SUPPLEMENTARY FIGURE LEGENDS

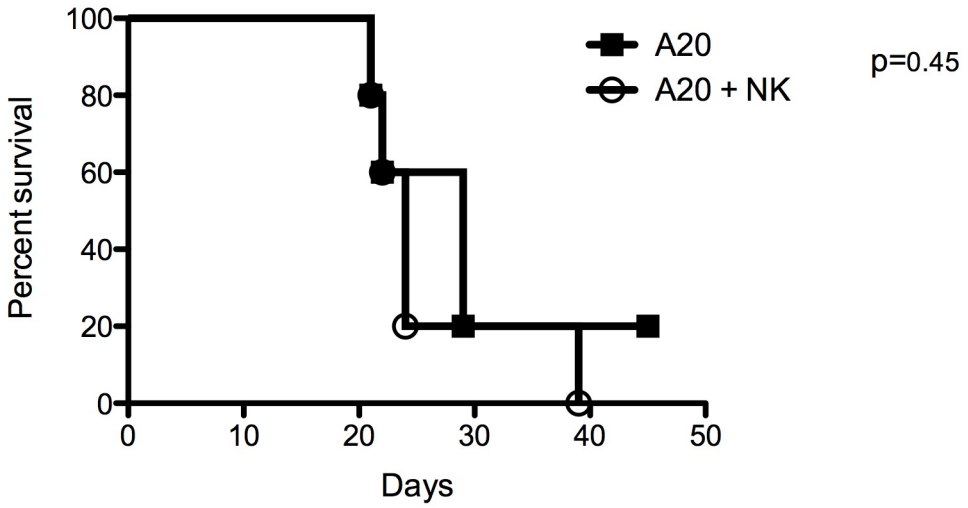
Supplementary Figure 1. NK cells fail to eradicate tumor when co-injected at high E:T ratios. $1x10^6$ C57BL/6 or FVB NK cells were co-injected at the same time as $1x10^4$ A20 cells into Balb/c mice and followed for survival' p-0.45. Representative of two independent experiments.

Supplementary Figure 2.Tumor-infiltrating NK cells downregulate activating receptors and demonstrate impaired function (**A**) Phenotype and sorting strategy of naïve NK cells derived from $CD45.1^+$ donor mice; top panel shows phenotype post-magnetic enrichment and the bottom panel shows the post-FACS sorting phenotype and purity. (**B**) Expression of activating and inhibitory Ly49 receptors and the inhibitory NKG2A/C/E receptors among transferred NK cells is similar to naïve or cultured NK cells. (**C**) FACS sorted $CD3^-$ $DX5^+$ or $CD3^-NK1.1^+$ NK cells from $CD45.1^+$ congenic donors were transferred into $CD45.2^+$ recipients along with a T cell-depleted bone marrow transplant from $CD45.2^+$ donors. All analyses are gated as indicated above each plot. Within the spleen of tumor-bearing mice that received NK cells, $NKp46^+$, $NKG2D^+$ and most $DX5^+$ cells are absent in the transferred $CD45.1^+$ population when compared with the bone-marrow derived $CD45.2^+$ population. (**D**). Expression pattern of KLRG1, CD27 and CD11b shows a pattern of maturation of NK cells after in vivo transfer. Results are representative of 2-3 independent experiments.

Supplementary Figure 3. Prolonged exposure to tumor is sufficient to induce NK cell dysfunction and proliferation; loss of function correlates with proliferation (**A**) Inhibition of NK cell function by tumor exposure is contact-dependent. Balb/c NK were cultured for 5 days alone, in contact with irradiated A20 cells, or separated from irradiated A20 cells by a transwell, then harvested for use in a standard chromium release assay. (**B**) Signaling through NKG2D is not required for the observed cytotoxic dysfunction. Sorted wild-type (left) or NKG2D$^{-/-}$ (middle) NK cells were cultured alone or with irradiated A20 cells for five days then harvested for use in a standard chromium release against A20 targets at the indicated effector:target ratios (at right, statistical analysis at an ET ratio of 5:1); all indicated significance symbols indicate $p < 0.001$ (**C**) IFNγ and NKG2D expression on proliferated (CFSE low) or unproliferated (CFSE high) NK cells after a 5 day co-culture with tumor. Analyses were in triplicate (**A,B**) and represent 2 independent experiments.

