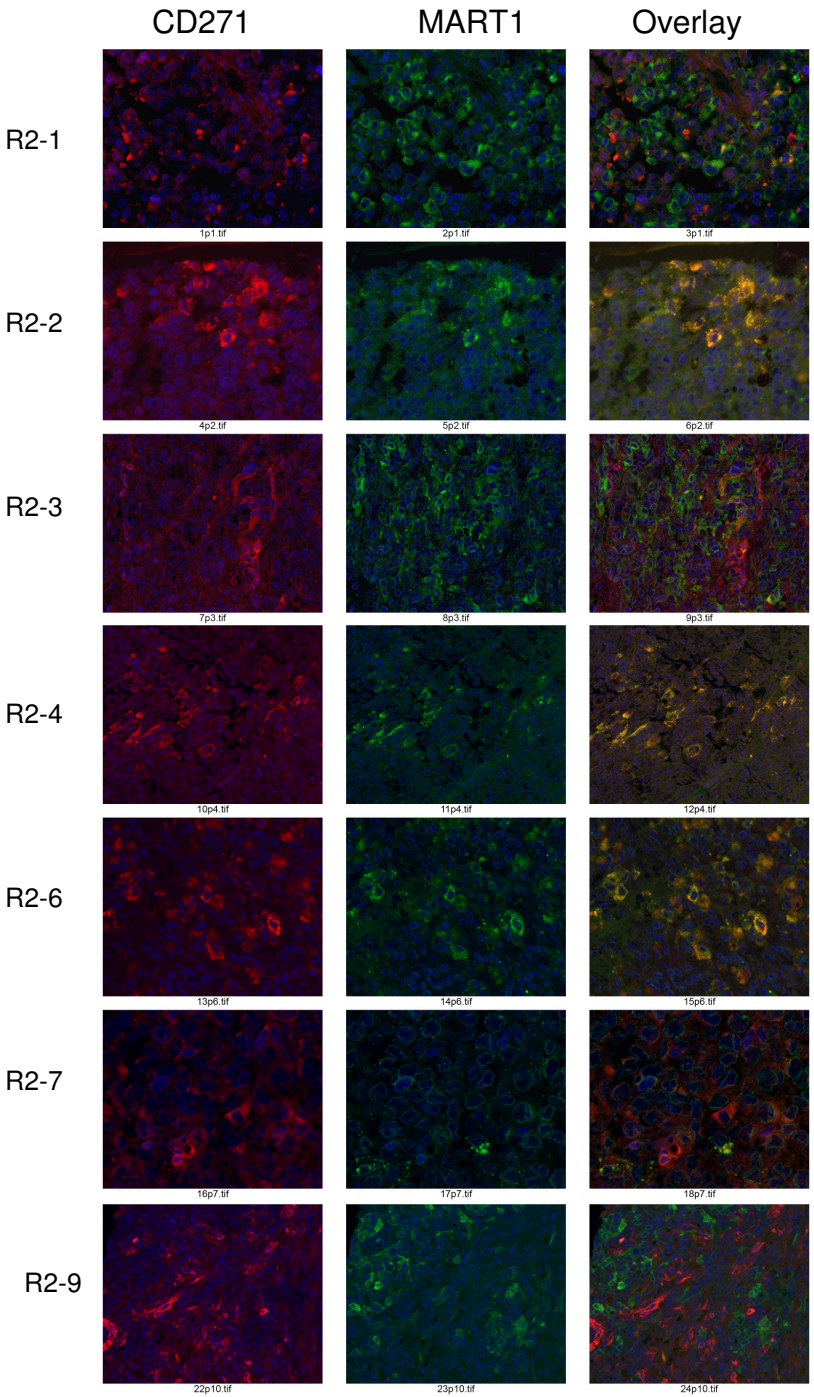
Supplementary Figure 17.





Supplementary Figure 18.





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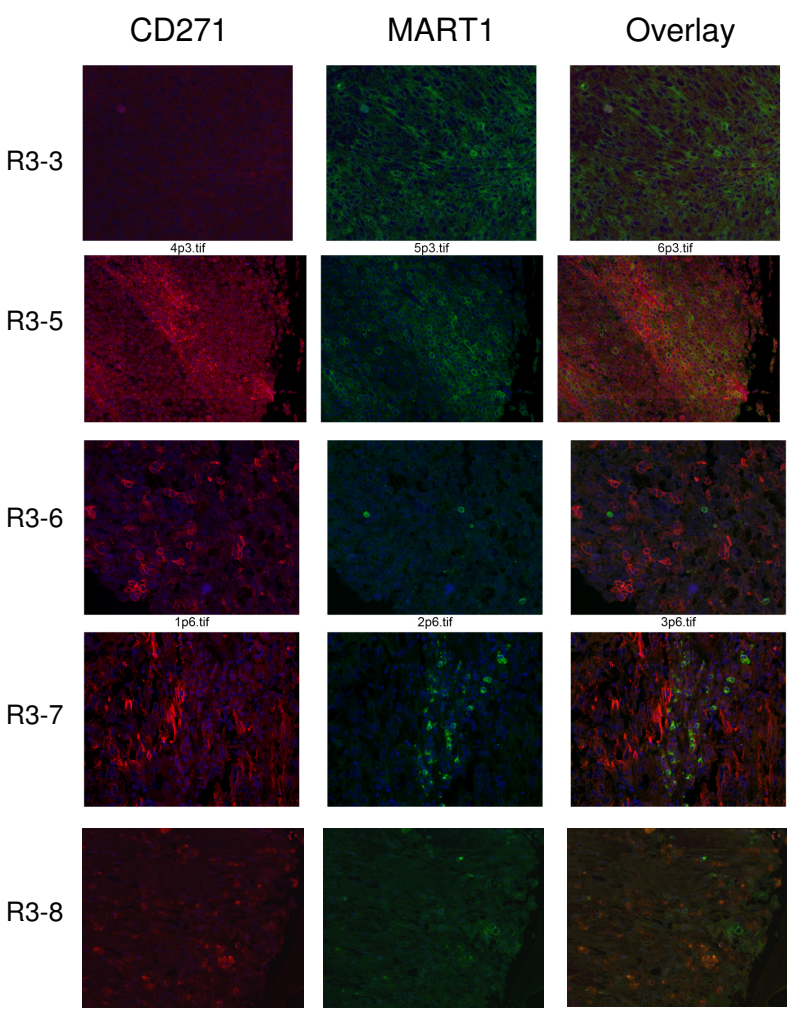

