Supporting Information

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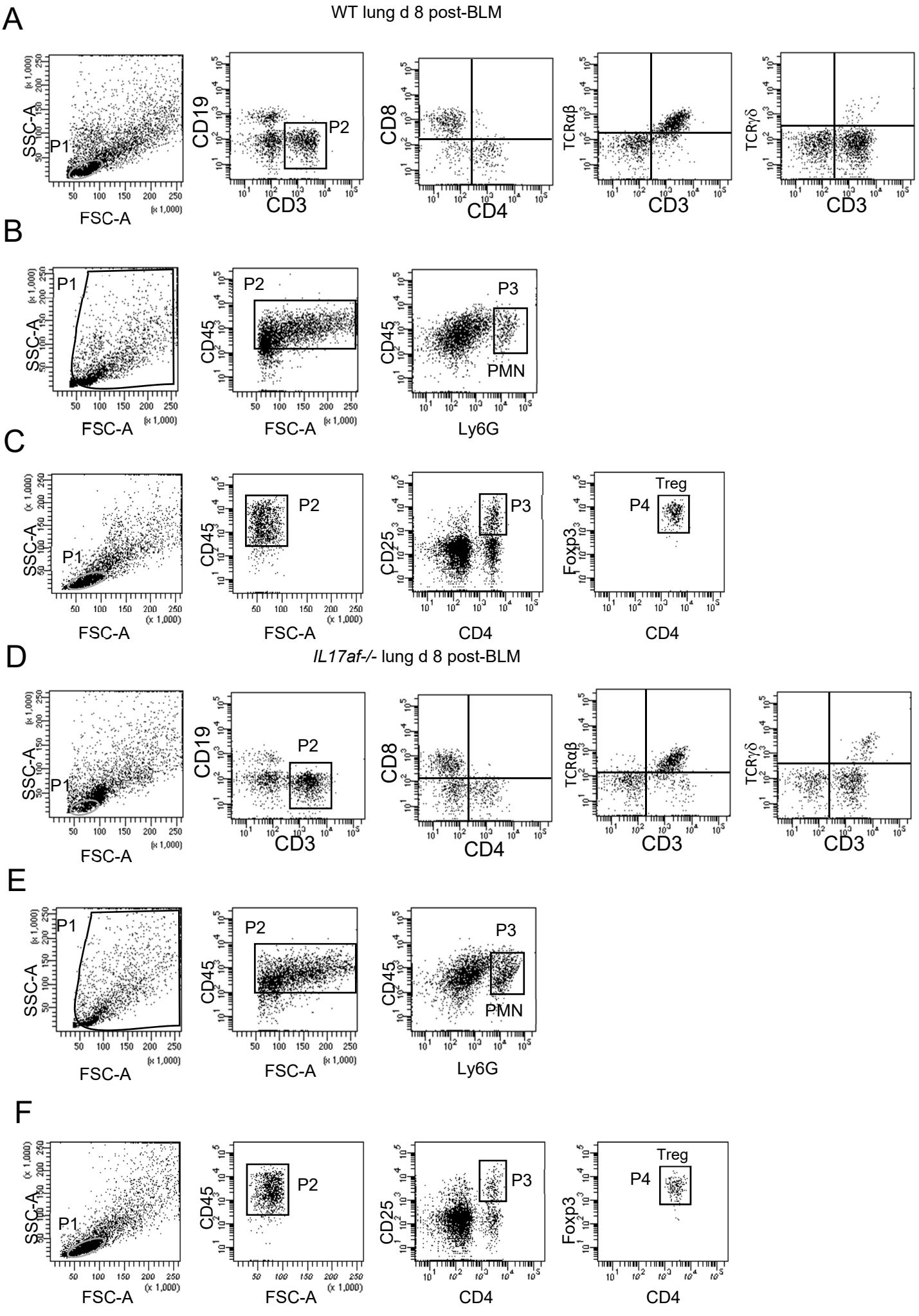
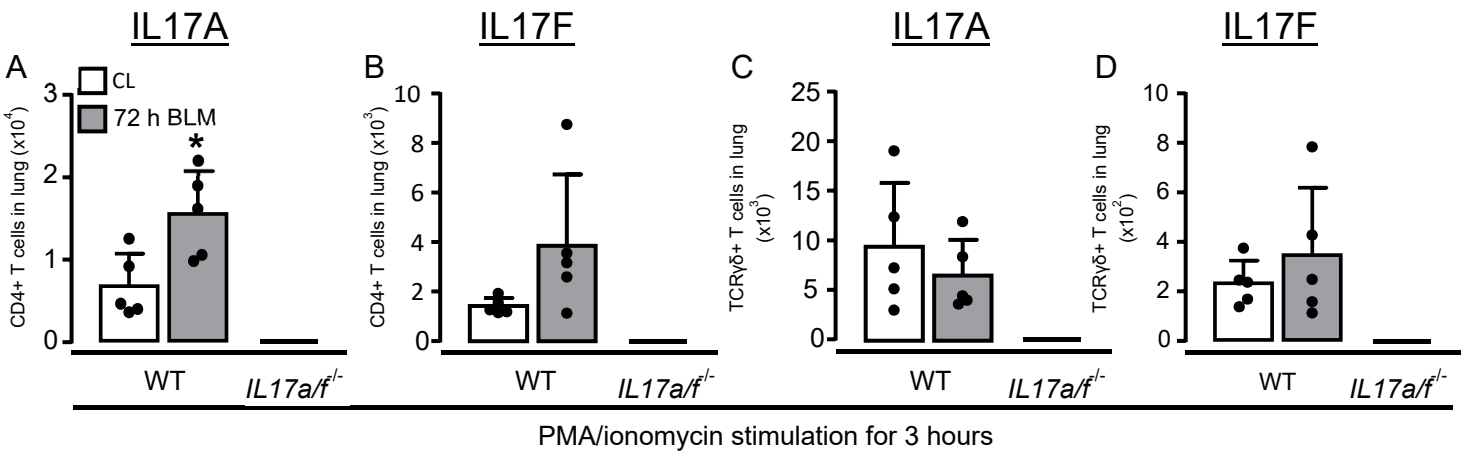
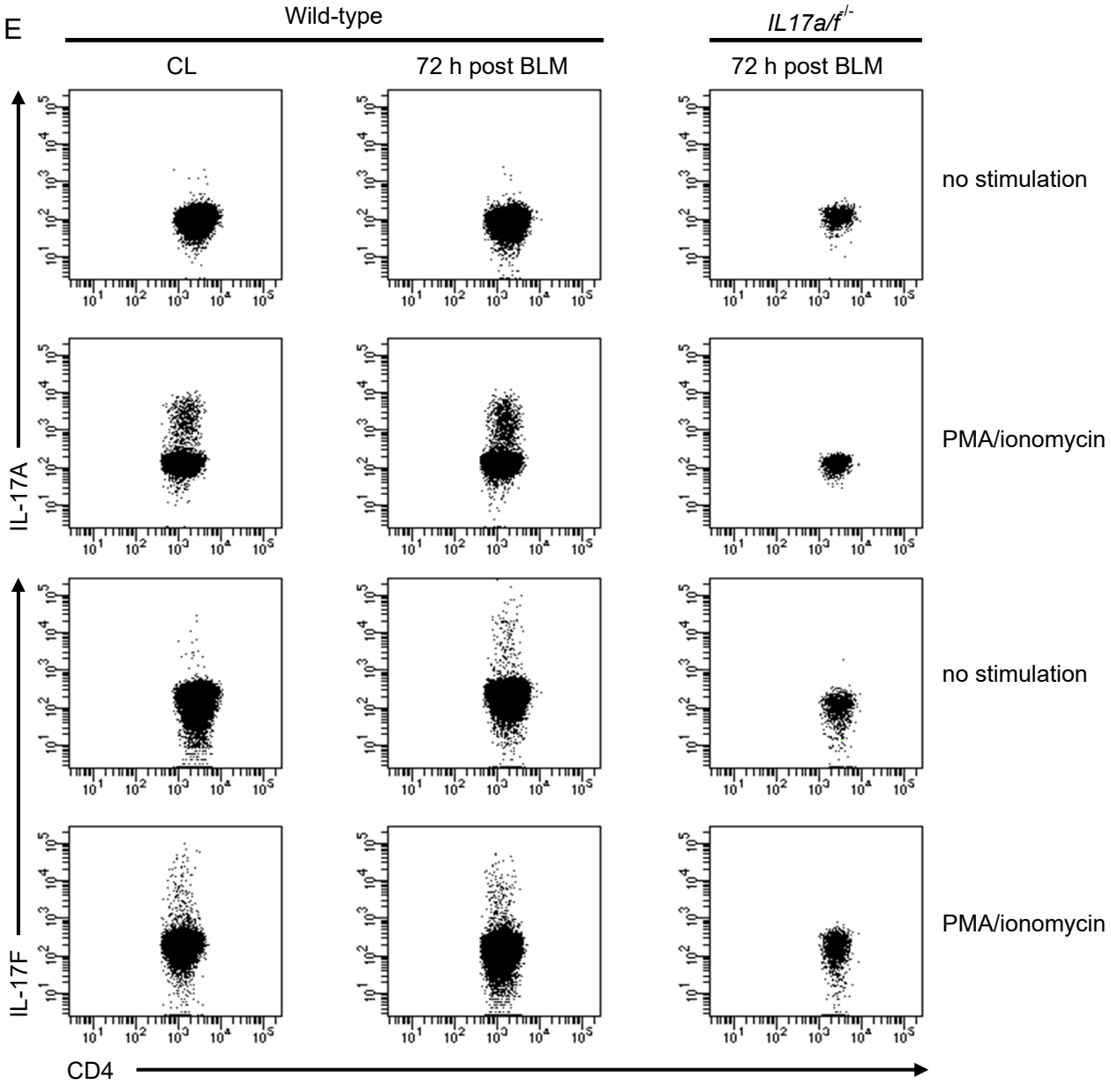
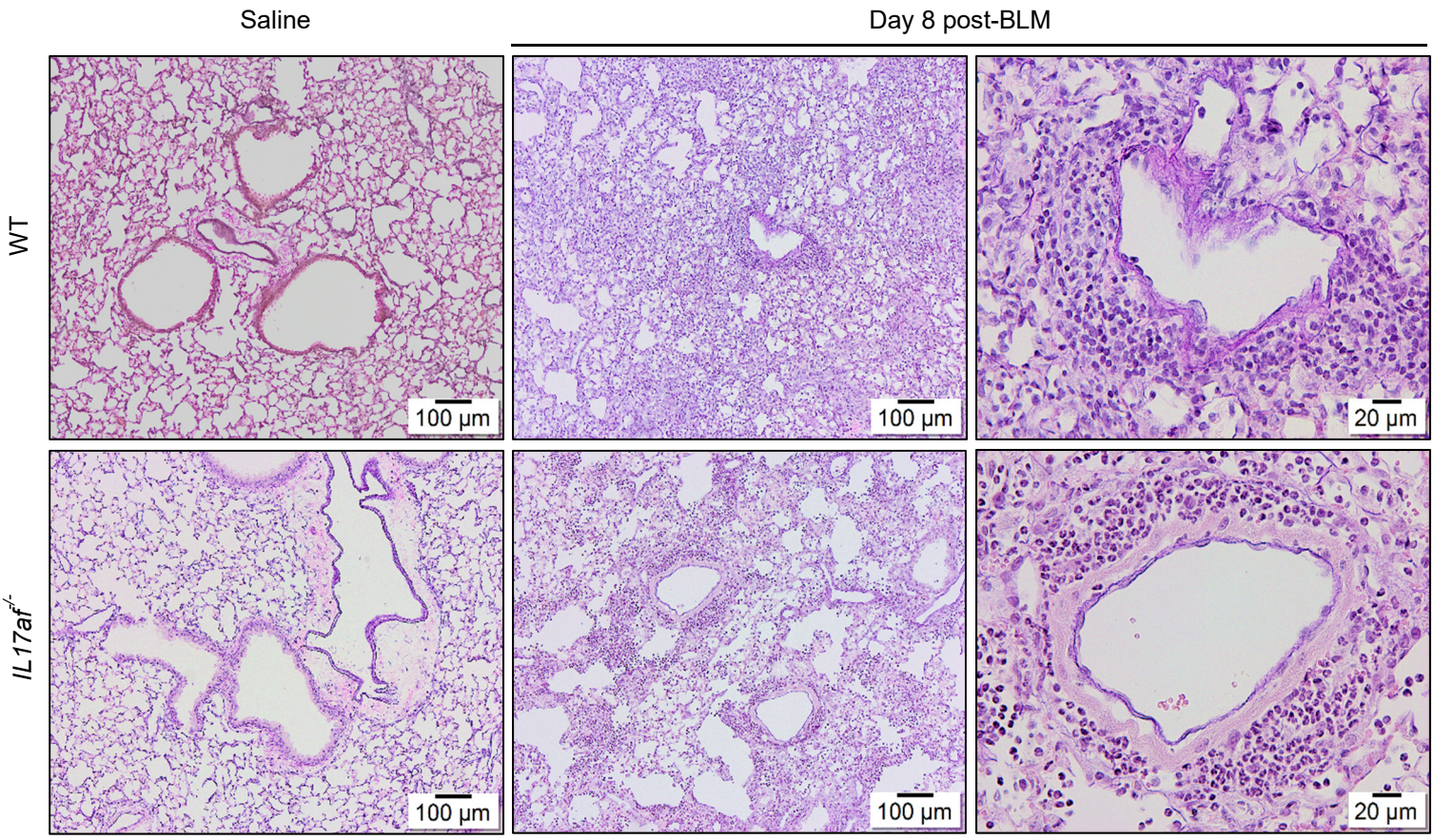

A
WT lung d 8 post-BLM
SSC-A
FSC-A
P1
CD19
CD3
P2
CD8
CD4
TCRαβ
TCRγδ
B
CD45
Ly6G
P3
PMN
C
CD25
Foxp3
Treg
P4
D
IL17af-/- lung d 8 post-BLM
E
F