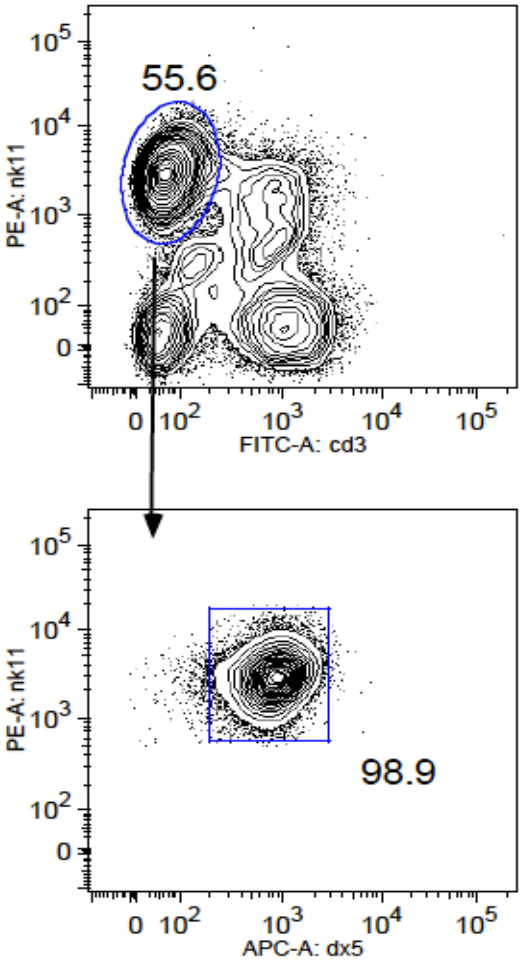
Supplementary Figure 4. Expression of Eomes, Tbet and IFNγ in naïve or IL2 stimulated cultured NK cells. NK cells were analysed for expression of transcription factors Eomes and T-bet as well as production of IFNγ upon stimulation with PMA and ionomycin. NK cells were harvested from naïve spleens or from a 5 day culture with IL-2 1500 U/mL. Analyses represent 3 independent experiments.

Suppl Fig 1


100
80
60
40
20
0
Percent survival
0
10
20
30
40
50
Days
A20
A20 + NK
p=0.45

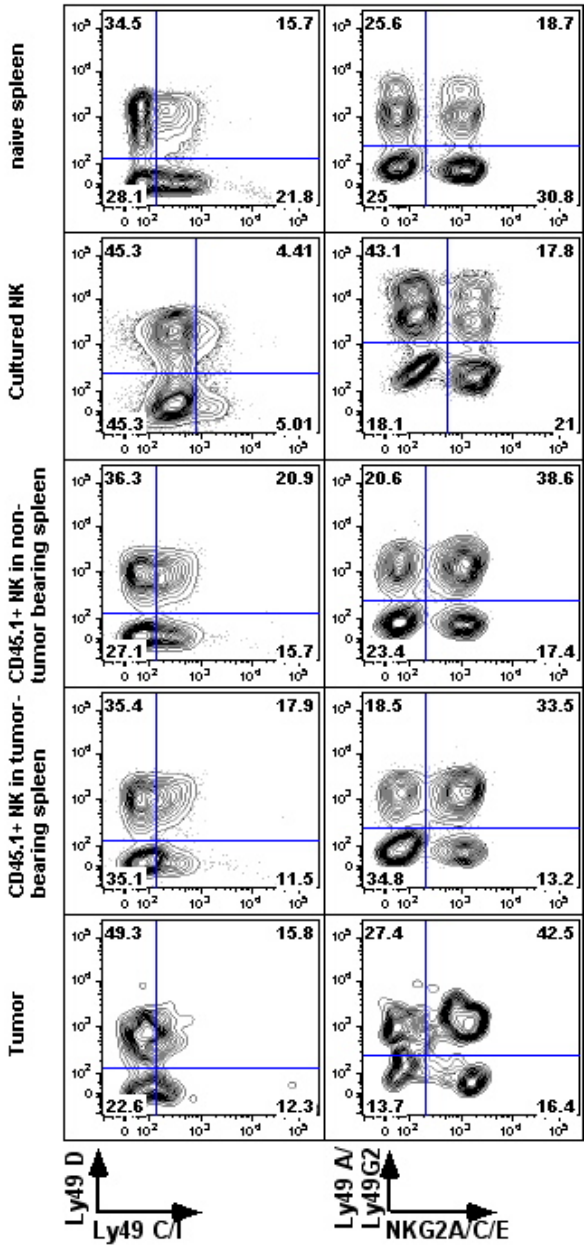
Supp Figure 2

A.