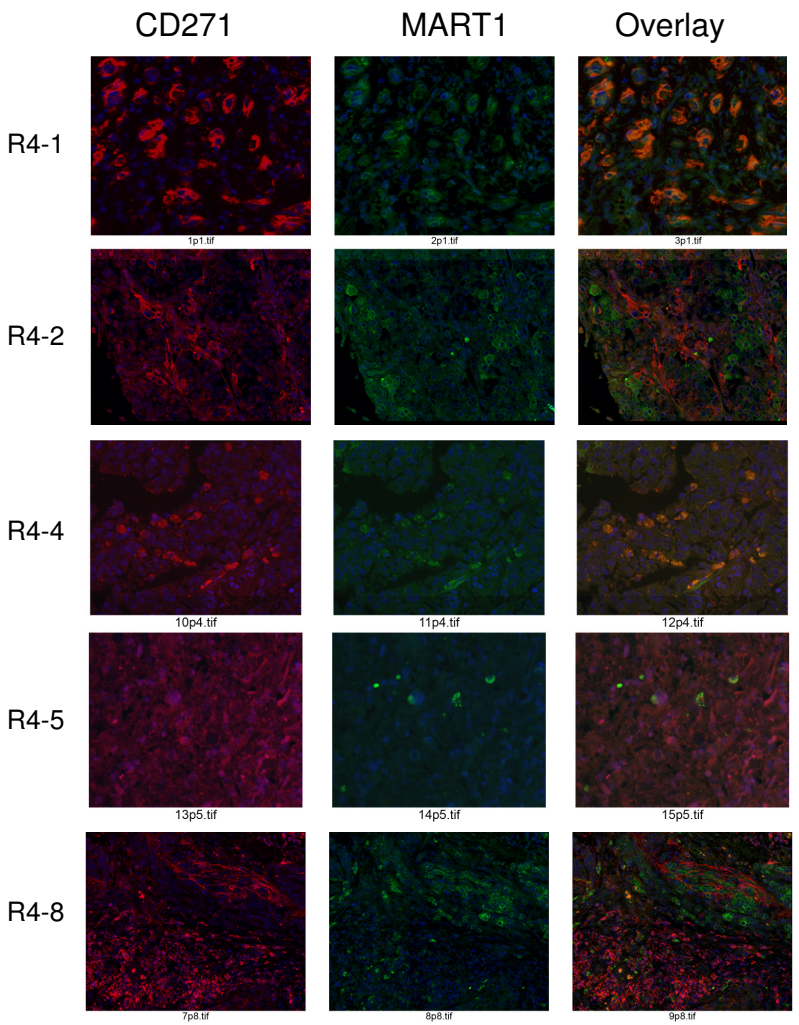

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