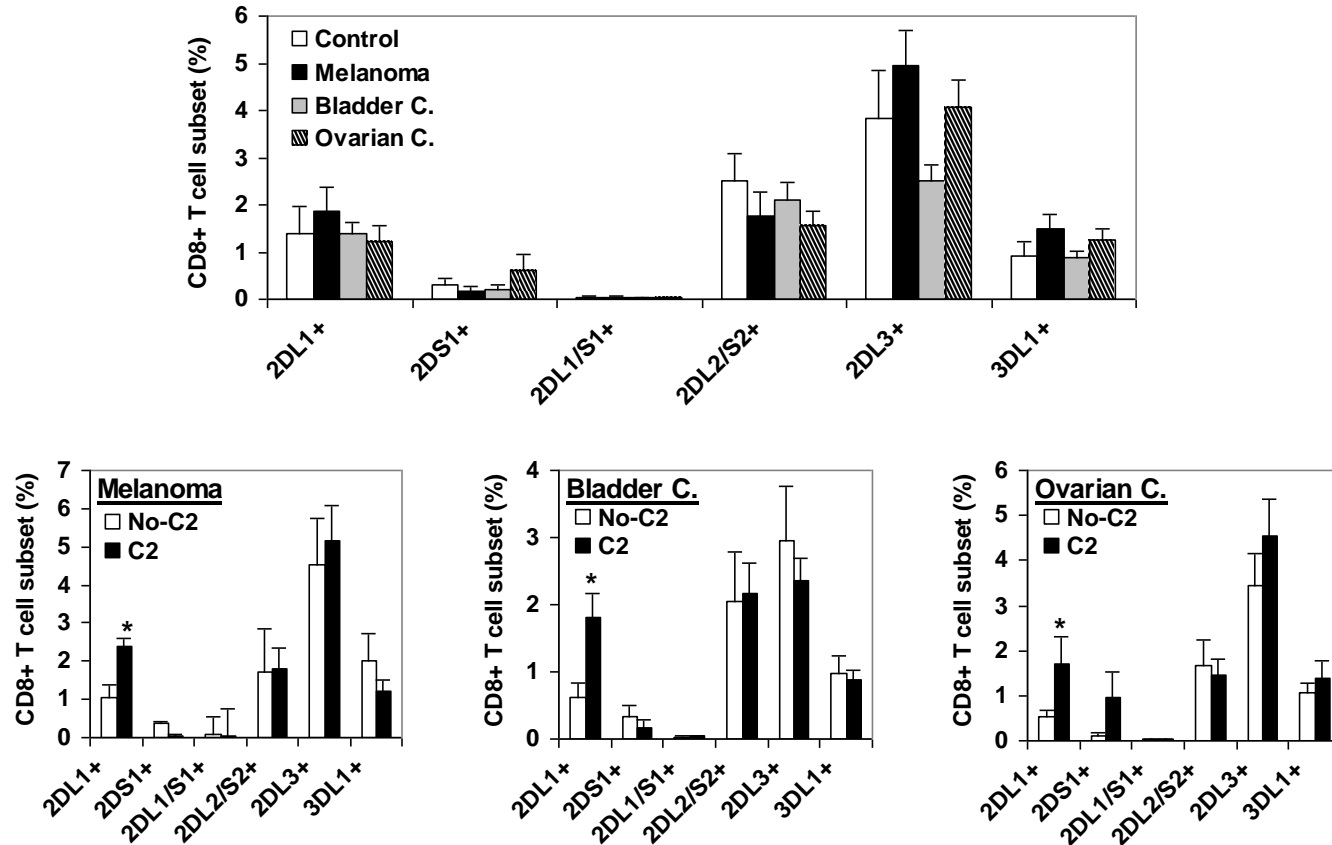
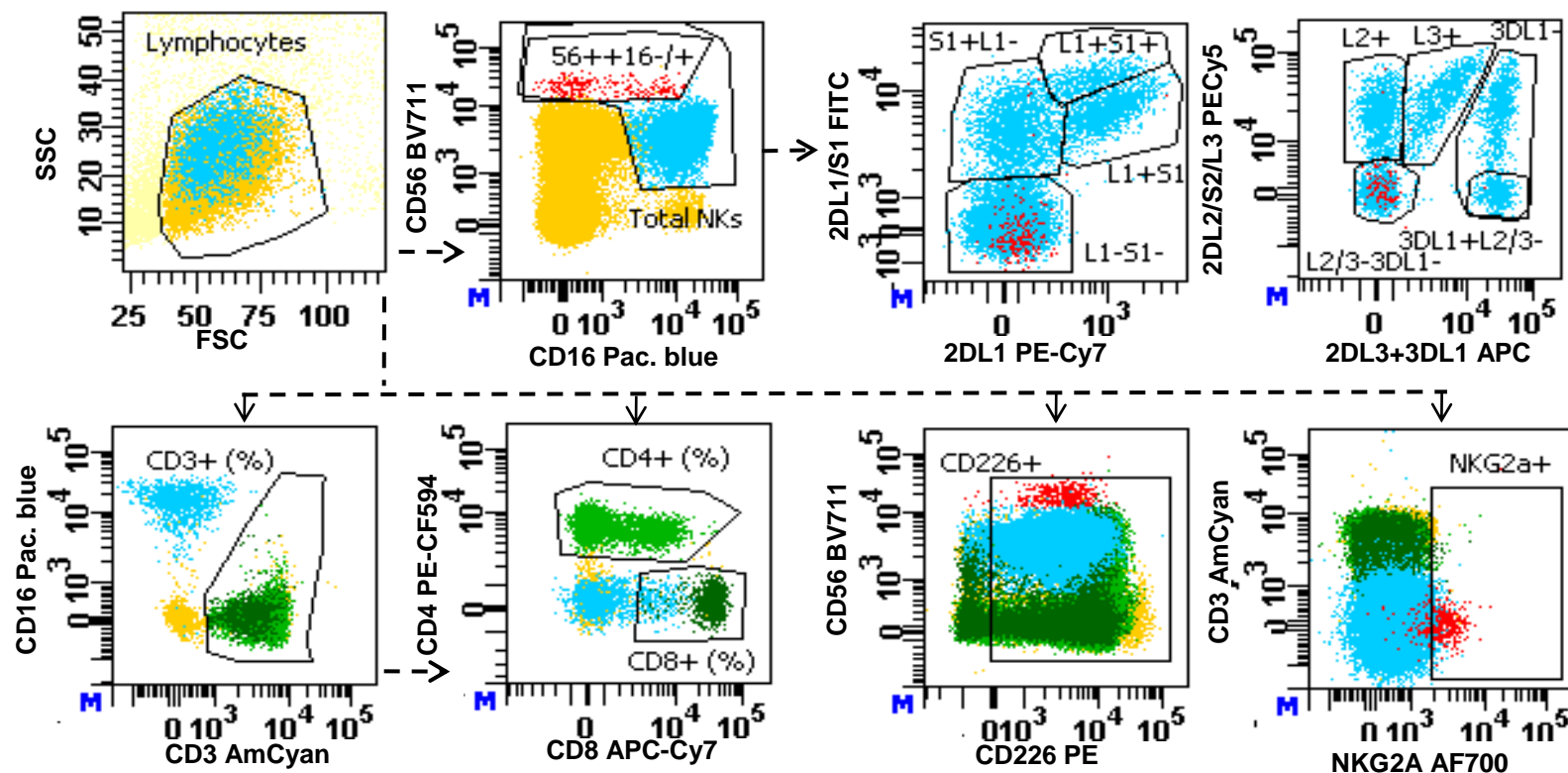