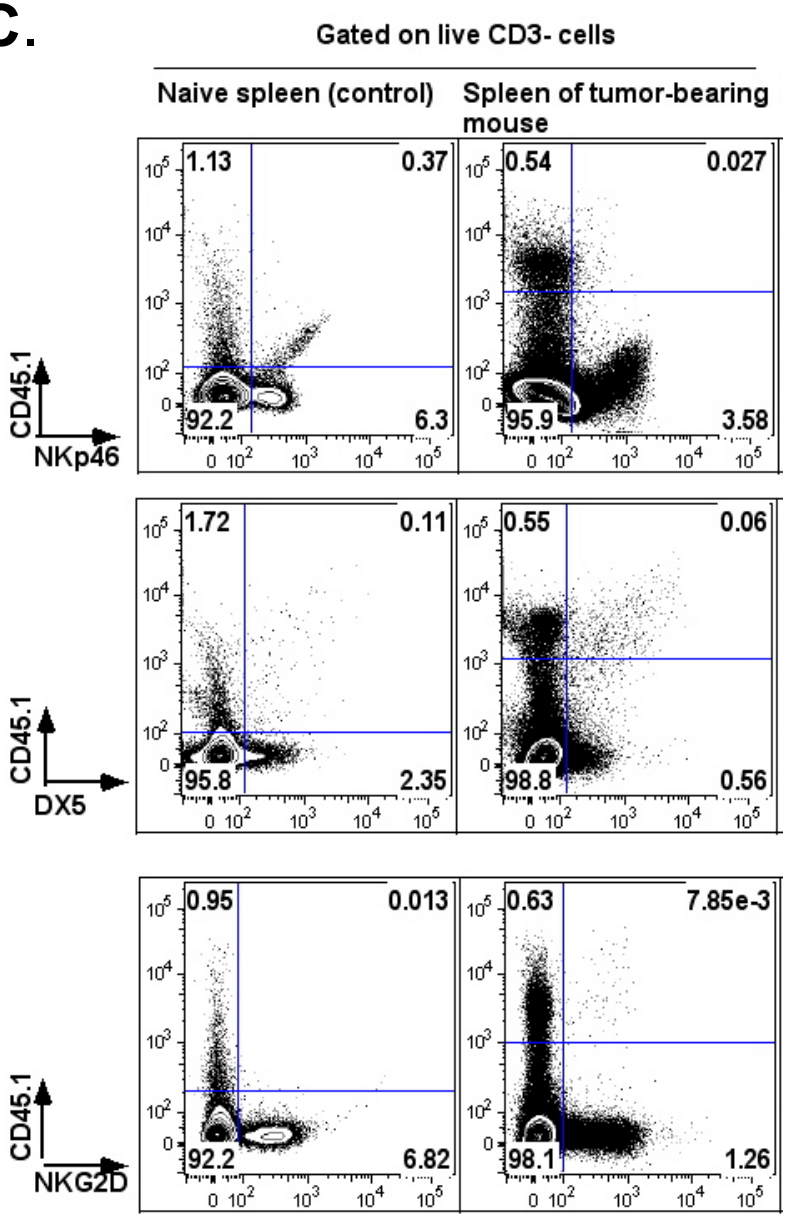
Supp Figure 2

B.