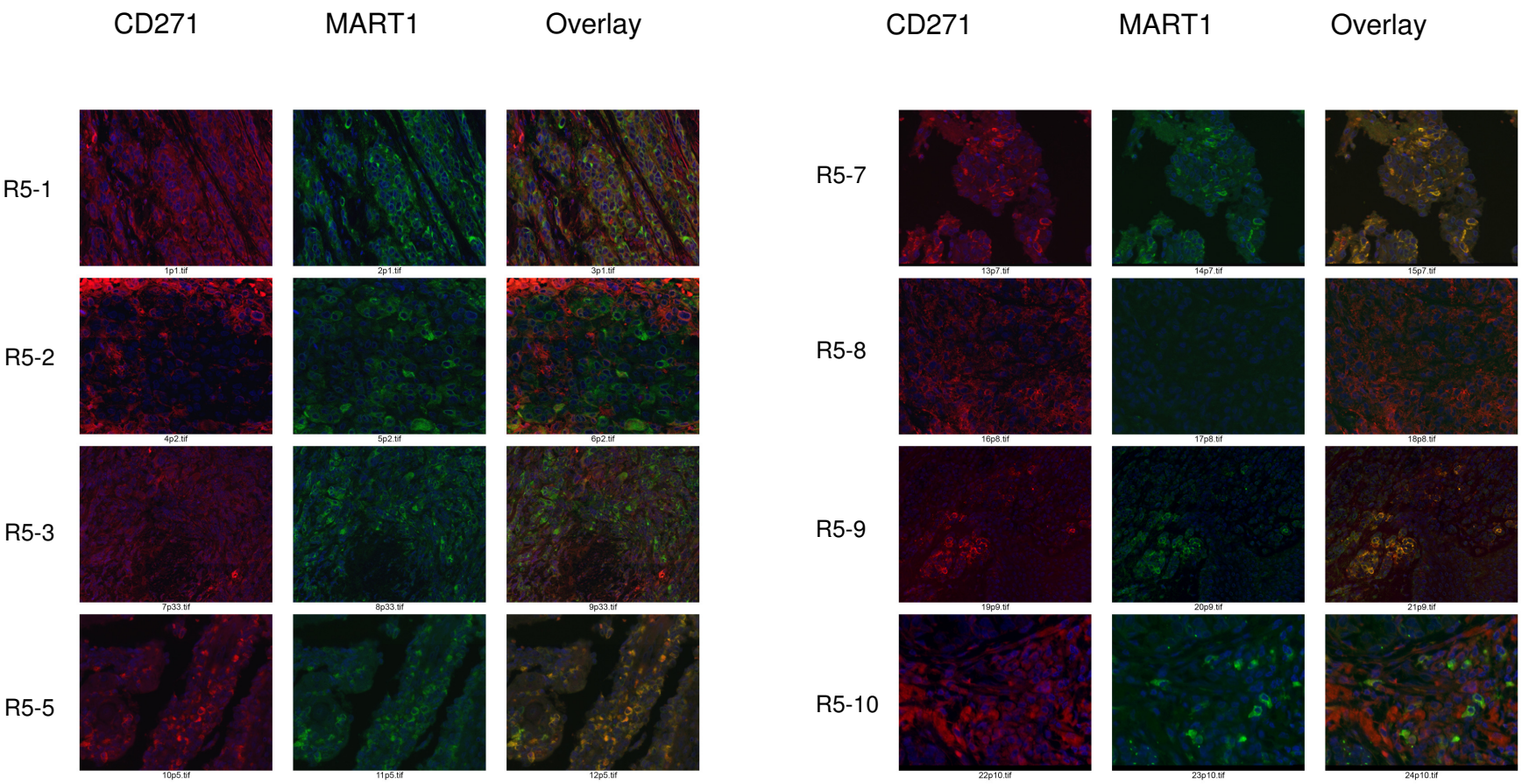
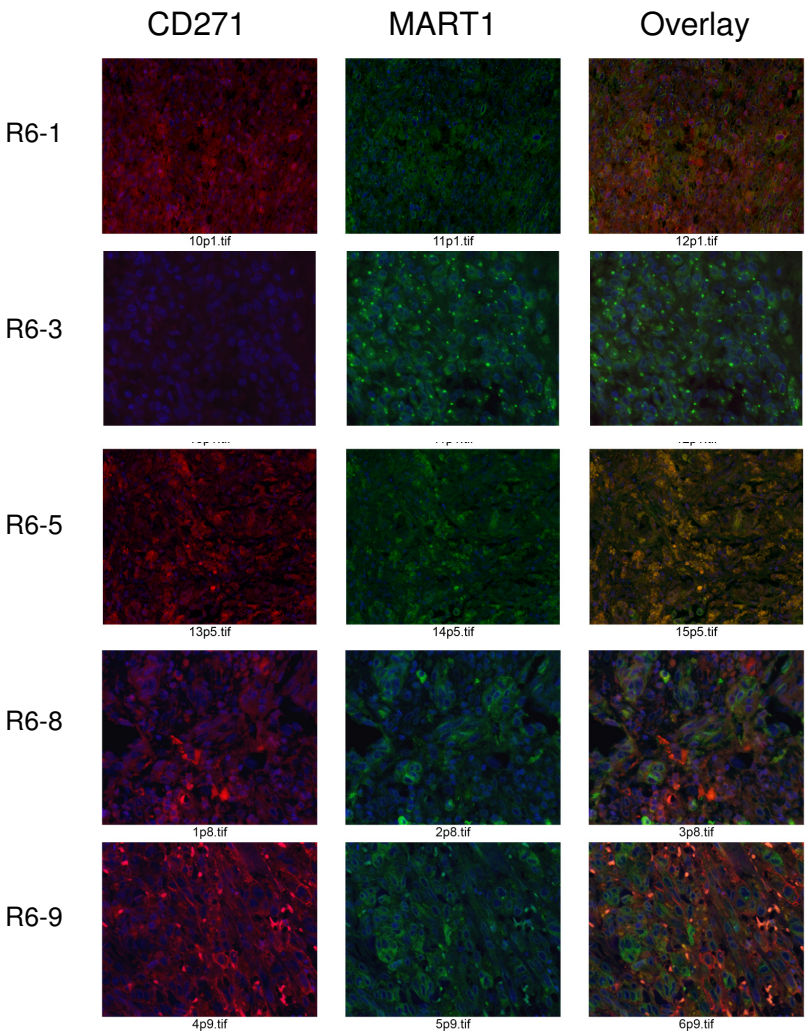
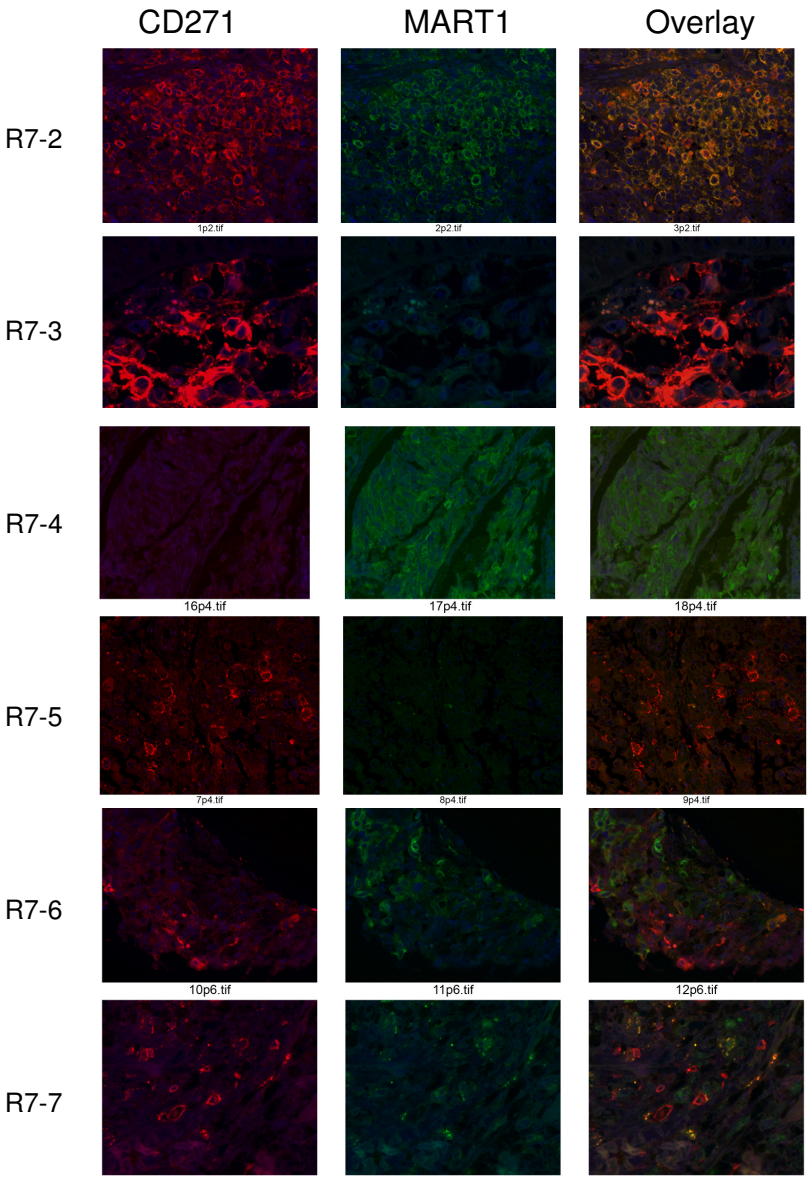


