#### **Supplemental Figure Legends**

**S. Figure 1. Transcriptomic analysis of type 1 ILC in liver metastasis. (A)** Gating strategy for cell sorting of ILC1, CD49a<sup>+</sup> and CD49a<sup>-</sup>NK cells from metastasis-free liver (MFL) and liver metastasis (LM) in the MC38 metastasis mouse model. Cells first gated based on forward and side scatter area parameters (FSC-A and SSC-A) were then selected for doublet exclusion (Heights, H versus Area, A). NKp46<sup>+</sup> cells were finally collected based on CD49a and CD49b expression levels. **(B)** Volcano plots of differentially expressed genes between LM and MFL NK cells and CD49a<sup>+</sup>NK cells and MFL or LM CD49a<sup>-</sup>NK cells. FDR < 0.1 was considered significant. **(C)** Gene Ontology (GO) of upregulated (top) and downregulated (below) genes.

**S. Figure 2. MC38-induced liver metastasis microenvironment comprises a variety of cellstate transitions in NK clusters. (A)** Top: Umap of cell clusters from analysis of scRNA-seq from (8). Bulk RNAseq was used to produce a unique gene signature for CD49a<sup>+</sup>NK cells (*1700025G04Rik, Prss16, Tpx2, H2bc23, Cdk6, Plac8, kKif20a, Cenpf, Atad5, Depdc1b, Smtn, Nt5e, 2410022M11Rik, H34, Emb, Tmprss6, 5830408C22Rik, H2aCc10, Spag5, Nsl1, Cxcl10, Sfxn5*) generated by extracting the genes uniquely expressed by CD49a<sup>+</sup>NK cells from the liver metastatic samples; Violin plots show scoring of clusters for in house signatures. **(B)** Pseudotime analysis on clusters. Schematic of cluster trajectory in pseudotime analysis. **(C)** Heatmap showing top 60 DEGs on cell clusters along with trajectory analysis. **(D)** Most upregulated genes at transition and branching points of pseudotime analysis.

S. Figure 3. Distinctive type 1 ILC markers in healthy liver and MC38-induced metastasis-free liver and liver metastasis. (A) Histogram plots show gMFI  $\pm$  SEM of CXCR3, CD11b, CD69, CD62L, TIGIT, PD-L1, CD73 and CX<sub>3</sub>CR1 expression and mean frequency  $\pm$  SEM of CD39<sup>+</sup> and CXCR6<sup>+</sup> cells among ILC1 (blue), CD49a<sup>+</sup>NK (orange) and CD49a<sup>-</sup>NK (red) cells in MFL and LM (CXCR3 n=7, \*\*p=0.002 one-way ANOVA in LM,

CD62L n=9 \*\*p=0.003, CD69 n=3 \*\*p=0.0013, CD73 n≥3 \*p=0.06, CXCR6 n≥8 \*\*\*p=0.0002, one-way ANOVA). (B) Representative FACS histogram plots show CD62L, CXCR3, CD73, CD39, TIGIT, CXCR6, KLRG1, CX<sub>3</sub>CR1 and CD69 expression on ILC1, CD49a<sup>+</sup>NK and CD49a<sup>-</sup>NK cells in healthy liver (HL), MFL and LM. At least three independent experiments were performed. (C) Representative histograms of Ki-67 expression and relative mean frequency  $\pm$  SEM in ILC1, CD49a<sup>+</sup>NK and CD49a<sup>-</sup>NK from MFL and LM (n=4, \*p=0.01, \*\*p=0.009, one-way ANOVA). (D) Histogram shows mean frequency  $\pm$  SEM of BrdU<sup>+</sup> cells among ILC1, CD49a<sup>+</sup>NK and CD49a<sup>-</sup>NK in LM upon *in vivo* staining with BrdU (n=3).

# S. Figure 4. CXCR3 identifies a liver metastasis-associated NK cluster in human patients.

(A) Umap of NK cell clusters in MFL, LM and peripheral blood mononuclear cells (PBMCs) from a dataset of human liver metastasis colorectal cancer patients (32). (B) Heatmap of top 10 DEGs in NK cell clusters.

