SUPPLEMENTAL DATA

Figure. S1. Naïve WT and $H_{1\text{-}4}$RKO mice exhibit normal immune system. Single-cell suspensions of thymus, lymph node, and spleen were prepared from WT and $H_{1\text{-}4}$RKO mice. (A) The total number of thymic, lymph node, and spleen cells was enumerated and shown as mean ± SEM of n = 10 per strain. Flow cytometric analysis of immune cell subpopulations in (B) thymus, (C) a representative dot plot of thymic double negative, double positive, and single positive CD4 and CD8, (D) lymph node, and (E) spleen of WT and $H_{1\text{-}4}$RKO mice was also performed and shown as mean ± SEM of n = 10 per strain. (F) The frequency of $Foxp3^+$ cells in thymus, spleen, and lymph node was also determined and shown as mean ± SEM of n = 10 per strain. (G) The total number of peripheral WBCs and (H) peripheral WBC differentials between WT and $H_{1\text{-}4}$RKO mice was determined and shown as mean ± SEM of n = 5 per strain. In (A, B, C, D, E, and G), significance of differences was determined by two-way ANOVA followed by Bonferroni post-hoc multiple comparison test (**, $p<0.01$; ****, $p<0.0001$) and in (F), by Mann Whitney test.

Figure. S2 HA concentration in plasma of WT and $H_{1\text{-}4}$RKO mice. HA concentrations were assessed in the plasma using an enzyme immune assay kit. Significance of differences in HA concentrations between strains were determined using the Mann Whitney test (****, $p<0.0001$). Data are shown as mean ± SEM of n = 5 per strain.

Figure. S3. Frequency of inflammatory immune cell types in the CNS and DLN of immunized $H_{1\text{-}4}$RKO mice. Mononuclear cells were isolated from the CNS and DLN of 1× immunized mice on d15 post-immunization, stimulated with PMA/ionomycin for 4 hours in the presence of Brefeldin A, stained, and analyzed by flow cytometry. (A) Total number of infiltrating cells in the CNS of immunized WT and $H_{1\text{-}4}$RKO mice. (B-E) Flow cytometric data

of the frequency of cells from CNS and (F and G) the percentage of IFN-γ^+ and IL-17^+ (gated on TCR$\gamma\delta^+$ and TCR$\alpha\beta^+CD8^+$) cells from CNS of WT and $H_{1\text{-}4}$RKO mice. (H-K) Flow cytometric data of the frequency of cells from DLN and (L) the percentage of IFN-γ^+ and IL-17^+ (gated on TCR$\alpha\beta^+CD8^+$) cells from the DLN of WT and $H_{1\text{-}4}$RKO mice. Data are shown as mean ± SEM of n = 8 per strain. In (B, C, E, F, G, H, and K) significance of differences observed was determined by two-way ANOVA followed by Bonferroni's post hoc multiple comparison test and in (A, B, E, J, and K) by Mann Whitney test.