Supplementary Figure 21.





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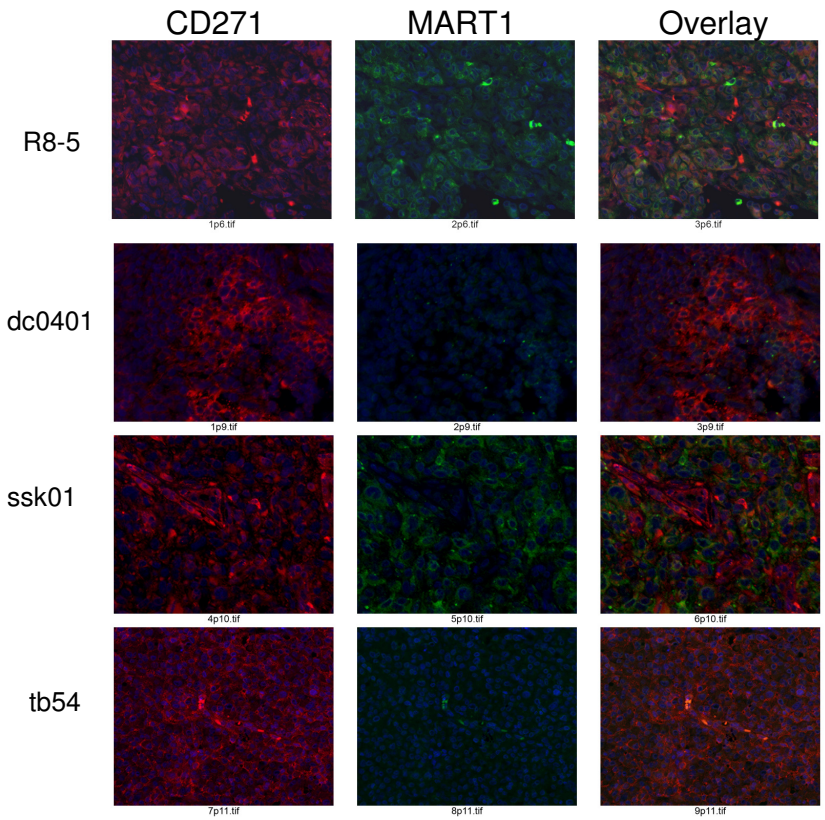

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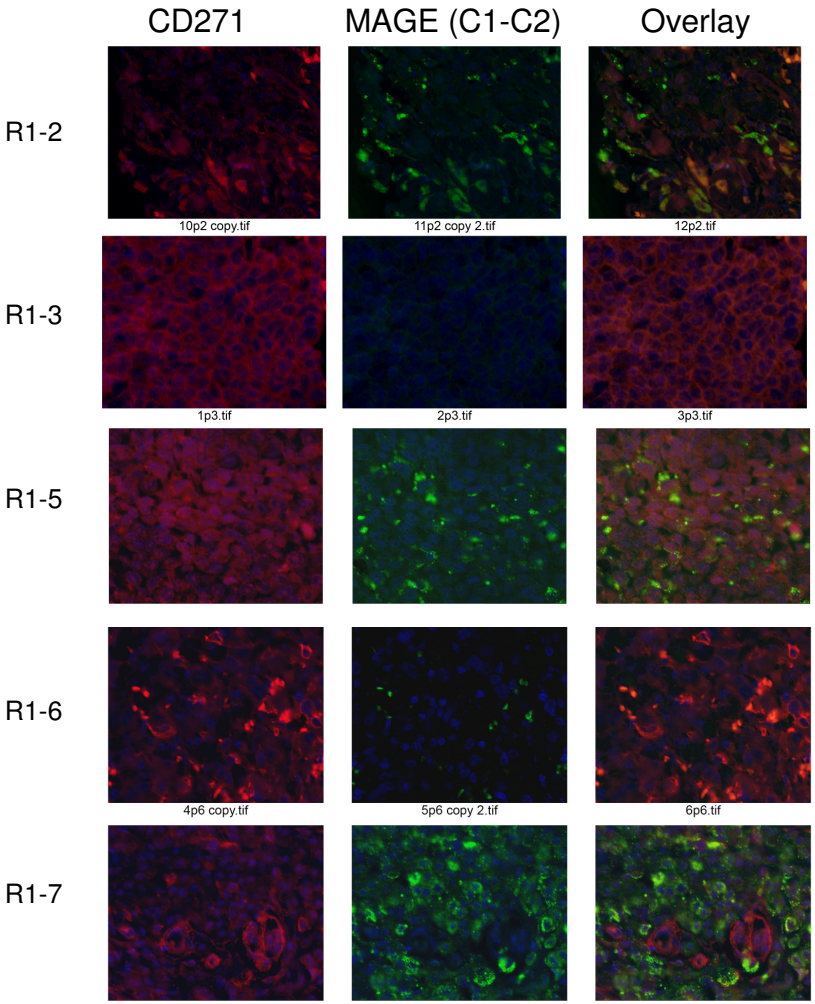