Supplemental Figure S1: Representative FACS-based gating of lung leukocyte subsets of BLM treated WT and *IL17af*$^{-/-}$ mice. WT and *IL17af*$^{-/-}$ mice were treated with either saline (CL) or BLM for 6, 8 and 10 days. The Figure shows representative FACS analyses of lung leukocytes of BLM treated WT and *IL17af*$^{-/-}$ mice on day 8 post-BLM. B and T cells were identified according to their forward scatter area (FSC-A) versus side scatter area (SSC-A) (P1) followed by hierarchical sub-gating of CD3^{pos} (P2 in A,D) or CD19^{pos} lymphocyte subsets. Lung neutrophils were gated in both experimental groups according to their FSC-A/SSC-A characteristics followed by FSC-A/CD45 antigen expression (P2 in B,E) and their CD45/Ly6G antigen expression characteristics (P3 in B,E). Regulatory T cells in lungs of BLM treated WT (C) and *IL17af*$^{-/-}$ (F) mice were identified according to their FSC-A/SSC-A characteristics followed by hierarchical sub-gating of cells according to their FSC-A/CD45 (P2), CD3/CD4 (P3), CD4/CD25 (P4) and CD4/Foxp3 antigen expression (P5). Dot plots are representative of at least 5 independent analyses per experimental group.

