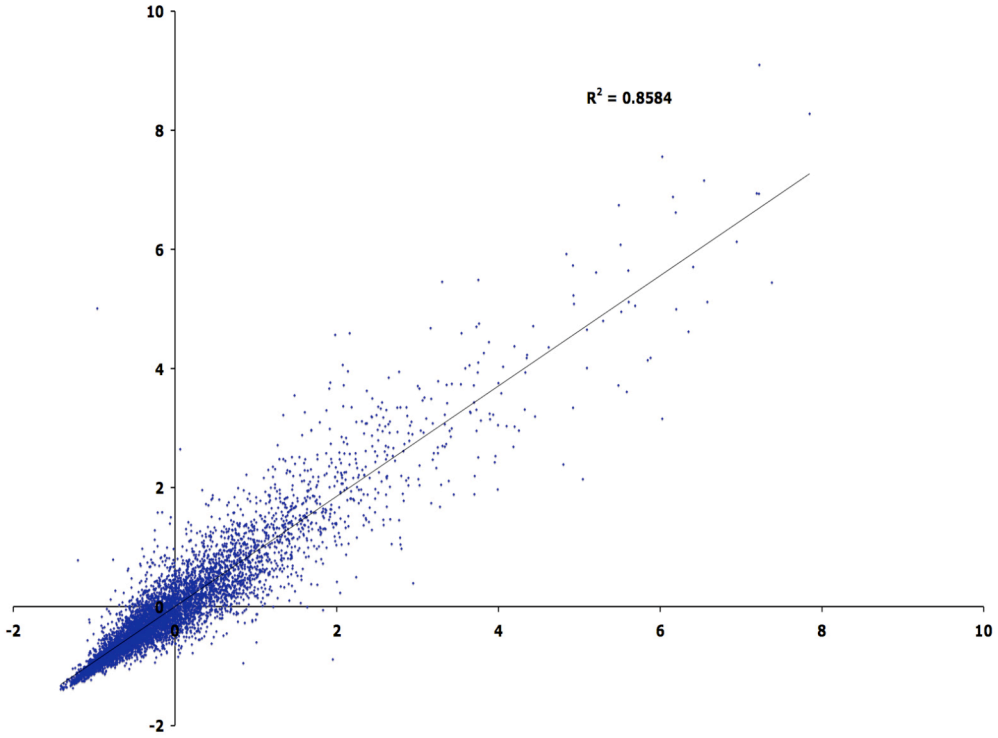
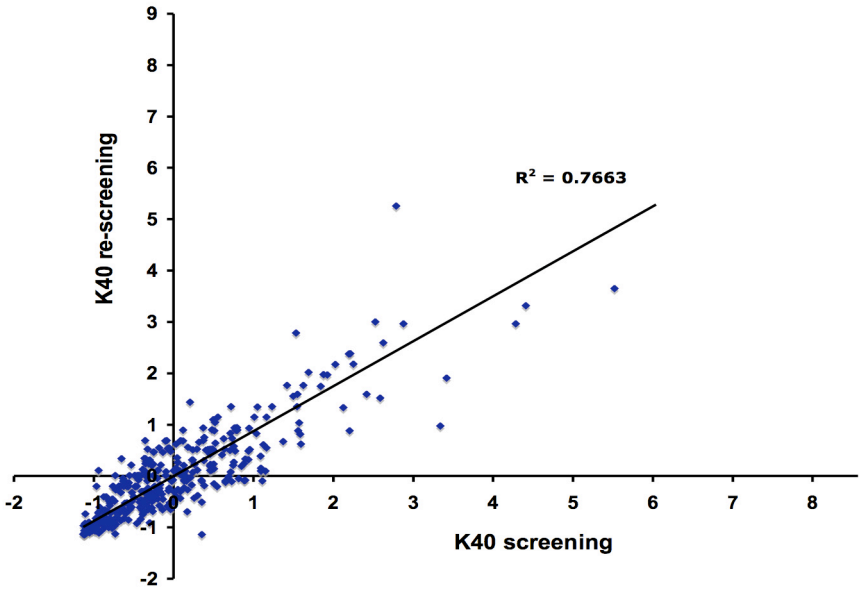
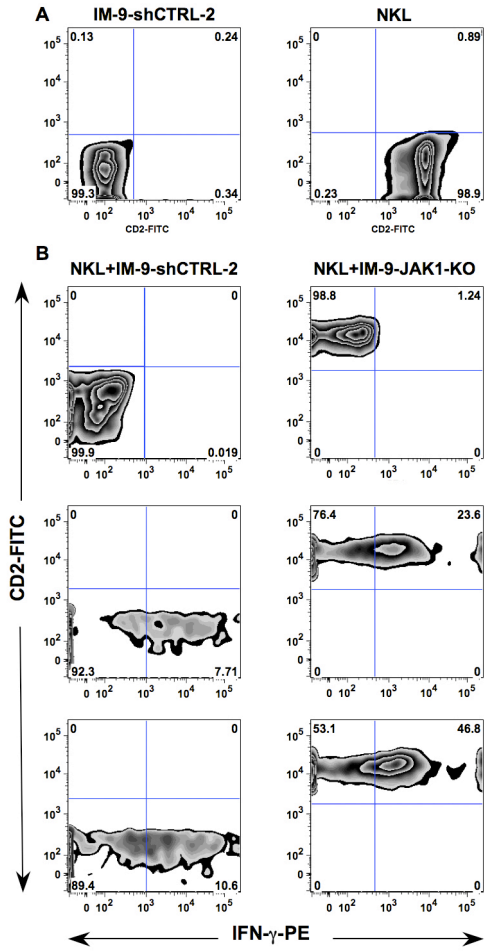
Supplementary Figure 1: **Correlation of INF-γ values in 2 replicate sets.** INF-γ values were compared from 2 replicate sets (12,288 wells) after incubation with NKL effector cells.