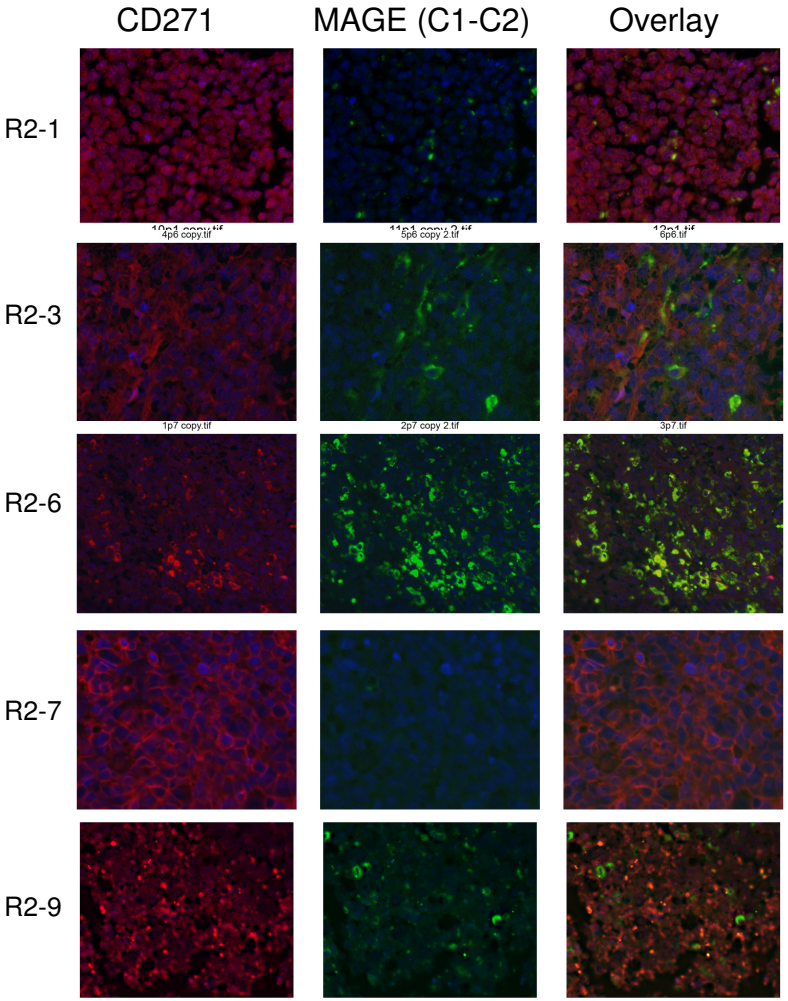

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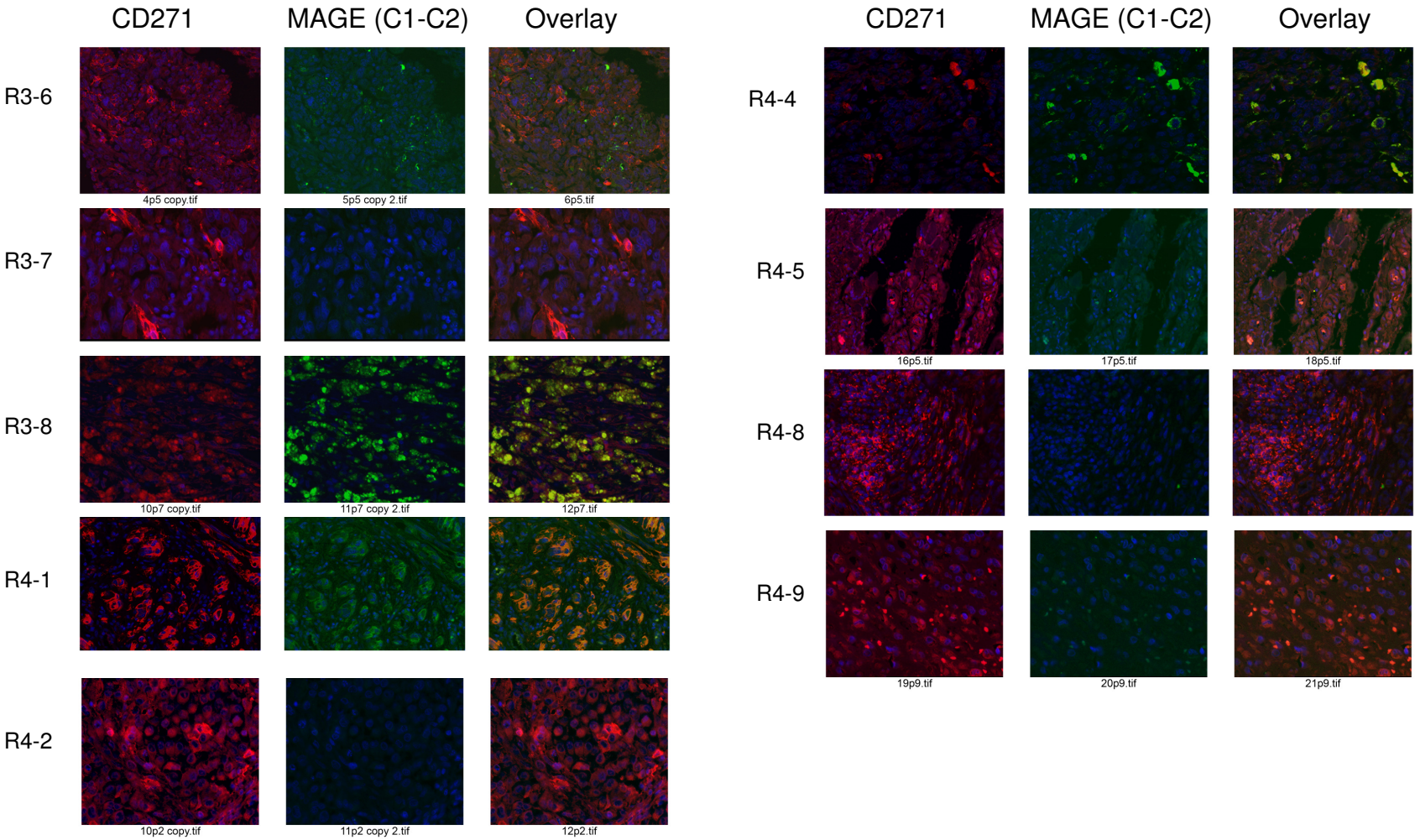