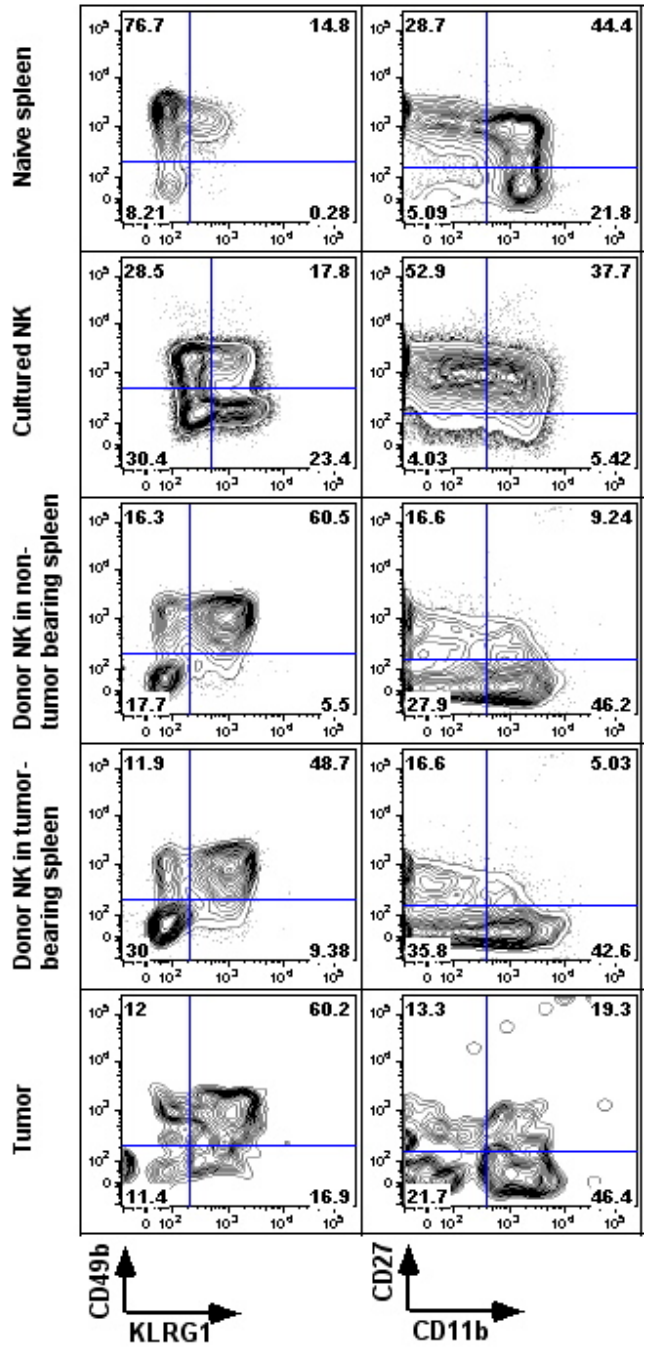


Supp Figure 2

C.

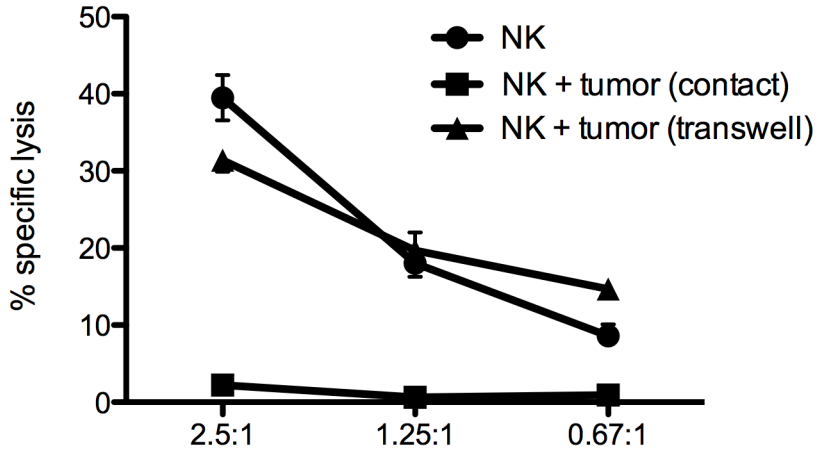


D.

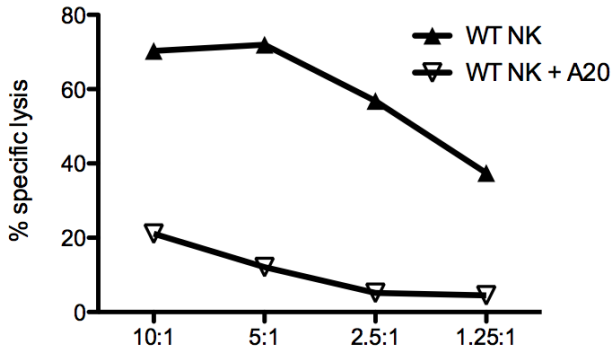

Naive spleen
76.7
14.8
8.21
0.28
28.7
44.4
5.09
21.8
Cultured NK
28.5
17.8
30.4
23.4
52.9
37.7
4.03
5.42
Donor NK in non-tumor bearing spleen
16.3
60.5
17.7
5.5
16.6
9.24
27.9
46.2
Donor NK in tumor-bearing spleen
11.9
48.7
30
9.38
16.6
5.03
35.8
42.6
Tumor
12
60.2
11.4
16.9
13.3
19.3
21.7
46.4
CD49b
KLRG1
CD27
CD11b