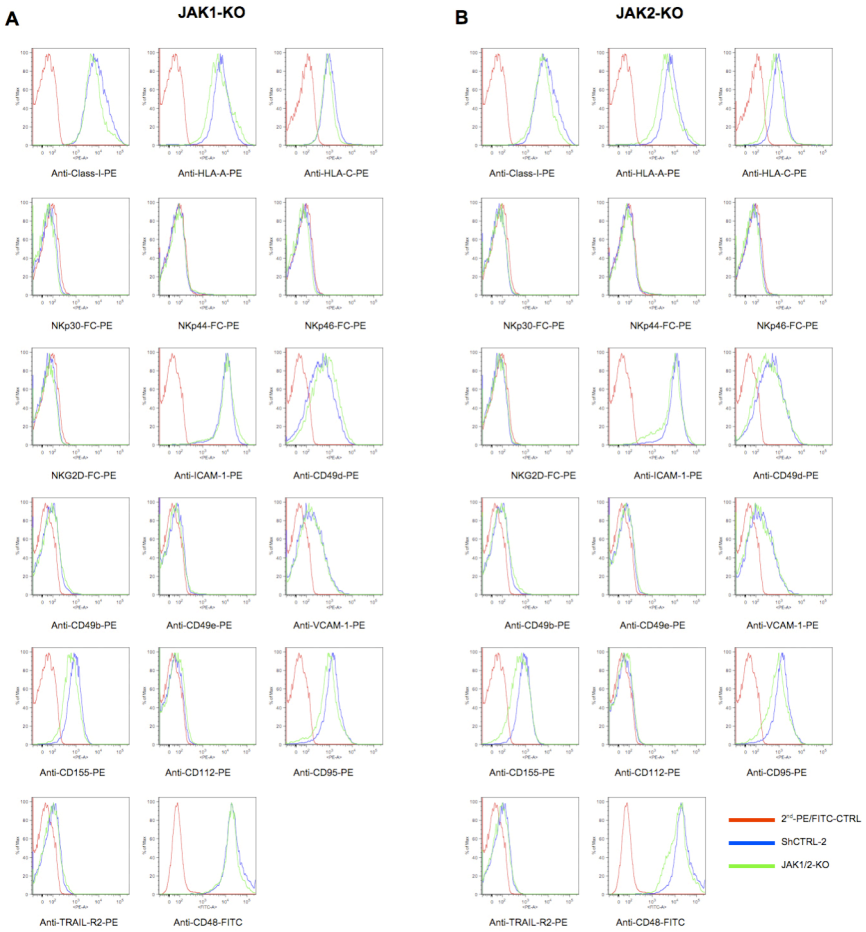
Supplementary Figure 2: **Reproducibility of the cell-cell interaction screen.** A random plate of the TRC library (K40) was used to repeat the entire screen with different batches of IM-9 target cells and NKL effector cells. INF-γ values obtained in the original plate (K40 screen) and the repeated screen (K40 re-screening) were correlated to validate the reproducibility of the genetic screen.



Supplementary Figure 3: **IFN-γ intracellular staining of IM-9-JAK1-KO co-cultured with NKL. (A)** Representative example of IM-9-shCTRL-2 transduced or IM-9-JAK1-KO co-incubated for 4 hours with NKL at 1:1 E/T ratio in the presence of brefeldin A. Cells were subsequently harvested and stained on the surface with CD2-FITC, fixed, permeabilized and stained with IFN-g-PE. NKL are 100% CD2 positive while IM-9 are negative. **(B)** The analysis of IFN-γ secretion was performed on both populations gated based on their CD2 expression.



Supplementary Figure 4: **Flow cytometry validation of inhibitory/activating ligands expression in JAK1 and JAK2-KO.** The expression of several inhibitory/activating ligands and several adhesion molecules on IM-9-JAK1 and JAK2-KO cells was also evaluated using flow cytometry.