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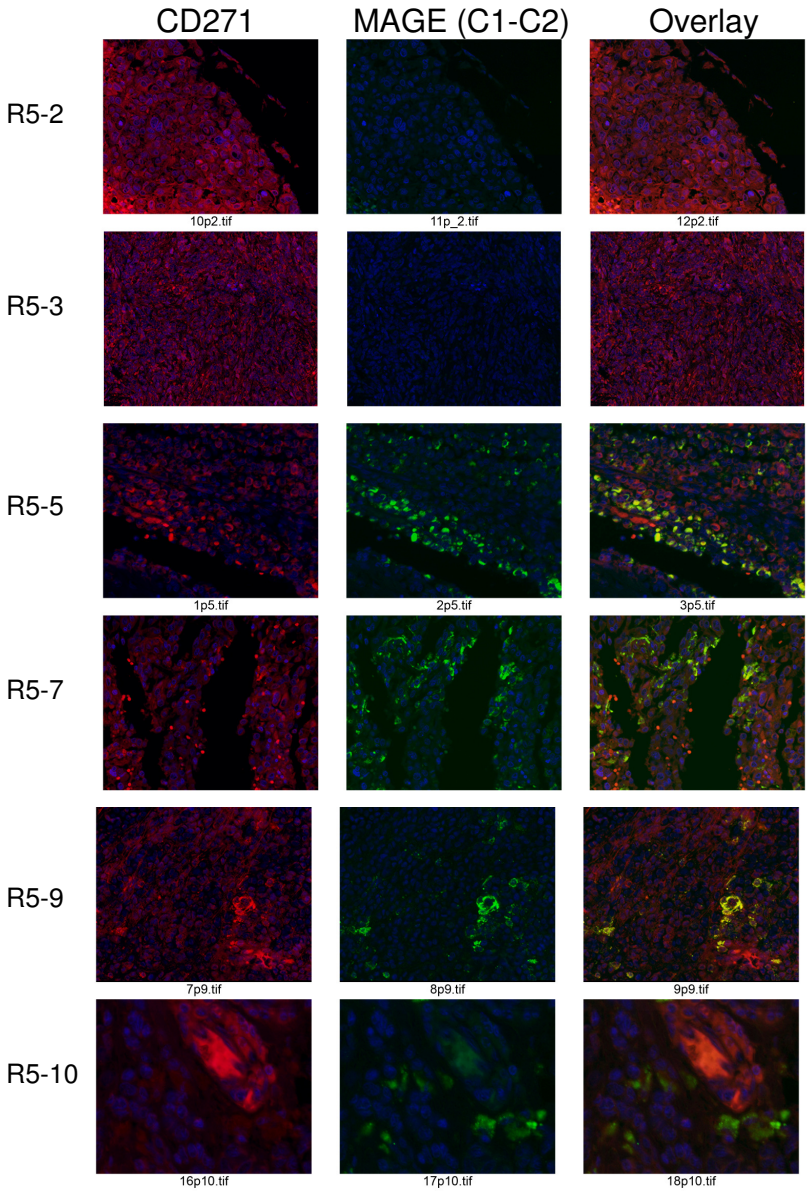

