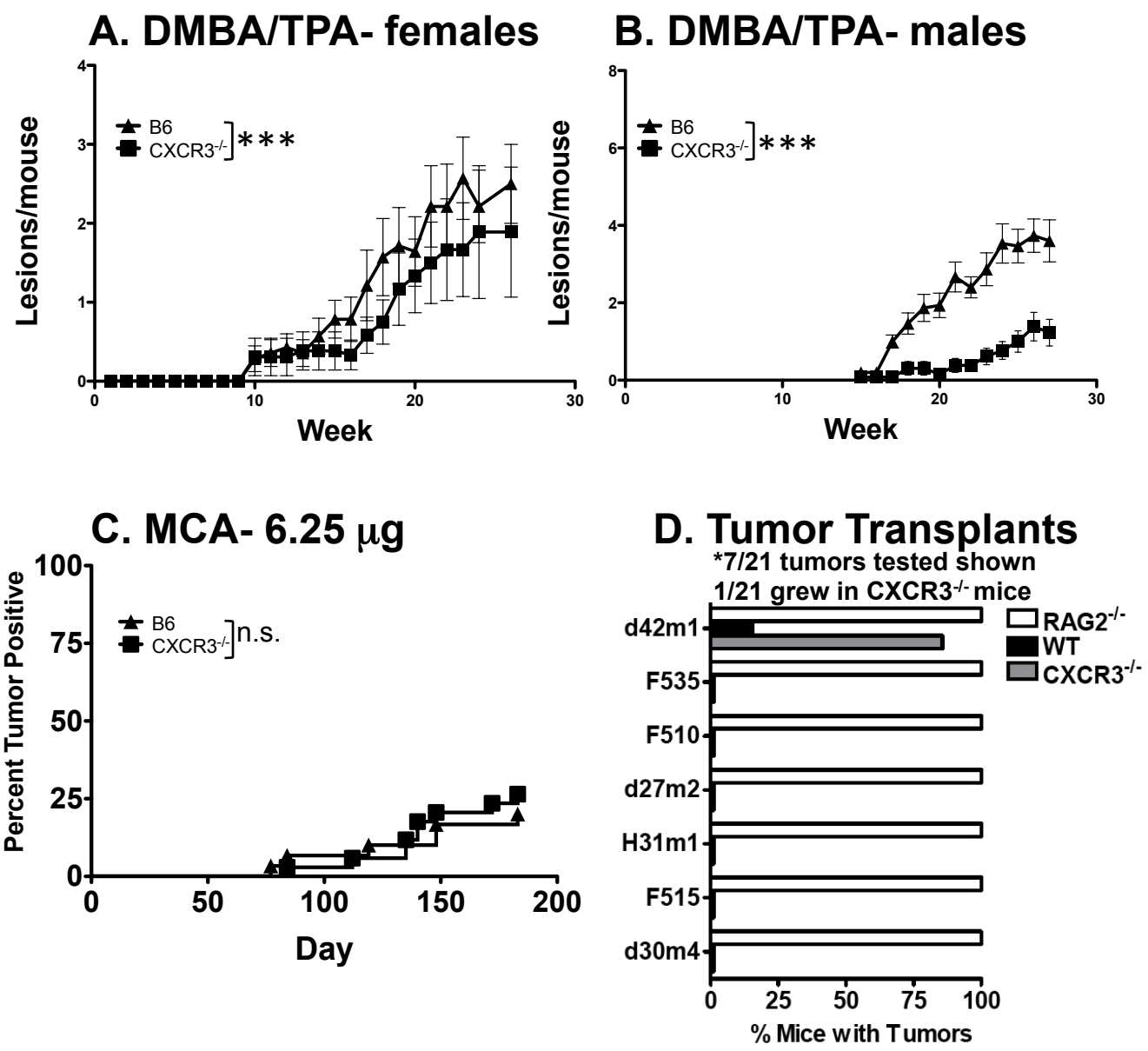
Supplementary Figure Legends

Supplementary Figure 1 CXCR3 promotes DMBA/TPA tumorigenesis but has no role in MCA primary tumorigenesis or in rejection of RAG regressor transplantable fibrosarcomas. (A) Separate experiments from those in Figure 1 show decreased DMBA/TPA tumor development in $CXCR3^{-/-}$ female mice compared to WT females (***$p<0.001$, n=14 for $CXCR3^{-/-}$(filled squares) and n=12 for WT females (filled triangles) and (B) in an independent experiment with $CXCR3^{-/-}$ male and WT male mice (***$p<0.001$, n=15 for $CXCR3^{-/-}$ and n=13 for WT males). Note the difference between WT and $CXCR3^{-/-}$ mice is more robust for males than females. In addition, the experiments in A and B were performed independently and in experiments where male and female cohorts were used concurrently, no significant difference in tumor development between $CXCR3^{-/-}$ male and female mice was noted (data not shown). (C) MCA tumorigenesis at 6.25 μg dose shows no difference between $CXCR3^{-/-}$ and WT mice (n.s.= not significant, n=33 for $CXCR3^{-/-}$ and n=30 for WT). (D) CXCR3 is not required for the rejection of a majority of RAG regressor tumors (note, data shown for 7/21 tumors). Twenty-one immunogenic RAG regressor tumors, which grow progressively in $RAG2^{-/-}$ mice and are rejected in WT mice, were transplanted into the flanks of the indicated mice and monitored for tumor growth (1×10^6 cells/mouse, n=2 for $RAG2^{-/-}$, n=4-5 for WT and $CXCR3^{-/-}$ mice). The d42m1 tumor is the only one that grows in $RAG2^{-/-}$ and $CXCR3^{-/-}$ mice whereas it is rejected in WT mice. Twenty other tumor cell lines tested rejected in both WT and $CXCR3^{-/-}$ mice and grew in $RAG2^{-/-}$ mice (tumor cell lines F535, F510, d27m2, H31m1, F515 and d30m4 are shown).

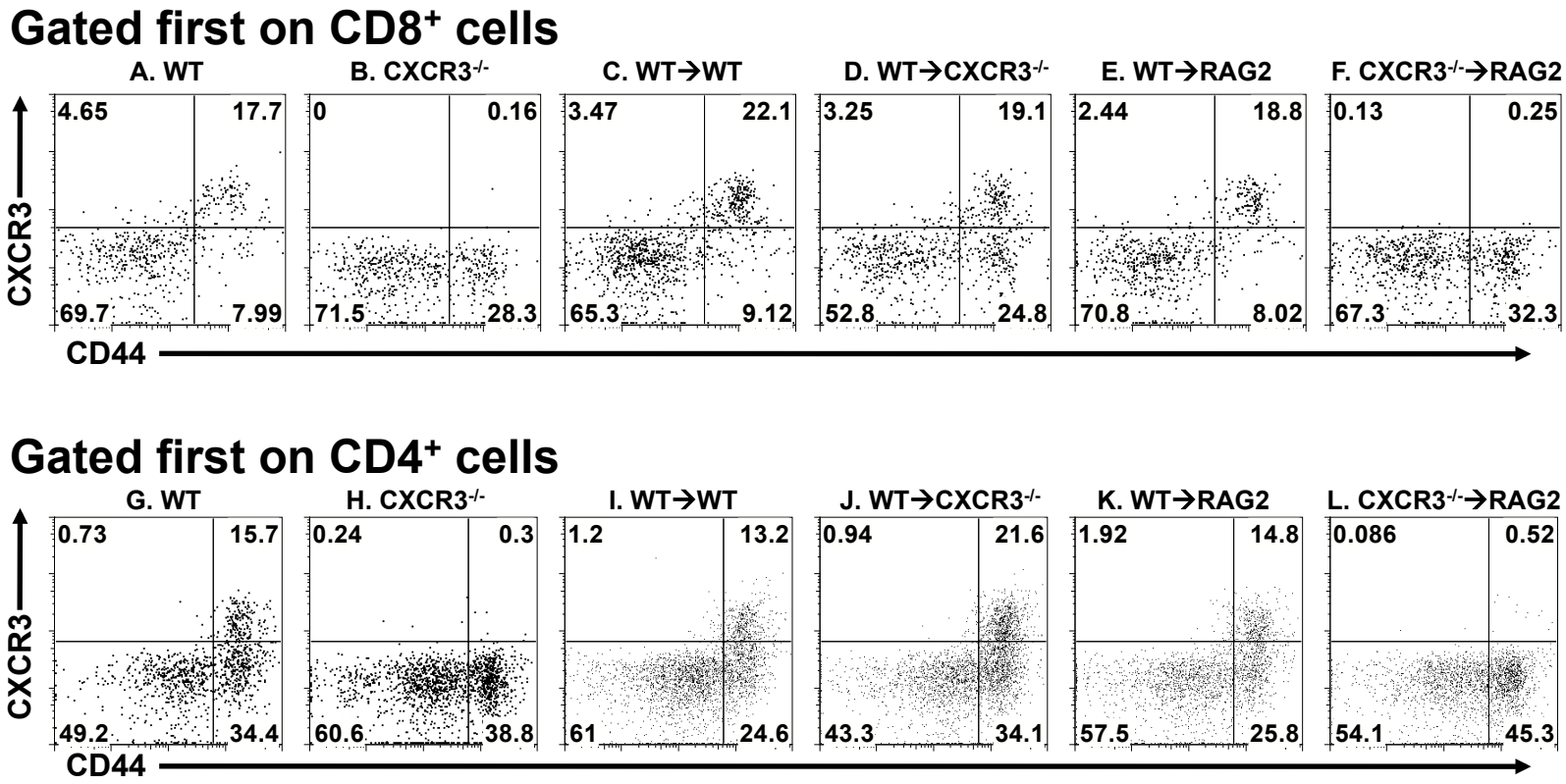
Supplementary Figure 2 Reconstitution of $CXCR3^{-/-}$ mice with WT fetal liver cells restores T and NK cell CXCR3 expression to WT levels. Recipient mice were radiated, adoptively transferred with either WT or $CXCR3^{-/-}$ fetal liver cells and were then rested for 12 weeks. FACS analysis of splenocytes at the end of experiments, with the indicated gating first either on $CD8^+$ or $CD4^+$, revealed CXCR3 expression in the T cell, $CD44^+$ population of WT mice (compare A to B and G to H). All recipients that received WT cells had similar CXCR3 expression (includes C, D and E for $CD8^+$ T cells and I, J and K for $CD4^+$ T cells). As expected, $RAG2^{-/-}$ mice that received $CXCR3^{-/-}$ cells had no CXCR3 expression (F and L). $RAG2^{-/-}$ mice were used as