S. Figure 5. Expression of effector molecules and killing capacity of type 1 ILC in liver metastasis. (A) Representative pictures of metastatic livers from MC38- and SL4-injected mice. (B) Quantification by ELISA of IFN- $\gamma$  concentration (pg/10<sup>6</sup> cells) in supernatants from sorted splenic NK (black), HL NK (grey), MFL NK (green), LM CD49a NK and LM CD49a<sup>+</sup>NK cells (\*p=0.04, \*\*\*p=0.0002, one-way ANOVA). (C) Top: representative FACS histogram plots of Perforin, Granzyme B (GzmB), Granzyme A (GzmA) and Granzyme C (GzmC) expression on MFL ILC1 and NK cells and on LM ILC1, CD49a<sup>+</sup>NK and CD49a NK cells. Dotted line represents FMO staining control. Below: histogram plots show mean frequency ± SEM of Perforin<sup>+</sup>, Granzyme B<sup>+</sup>, Granzyme A<sup>+</sup> and Granzyme C<sup>+</sup> cells on MFL and LM ILC1, CD49a<sup>+</sup>NK and CD49a<sup>-</sup>NK cells. Three independent experiments with 4 mice per group were performed. For Perforin expression, n=4 (GzmA n ≥ 8 \*\*p=0.002, GzmC n ≥14

\*\*\*\*p<0.0001, one-way ANOVA). **(D)** Histograms show comparison of frequency of granzyme A, B and C expression on LM ILC1, CD49a<sup>+</sup>NK and CD49a<sup>-</sup>NK from MC38- and SL4-derived liver metastasis (MC38 n=5, SL4 n=3, \*\*p=0.001, \*\*p=0.004, \*\*\*p=0.0002, one way ANOVA).

**S. Figure 6. Myeloid cell distribution and function in metastatic liver. (A)** Gating strategy of myeloid cells in LM. **(B)** Histogram plots show frequency of dendritic cells (Total DC) (CD11c<sup>+</sup>MHCII<sup>+</sup>), cDC1 (CD11c<sup>+</sup>MHCII<sup>+</sup>CD11b<sup>+</sup>Xcr1<sup>+</sup>) and cDC2 (CD11c<sup>+</sup>MHCII<sup>+</sup>CD11b<sup>+</sup>) among live lin<sup>-</sup>CD45<sup>+</sup> cells (\*\*p=0.001, one-way ANOVA) **(C)** TGF-β concentration (pg/ml) in supernatants of LM macrophages, MC38 and SL4 cells. Medium was used as control. **(D)** Representative FACS histograms of Arg1, MMR, PD-L1, LAP-1, MHCII and TREM2 expression on F4/80<sup>high</sup> and F4/80<sup>int</sup> LM macrophages. Light grey histograms show isotypic staining control, dark grey histograms show FMO control in HL, MFL and LM. **(E)** Histograms show gMFI ± SEM of Ly6C, CD11c, CD11b, CX<sub>3</sub>CR1 and frequency of Tim4<sup>+</sup> cells in LM F4/80<sup>int</sup> and F4/80<sup>high</sup> macrophages (n>4, Ly6C \*\*p=0.002, CD11c \*\*p=0.04, CX<sub>3</sub>CR1 \*\*p=0.002, Tim4 \*\*p=0.02, two-tailed Student's *t* test). **(F)** Frequency ± SEM of CXCL9<sup>+</sup> cells among F4/80<sup>int</sup> and F4/80<sup>high</sup> macrophages and dendritic cells from LM (n>3 in three independent experiments, \*\*p=0.07, \*\*p=0.01, one-way ANOVA).