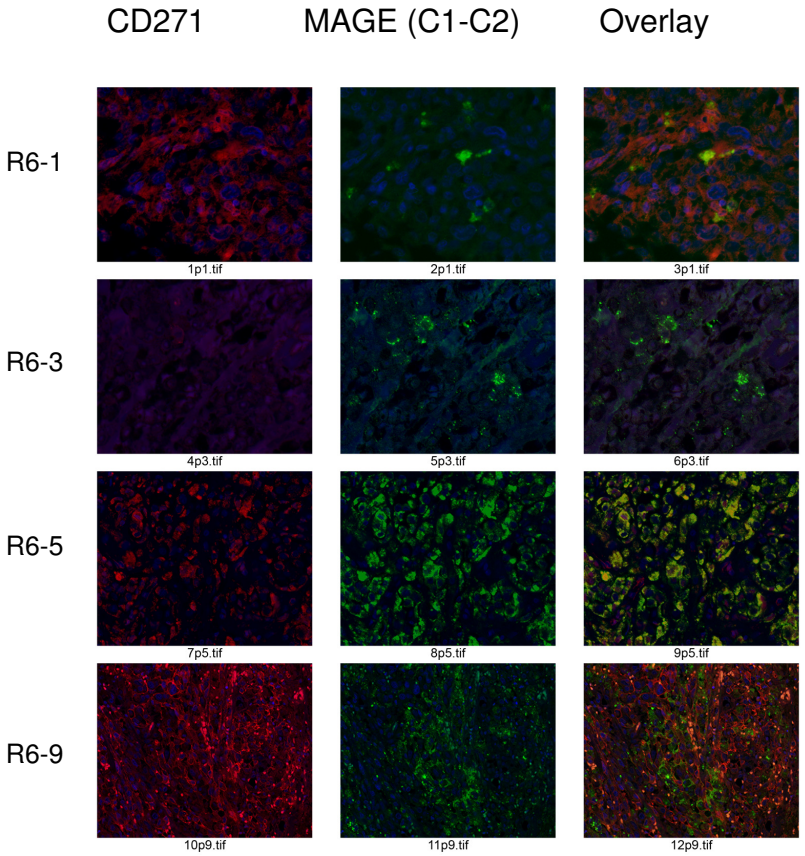

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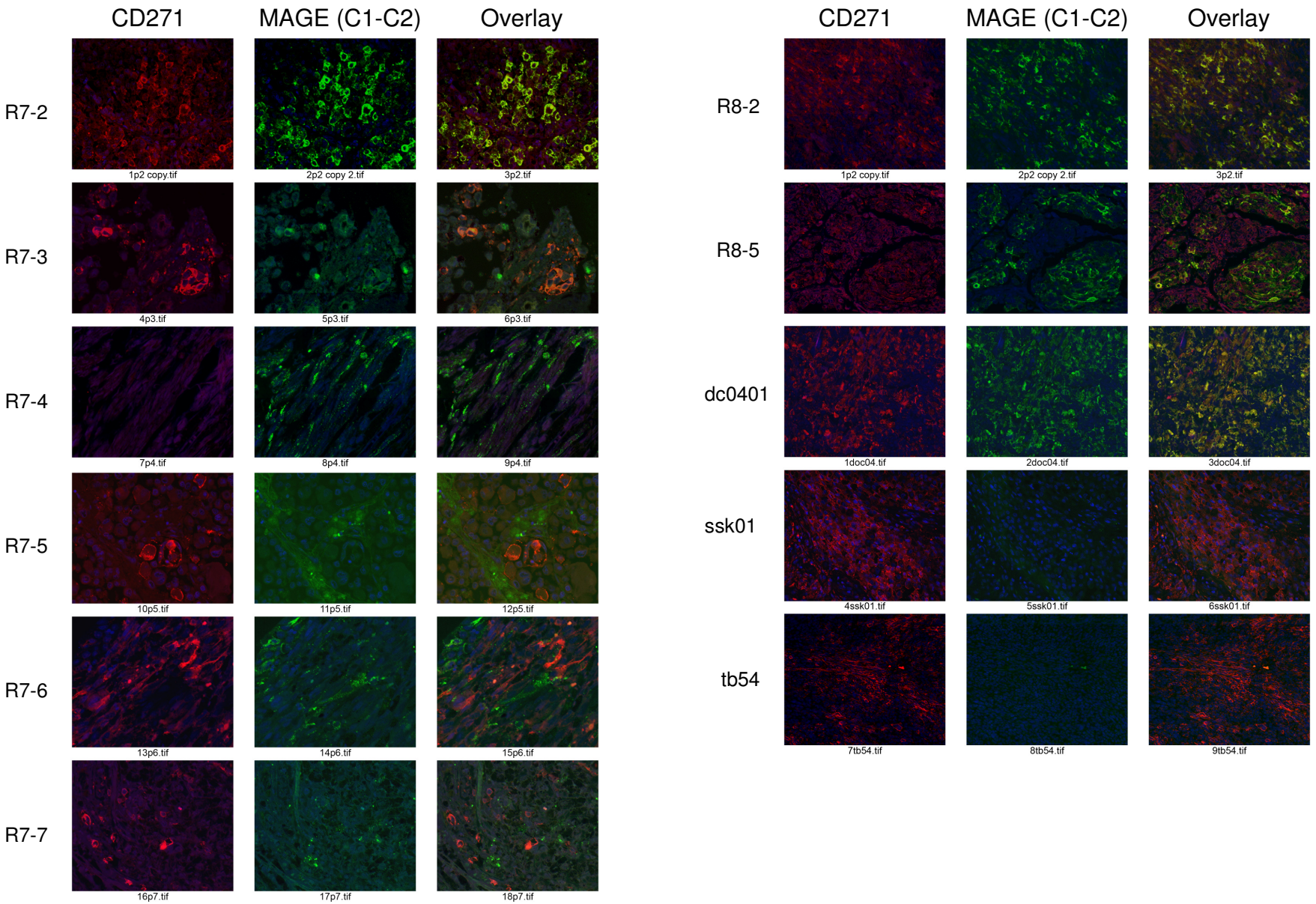
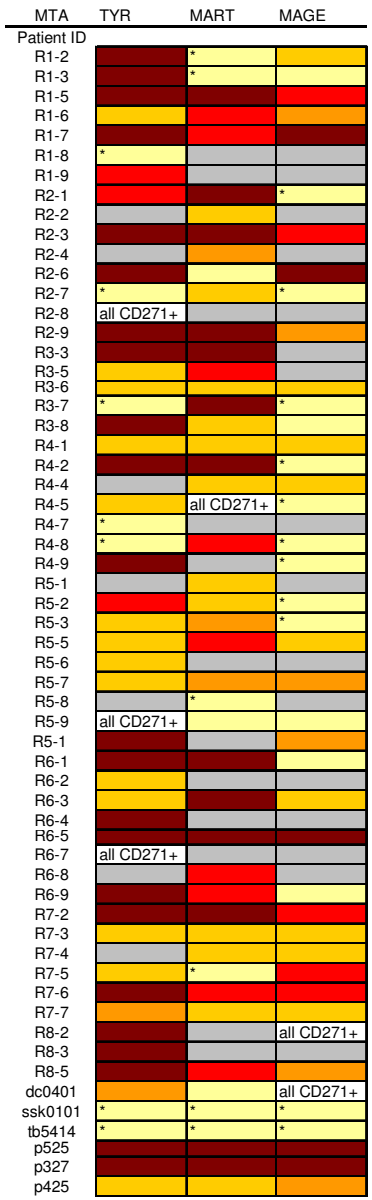
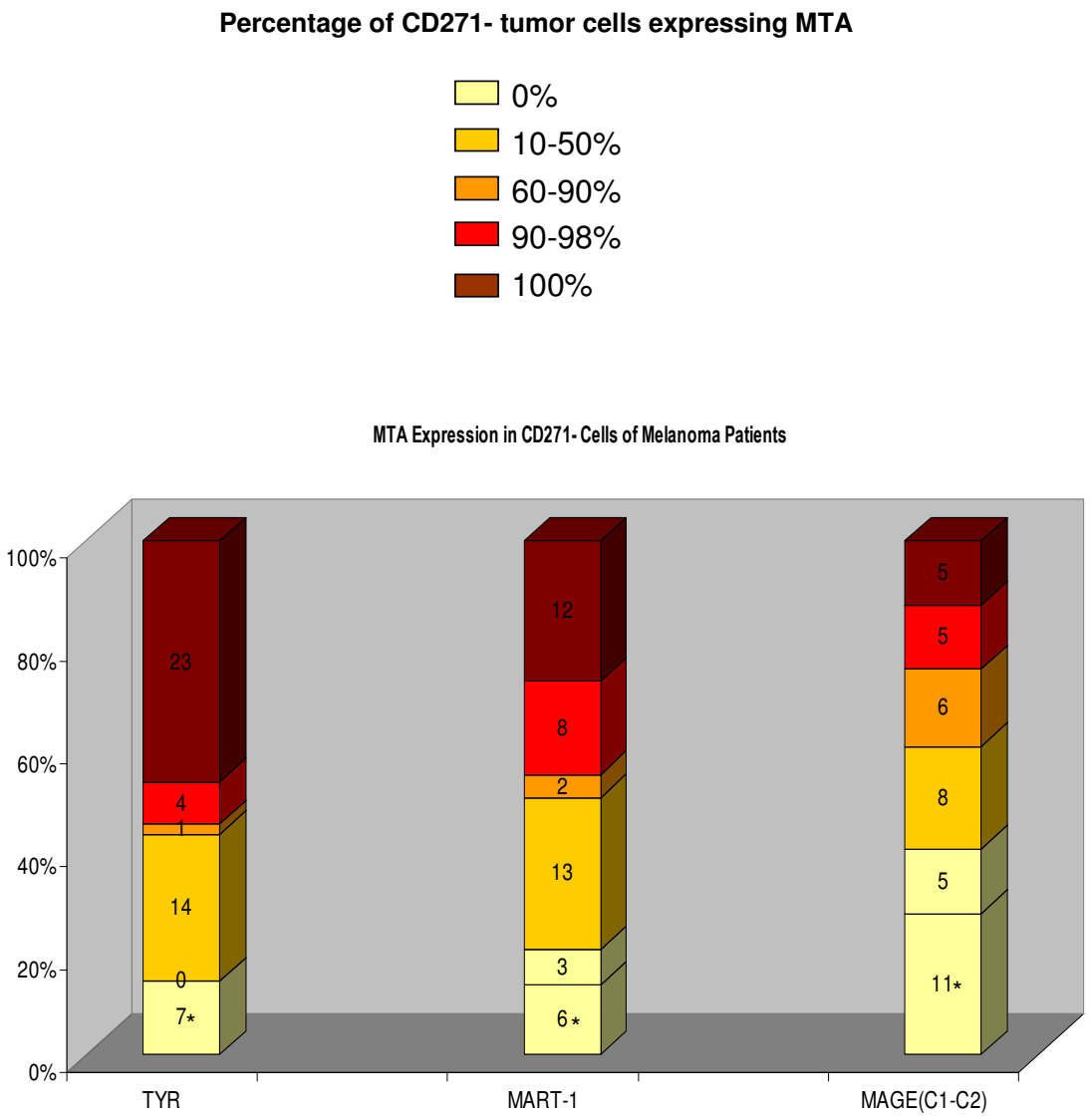