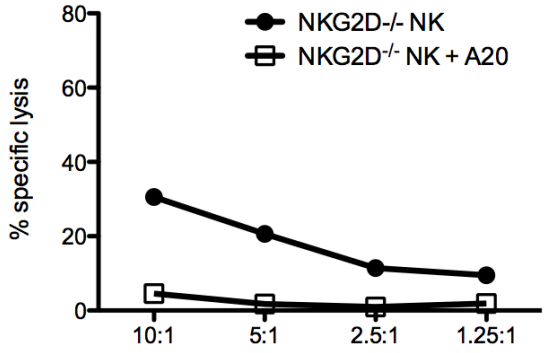
Supp Figure 3

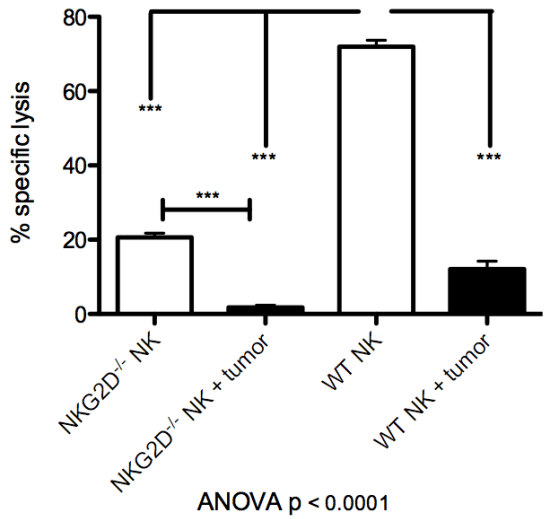
A.



B.

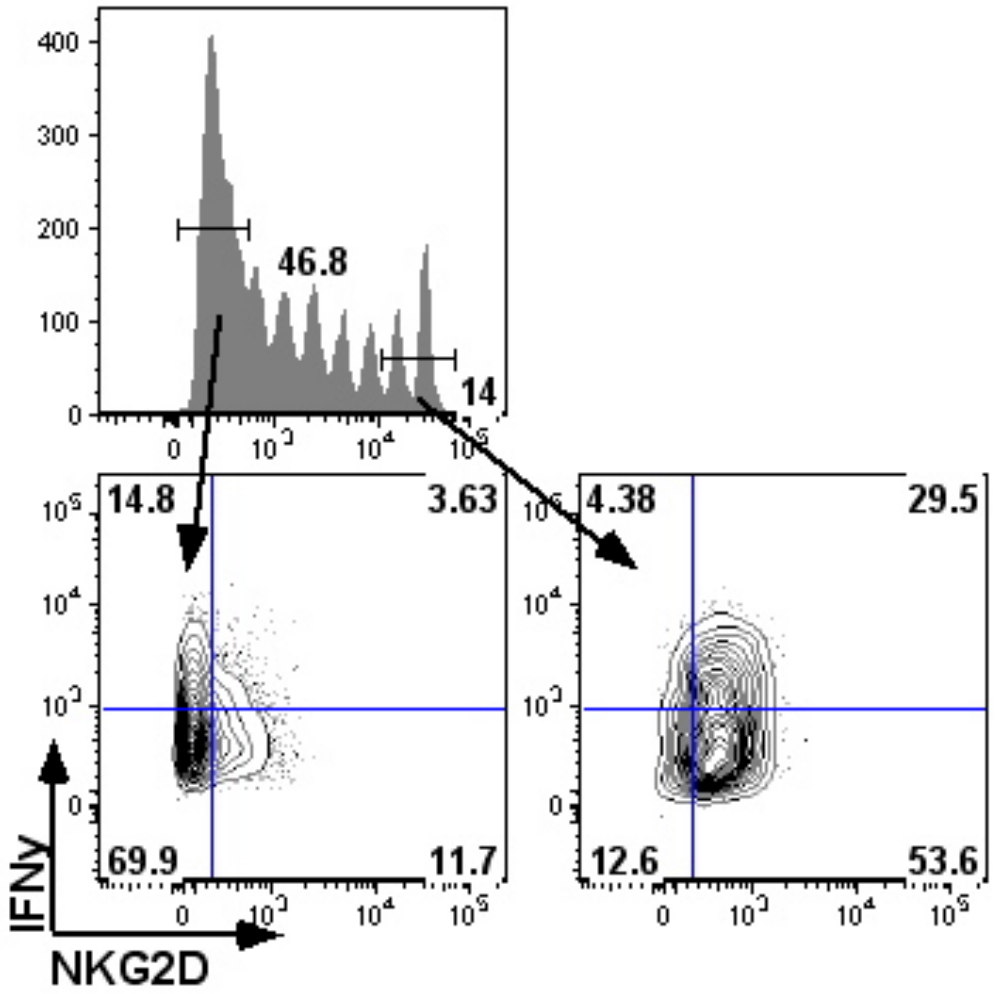






Supp Figure 3

C.



Supp Figure 4