# Supplementary Figure S1



**Supplementary Figure S1. Frequency of KIR+ CD8+ T cell subsets for Melanoma, Bladder, and Ovarian cancer patients.** A, Frequency of KIR+ CD8+ T cell subsets in peripheral blood of healthy controls (n=42) and cancer patients (80 melanomas, 80 bladder cancers and 89 ovarian cancers) and according to the presence of the HLA-C2 or Bw4 ligands, respectively. \* p<0.05, comparing KIR2DL1+ CD8+ T cells in HLA-C2 positive (C1C2 or C2C2) and negative (C1C1) cancer patients. No differences in KIR+ CD8+ T cell subsets were observed for HLA C1-ligands or Bw4-ligands in any of the three types of cancer (data not shown).







## Supplementary Figure S2



**Supplementary Figure S2. Analysis of activating and inhibitory KIR receptors on peripheral blood CD8<sup>+</sup> T cells and NK cells.** Gating strategy to identify total lymphocytes, CD3<sup>+</sup>, CD4<sup>+</sup> and CD8<sup>+</sup> T lymphocytes, CD56<sup>++</sup>CD16<sup>-/+</sup>CD3<sup>-</sup> (CD56<sup>bright</sup>) and CD56<sup>+</sup>CD16<sup>+</sup>CD3<sup>-</sup> (CD56<sup>dim</sup>) NK cells, CD226<sup>+</sup> cells, NKG2A<sup>+</sup> cells, as well as KIR2DS1<sup>+</sup>, KIR2DL1<sup>+</sup>, KIR2DL1<sup>+</sup>S1<sup>+</sup>, KIR2DL2/S2<sup>+</sup>, KIR2DL3<sup>+</sup>, and KIR3DL1<sup>+</sup> CD8<sup>+</sup> T cells and NK cells. Diva software combines hierarchical and logical gating strategies to define different cell subsets (see tables below the figure). Lymphocytes were selected based on low dispersion for FSC/SSC and all lymphocyte subsets were hierarchically dependent on this gate. CD3<sup>+</sup> lymphocytes were gated as CD3<sup>+</sup>CD16<sup>-/+</sup> cells. CD4<sup>+</sup> and CD8<sup>+</sup> T cell subsets were set within CD3<sup>+</sup> lymphocyte. Total NKs gate included CD56<sup>++</sup>CD16<sup>-/+</sup> events combined with a NOT(CD3<sup>+</sup>) gate. To exclude M2-monocytes in the NK cell subset, CD16<sup>++</sup>CD56<sup>-</sup> events were gated-out. The expression of KIR receptors was estimated on CD3<sup>+</sup>CD8<sup>+</sup> T Lymphocytes ("CD8<sup>+</sup>") and on total NK cell ("NKs totales (%)") subsets. A special analysis described elsewhere [1] was applied to patients with KIR2DL3 alleles that were nonreactive with anti-KIR2DL3 antibody clone 180701 (KIR2DL3\*005 or \*015 alleles).