Figure. S1

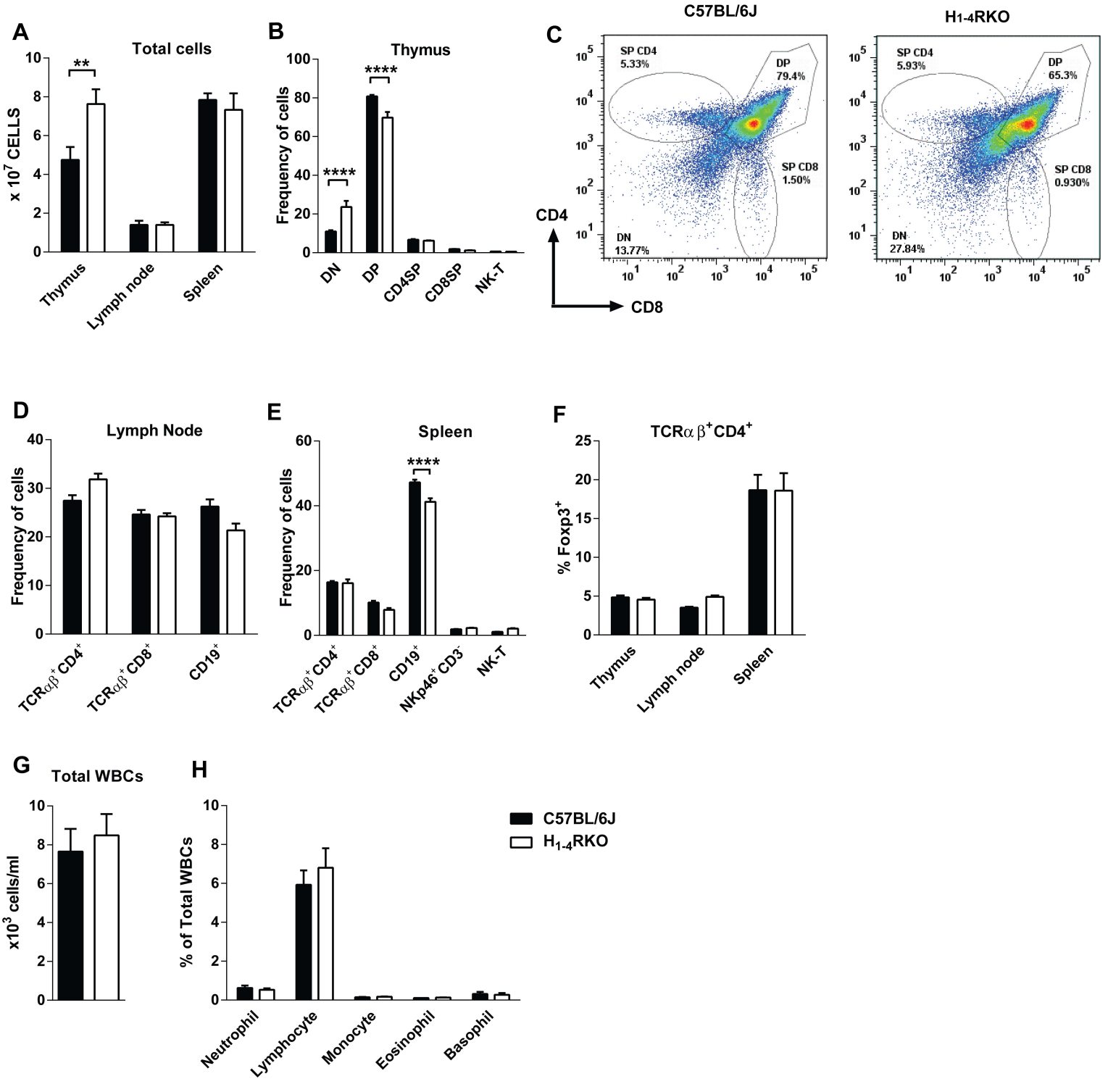

A
Total cells
x 10^7 CELLS
**
Thymus
Lymph node
Spleen
B
Thymus
Frequency of cells
****
****
DN
DP
CD4SP
CD8SP
NK-T
C
C57BL/6J
H1-4RKO
SP CD4
5.33%
DP
79.4%
SP CD8
1.50%
DN
13.77%
SP CD4
5.93%
DP
65.3%
SP CD8
0.930%
DN
27.84%
CD4
CD8
D
Lymph Node
Frequency of cells
TCRαβ+CD4+
TCRαβ+CD8+
CD19+
E
Spleen
Frequency of cells
****
TCRαβ+CD4+
TCRαβ+CD8+
CD19+
NKp46+CD3-
NK-T
F
TCRα β+CD4+
% Foxp3+
Thymus
Lymph node
Spleen
G
Total WBCs
x10^3 cells/ml
H
% of Total WBCs
Neutrophil
Lymphocyte
Monocyte
Eosinophil
Basophil
C57BL/6J
H1-4RKO