Supplementary Figure 27.

a





Supplementary Figure 28. a, Table indicating expression of melanoma tumor antigens (MTAs) TYR, MART1 and MAGE C1-C2 in CD271- cells of each patient's tumor. Color bars indicate percentage range of CD271- cells that expressed MTA; grey bars indicate that expression of CD271 and MTA was not detected during analysis of the tumor core. **b**, Stacked bar graph indicating proportion of melanoma patients with ranges of MTA positivity of CD271- cells.

Supplementary Methods

Antibody list used in this study

Mab Anti-	Conjugate	Company	Catalog ID
CD117	PE	BD	340867
CD133	APC	MACS	130090826
CD166	BIO	RD	6561
CD271	A647	BD	560326
CD271	BIO	BD	557195
EGFR	PE	BD	555997
CD51/61	FITC	BD	555505
CD49f	FITC	BD	555735
CD49d	APC	BD	559881
CD90	FITC	BD	555595
CD95	PE	BD	555674
CD44	PE	BD	550989
CD47	PE	BD	556046
CD105	APC	EB	17105773
CD146	PE	BD	550315
CD56	PE-Cy7	BD	57747
CD71	BIO	BD	557416
CD24	FITC	BD	555427
EpCAM	A647	BL	324212
FGF-R	APC	RD	684A
CXCR-4	APC	RD	170A
FZD-7	BIO	RD	1981
CD45	PB	INV	4528
CD31	PB	BL	303114
CD2	Unc	BD	555324
CD3	Unc	BD	555329
TER119	FITC	BD	557915
CD45.2	PB	BL	109820
H2-Kd	FITC	BD	553565

(BD) - BD Pharmigen
(RD) – R&D Systems
(MACS) – Myltenyi
(BL) – Biolegend
(INV) – Invitrogen