recipients instead of WT mice to avoid any contribution of radio resistant CXCR3 expressing B or T cells. Note that splenocyte counts were similar in all mice (data not shown) and these plots are representative of all mice examined (numbers of mice in Figure 4 legend).

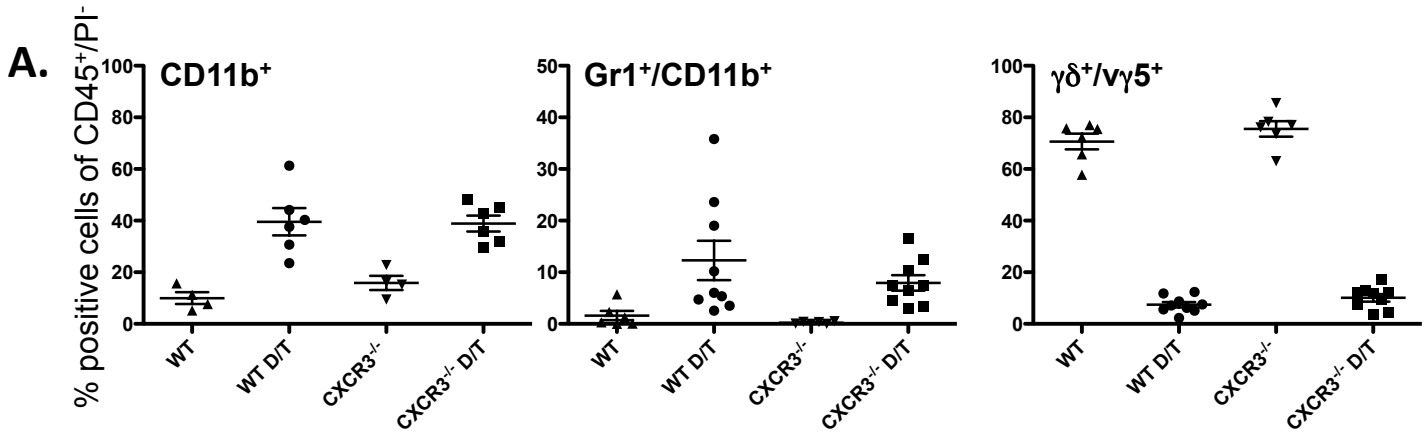
Supplementary Figure 3 FACS analysis of DMBA/TPA treated skin revealed no differences in $CXCR3^{-/-}$ compared to WT mice for $CD11b^{+}$ cells, $Gr1^{+}CD11b^{+}$cells and $\gamma\delta/v\gamma5^{+}$ cells. Epidermal preparations were generated from untreated WT and $CXCR3^{-/-}$ mice or short course DMBA/TPA treated skin (designated as D/T) and FACS analysis was performed for $CD11b^{+}$ cells, $Gr1^{+}CD11b^{+}$cells and $\gamma\delta/v\gamma5^{+}$ cells. Each point represents an individual mouse and data are expressed as percentage of cells relative to total $CD45^{+}/PI^{-}$ cells and revealed no significant reductions.



Winkler et al., Supplementary Figure 1



Winkler et al., Supplementary Figure 2



Winkler et al., Supplementary Figure 3