Supplementary Table 1: *Target sequences of the validated genes.*

Gene ID	Symbol	shRNAs	Target sequence
3480	IGF1R	IGF1R-1	AAACACATTTGGGATGTTCCT
		IGF1R-3	GCCGAAGATTTCACAGTCAAA
		IGF1R-4	CCAAATTATGTGTTTCCGAAA
3643	INSR	INSR-1	CACCTATTTCTACGTGACAGA
		INSR-3	CCTACCCTTCAAGAGATGATT
		INSR-4	CCTGTCTAATGAACAGGTGTT
5594	MAPK1	MAPK1-1	TATTACGACCCGAGTGACGAG
		MAPK1-2	TGGAATTGGATGACTTGCCTA
		MAPK1-5	TATCCATTCAGCTAACGTTCT
3716	JAK1	Jak1-1	GAGACTTCCATGTTACTGATT
		Jak1-2	CATGCCGTATCTCTCCTCTTT
		Jak1-3	CTGAGCTACTTGGAGGATAAA
3717	JAK2	Jak2-1	CAGTGTTAGATATGATGAGAA
		Jak2-3	CACAGTTTGAAGAGAGACATT
		Jak2-4	GCTTTGTCTTTCGTGTCATTA
3718	JAK3	Jak3-1	TCCTGCTAAGAAACTCCAATT
		Jak3-2	CTCTTCACCTACTGCGACAAA
		Jak3-3	CAGCGCCTATCTTTCTCCTTT
		Jak3-4	CAAAGAAGCAAGGAACCAAAT
7297	TYK2	TYK2-1	GAGATCCACCACTTTAAGAAT
		TYK2-2	TGGTATCACTCCTCCTTGCTT
		TYK2-3	GAGGCCATCATTCCGCACCAT
		TYK2-4	CGAGCACATCATCAAGTACAA

Supplementary Table 2: *Control sequences.*

Symbol	Control target	Target sequence
shCTRL-1	Estrogen receptor	CCTGAAGTCTCTGATTAAGTA
shCTRL-2	GFP	GCAAGCTGACCCTGAAGTTCA
shCTRL-3	LacZ	CGCGATCGTAATCACCCGAGT
shCTRL-4	Luciferase	CTTCGAAATGTCCGTTCGGTT

Supplementary Table 3: *Gene expression of activating/inhibitory ligands in JAK1-KO and CTRLs. (raw data).*

	CTRLs				JAK1-KO			
Gene	IM9 # 1	shCTRL-2 #1	IM9 #2	shCTRL-2 #2	JAK1-1 #1	JAK1-3 #1	JAK1-1 #2	JAK1-3 #2
HLA-A	10717	11825	10197	10815	10393	11229	10342	10318
HLA-A0226	12328	13283	11787	12335	11731	12383	12168	12451
HLA-A29.1	12170	13293	11654	12149	11611	12161	11933	12213
HLA-b	9550	10653	9234	9786	9421	10088	9625	8998
HLA-B0707	9705	11445	9511	10000	10439	10558	10475	9976
HLA-C	7810	8709	7672	7918	7825	8311	7938	7207
HLA-E	3181	3165	3211	2909	2904	2870	2921	2594
HLA-F	912	1127	894	1026	994	1031	1006	885
HLA-G	6949	7508	6641	7333	6959	6803	7169	6573
HLA-G2.2	154	193	151	186	191	188	181	190
MICA	79	92	103	93	87	98	126	107
MICB	472	664	726	766	698	835	800	904
ULBP1	62	59	59	42	45	48	53	57
ULBP2	40	55	51	40	46	51	50	48
ULBP3	52	34	54	41	44	43	42	37
CD48	7486	5495	8951	6172	6351	4912	6355	5115
PVR	237	247	251	272	265	229	244	222
PVRL1	230	193	240	179	146	225	156	166
PVRL2	120	126	89	111	114	126	120	90
TNFRSF6B	150	135	148	133	144	141	130	143
TNFRSF10A	192	192	195	214	317	341	295	307
TNFRSF10B	1150	1044	1041	766	1057	1130	873	1003
TNFRSF10C	55	51	45	52	50	48	48	69
TNFRSF10D	334	365	337	324	449	633	369	503
FAS	1924	1625	1687	1528	1537	1716	1363	1892
ICAM1	2190	2167	1670	1653	2028	1920	1631	1758
VCAM1	319	415	202	378	487	933	393	1030
ITGA4	387	737	360	497	964	859	761	566
ITGA5	84	82	89	68	85	97	89	81
ITGA6	33	34	31	29	27	30	34	32
ITGA2	29	34	38	38	32	40	34	43
ITGAL	275	352	264	324	168	415	181	305