**S Figure 7. Macrophages from MC38-induced metastasis contribute to CD49a<sup>+</sup> NK cell generation.** (A) Left: contour plots show F4/80 and CD11b expression on purified macrophages from LM and MFL. Representative contour plots and scatter plots show gMFI  $\pm$  SEM of Tim4, Ly6C and CD115 on F4/80<sup>high</sup> (green) and F4/80<sup>int</sup> (grey) subsets of macrophages from LM and MFL (Tim4 LM \*\*p=0.001, MFL \*p=0.02, Ly6C LM \*\*p=0.001, MFL \*p=0.01, CD115 MFL \*p=0.05, two-tailed Student's *t* test). (B) Fold change (F.C.) of CD49a<sup>+</sup>NK cell frequency upon 72 hours of co-culture with MC38-derived MAMs in presence

or absence of cell-cell contact (n=3, \*p=0.02, two-tailed Student's *t* test). (C) Frequency  $\pm$  SEM of CD107a<sup>+</sup> expression induced by anti-NK1.1 mAb stimulation on NK cells cultured with IL-15 alone (grey) or in combination with TGF- $\beta$  (orange) or upon co-culture with LM macrophages (red) for 72 hours (n≥3, \*\*p=0.003, \*\*\*p=0.0006, one-way ANOVA). (D) Frequency  $\pm$  SEM of BrdU<sup>+</sup>NK cells upon 72 hours with IL-15 (10ng/ml) alone or in co-culture with MC38- or SL4-derived LM macrophages (\*\*\*\*p<0.0001, one-way ANOVA). From panel (A) to (D) at least three independent experiments are shown. (E) Cell interaction analysis of macrophage and NK cell clusters in human liver metastasis from analysis of (32).

S Figure 8. MC38-derived MAMs, but not mono-macrophages, contribute to CD49a<sup>+</sup>NK cell generation. (A) Representative picture of metastatic livers from Rat IgG- and anti-CSF1Rtreated mice. (B) Hematoxylin-eosin staining of liver sections from Rat IgG- and anti-CSF1Rtreated mice and relative quantification. Histogram plot shows metastasis weight (gr) in Rat IgG- and anti-CSF1R-treated mice. Mean  $\pm$  SEM of two independent experiments. (C) Top: representative contour plots of F4/80<sup>int</sup> and F4/80<sup>high</sup> macrophages in Rat IgG- and anti-CSF1Rtreated LM. Below: histogram plots show frequency  $\pm$  SEM of F4/80<sup>int</sup> and F4/80<sup>high</sup> macrophages in LM (n=9 \*\*\*\*p<0.0001, two-tailed Student's t test). (D) Violin plot shows concentration of CXCL10 in LM from Rat IgG- and anti-CSF1R-treated tumor-bearing mice. Histogram plots show gMFI ± SEM of CXCL9 and LAP-1 in leftover macrophages from Rat IgG- and anti-CSF1R-derived LM (n>2, \*p=0.02, two-tailed Student's t test). (E) Mean frequency ± SEM of monocytes, macrophages and dendritic cells in bone marrow from Rat IgG- or anti-CSF1R-treated tumor-bearing mice (n=4). (F) Representative pictures of metastatic livers in Rat IgG- and MC-21-treated mice. Histogram plots show number of metastasis, % of inhibition  $\pm$  SEM of F4/80<sup>int</sup> and F4/80<sup>high</sup> macrophage frequency in LM of Rat IgG- and MC-21-treated tumor-bearing mice (n>5, \*\*p=0.07, two-tailed Student's *t* test).

**S. Figure 9. Immune cell distribution in the liver from Ncr1**<sup> $\Delta$ Cxcr3</sup> **mice. (A)** Left: schematic representation of mouse breeding to obtain Ncr1<sup> $\Delta$ Cxcr3</sup>; right: representative FACS histogram plots of CXCR3 expression on ILC1, NK cells and CD3<sup>+</sup> cells from liver of Cxcr3<sup>fl/fl</sup> and Ncr1<sup> $\Delta$ Cxcr3</sup> mice. **(B)** Histogram plots show frequency ± SEM of liver type 1 ILCs (live lin-CD45<sup>+</sup>NK1.1<sup>+</sup>NKp46<sup>+</sup>), NK cells and ILC1 and of Kupffer cells, dendritic cells (DCs) and macrophages in Cxcr3<sup>fl/fl</sup> and Ncr1<sup> $\Delta$ Cxcr3</sup> mice at steady state (n=3/group).





