Figure. S2

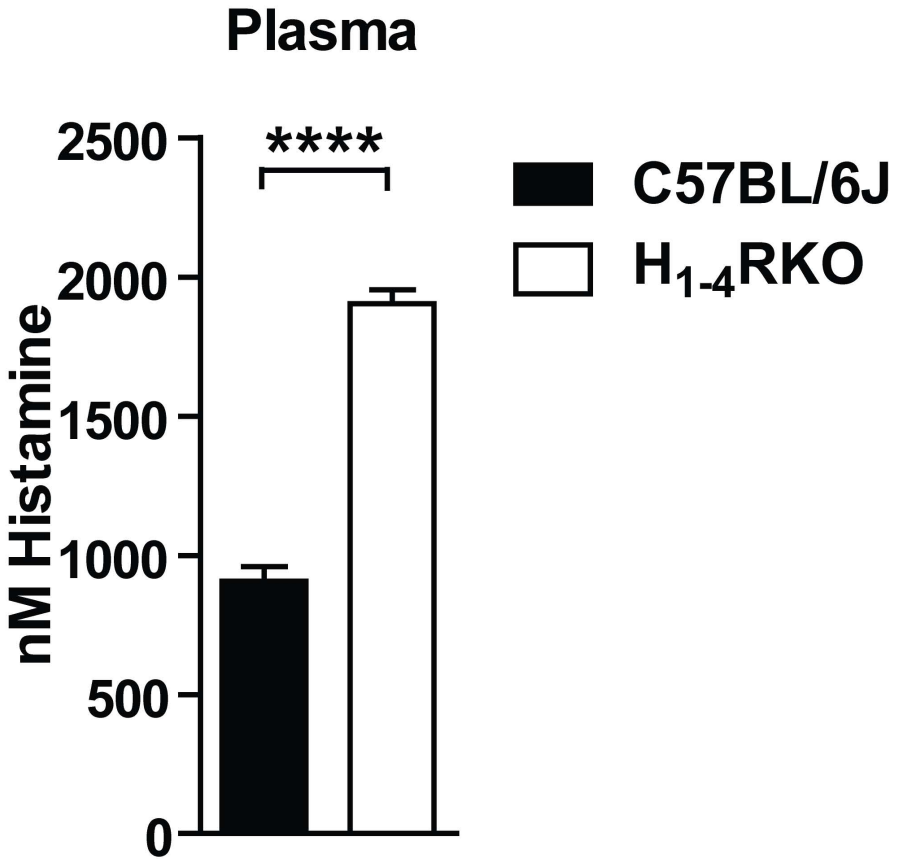
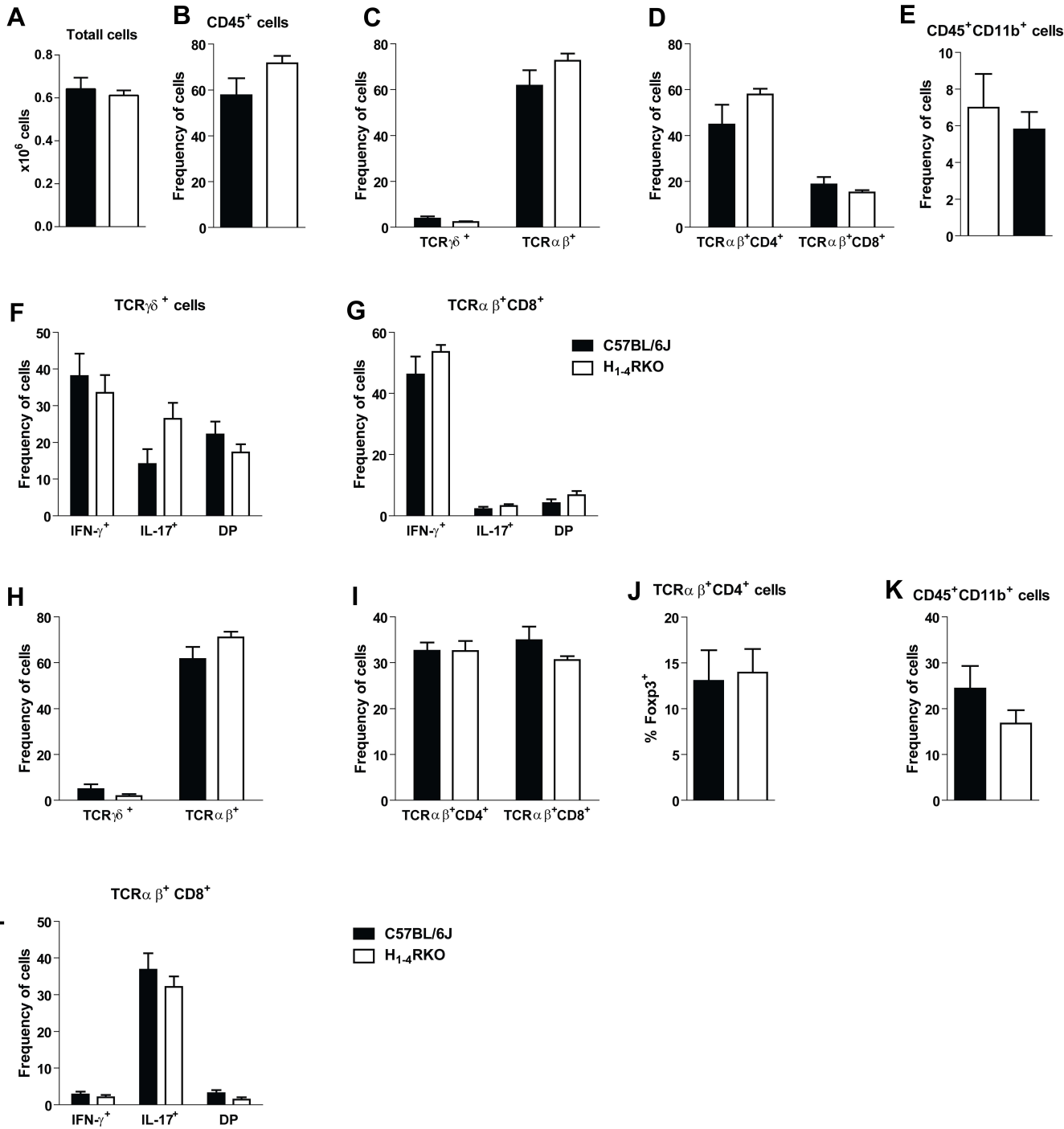

Plasma
****
2500
2000
1500
1000
500
0
nM Histamine
C57BL/6J
H1-4RKO

Figure. S3


A
Totall cells
x10^6 cells
B
CD45+ cells
Frequency of cells
C
Frequency of cells
TCRγδ+
TCRαβ+
D
Frequency of cells
TCRαβ+CD4+
TCRαβ+CD8+
E
CD45+CD11b+ cells
Frequency of cells
F
TCRγδ+ cells
Frequency of cells
IFN-γ+
IL-17+
DP
G
TCRαβ+CD8+
Frequency of cells
IFN-γ+
IL-17+
DP
C57BL/6J
H1-4RKO
H
Frequency of cells
TCRγδ+
TCRαβ+
I
Frequency of cells
TCRαβ+CD4+
TCRαβ+CD8+
J
TCRαβ+CD4+ cells
% Foxp3+
K
CD45+CD11b+ cells
Frequency of cells
L
TCRαβ+ CD8+
Frequency of cells
IFN-γ+
IL-17+
DP
C57BL/6J
H1-4RKO