[1] Falco M, Romeo E, Marcenaro S, Martini S, Vitale M, Bottino C, et al. Combined genotypic and phenotypic killer cell Ig-like receptor analyses reveal KIR2DL3 alleles displaying unexpected monoclonal antibody reactivity: identification of the amino acid residues critical for staining. *J Immunol* 2010;185:433-41.

Tube: Tube\_001 **Gating strategy**

Population	#Events	%Parent	%Total
 All Events	505,859	###	100.0
 Lymphocytes	388,043	76.7	76.7
<input checked="" type="checkbox"/> CD3+	287,332	74.0	56.8
 CD4+	231,088	80.4	45.7
 CD8+	44,517	15.5	8.8
<input checked="" type="checkbox"/> CD8+L1+	218	0.5	0.0
<input checked="" type="checkbox"/> CD8+S1+	297	0.7	0.1
<input checked="" type="checkbox"/> CD8+L1+S1+	20	0.0	0.0
<input checked="" type="checkbox"/> CD8+3DL1+	241	0.5	0.0
<input checked="" type="checkbox"/> CD8+L2+	1,588	3.6	0.3
<input checked="" type="checkbox"/> CD8+L3+	3,298	7.4	0.7
<input checked="" type="checkbox"/> Ts CD226+	272,443	94.8	53.9
<input checked="" type="checkbox"/> CD4+CD226+ (%)	224,595	78.2	44.4
<input checked="" type="checkbox"/> CD8+CD226+ (%)	37,480	13.0	7.4
<input checked="" type="checkbox"/> Total NKs	60,766	15.7	12.0
<input checked="" type="checkbox"/> NKG2a+	1,782	2.9	0.4
<input checked="" type="checkbox"/> CD226+	59,680	98.2	11.8
<input checked="" type="checkbox"/> NOT(CD3+ (%))	100,711	26.0	19.9
 NKs totales (%)	59,746	15.4	11.8
 56++16-/+	1,020	1.7	0.2
<input checked="" type="checkbox"/> L2/3-3DL1-	23,727	39.7	4.7
<input checked="" type="checkbox"/> L1-S1-	39,127	65.5	7.7
<input checked="" type="checkbox"/> KIRneg	18,356	30.7	3.6
<input checked="" type="checkbox"/> NKs CD226+	58,061	97.2	11.5
<input checked="" type="checkbox"/> NKG2a++	5,732	9.6	1.1
<input checked="" type="checkbox"/> NKG2a-	54,014	90.4	10.7
<input checked="" type="checkbox"/> KIRnegNKG2a-	16,340	27.3	3.2
<input checked="" type="checkbox"/> L1+S1-	7,064	11.8	1.4
<input checked="" type="checkbox"/> S1+L1-	10,581	17.7	2.1
<input checked="" type="checkbox"/> L1+S1+	2,907	4.9	0.6
<input checked="" type="checkbox"/> 3DL1+	13,642	22.8	2.7
<input checked="" type="checkbox"/> L2/S2+	12,852	21.5	2.5
<input checked="" type="checkbox"/> L3+	9,095	15.2	1.8
<input checked="" type="checkbox"/> 3DL1+L2/3-	5,314	8.9	1.1
<input checked="" type="checkbox"/> L1+total	9,924	16.6	2.0
<input checked="" type="checkbox"/> 3DL1+L2/3-L1-S1-NKG2a-	3,425	5.7	0.7
<input checked="" type="checkbox"/> L1+S1-L2/3-3DL1-NKG2a-	2,075	3.5	0.4
<input checked="" type="checkbox"/> L2+L1-S1-3DL1-NKG2a-	7,236	12.1	1.4
<input checked="" type="checkbox"/> L3+L1-S1-3DL1-NKG2a-	4,285	7.2	0.8
<input checked="" type="checkbox"/> S1+L1-L2/3-3DL1-NKG2a-	2,401	4.0	0.5
<input checked="" type="checkbox"/> 16-56-	204,020	52.6	40.3
<input checked="" type="checkbox"/> CD226-	26,416	6.8	5.2
<input checked="" type="checkbox"/> Monocyte vs NKs CD56-CD16+	1,631	0.4	0.3