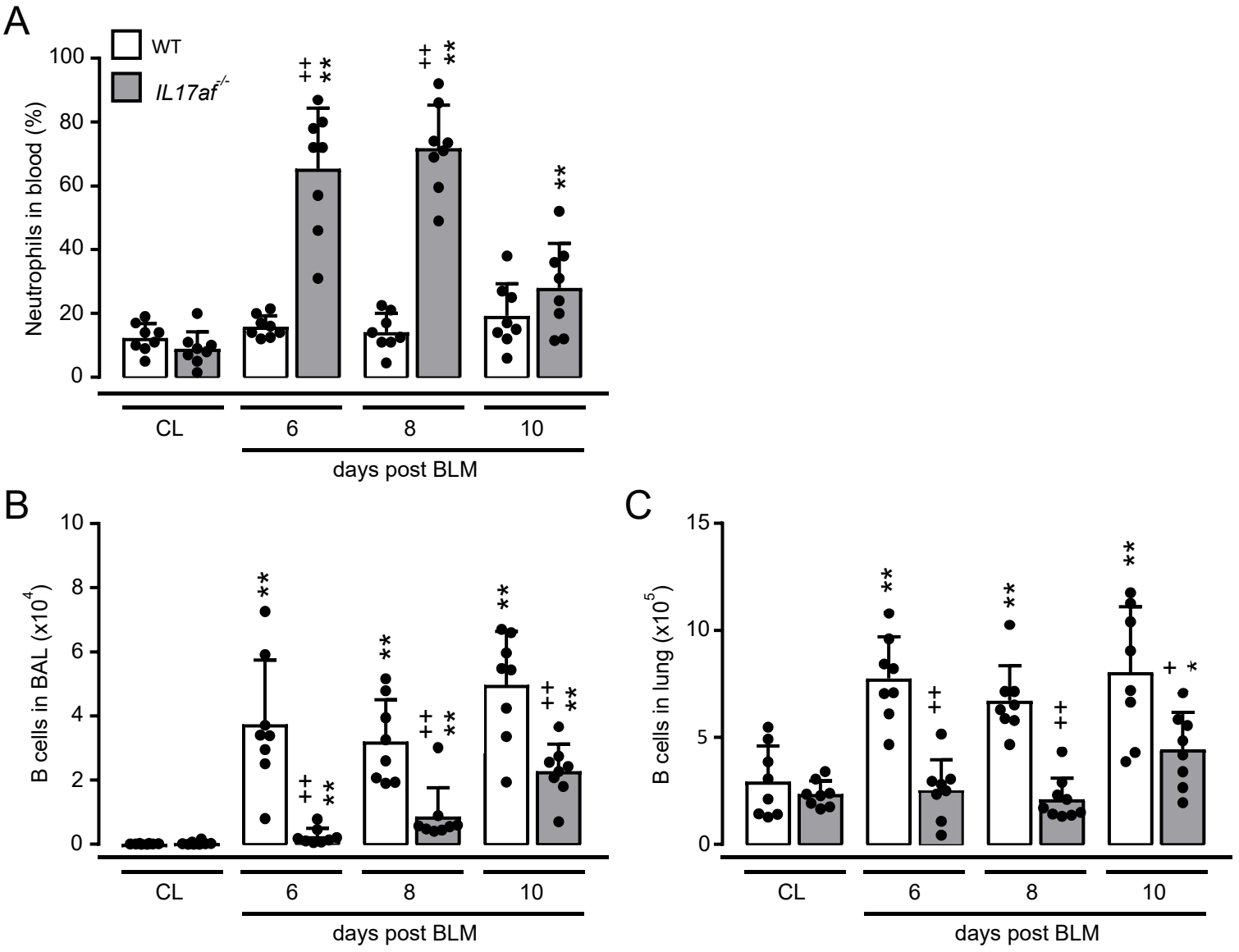

IL17A
IL17F
IL17A
IL17F
A
B
C
D
CL
72 h BLM
CD4+ T cells in lung (x10^4)
CD4+ T cells in lung (x10^3)
TCRγδ+ T cells in lung (x10^3)
TCRγδ+ T cells in lung (x10^2)
WT
IL17a/f-/-
PMA/ionomycin stimulation for 3 hours


E
Wild-type
IL17a/f-/-
CL
72 h post BLM
72 h post BLM
no stimulation
PMA/ionomycin
no stimulation
PMA/ionomycin
IL-17A
IL-17F
CD4

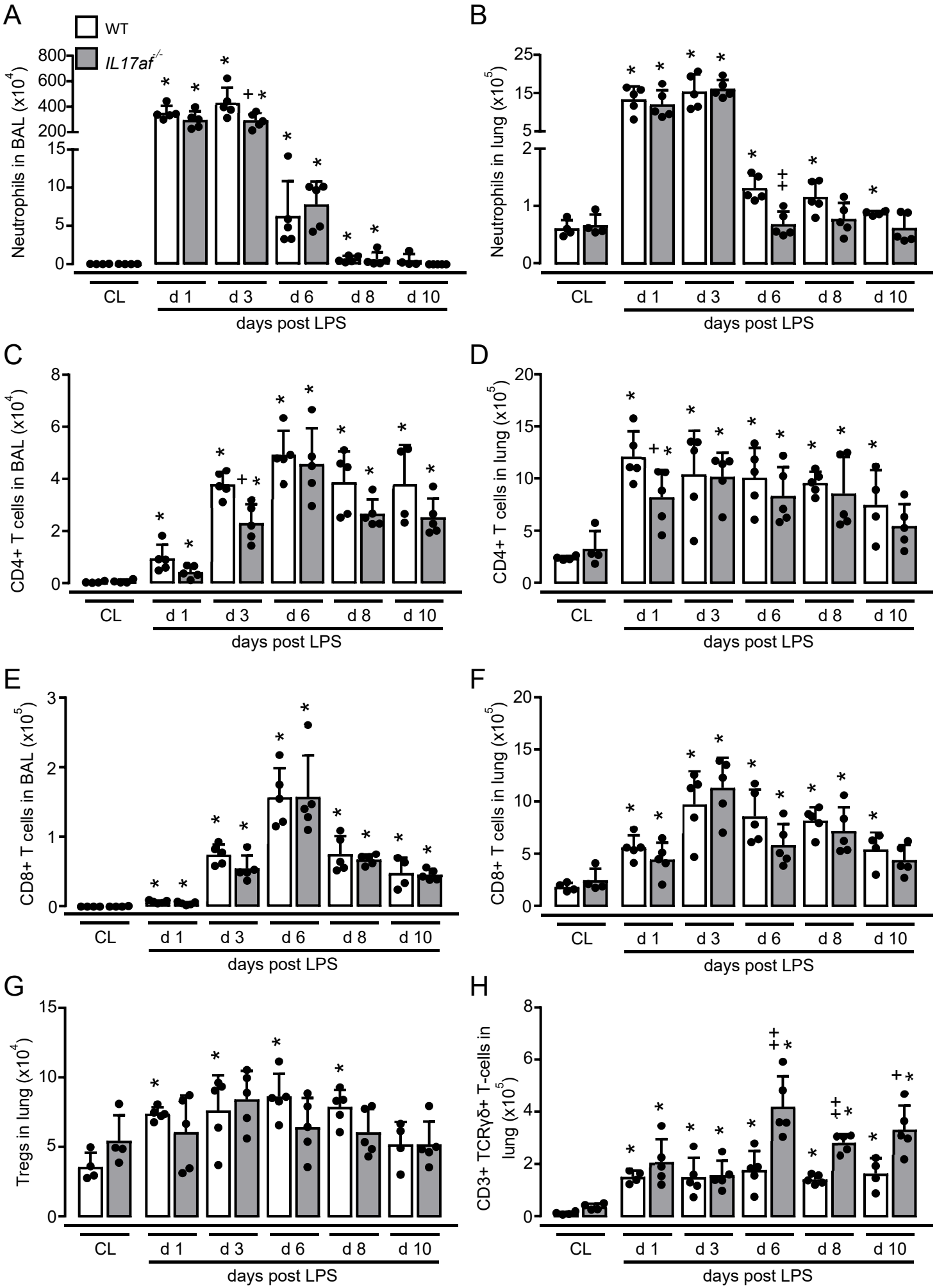
Supplemental Figure S2: Effect of PMA/ionomycin stimulation on intracellular IL17A and IL17F protein expression in T cells of naïve or BLM challenged mice. WT mice and *IL17af*$^{-/-}$ mice were treated with saline (CL, white bars) or BLM (grey bars) for 72 h. Subsequently, intracellular IL17A and IL17F protein expressions of CD4^{pos} T cells (A, B, E) and TCRγδ^{pos} T cells (C-E) were measured in lung T cell preparations by flow cytometry after stimulation of the cells with PMA/ionomycin for 3 hours. Data are shown as mean±SD of n=5 data points, with one data point representing a pool of T cells isolated from n=2-3 mice per experimental group and time point and are representative of at least two independent experiments. *$p<0.05$ versus CL (Mann-Whitney U Test).


Saline
Day 8 post-BLM
WT
IL17af-/-
100 µm
100 µm
20 µm
100 µm
100 µm
20 µm

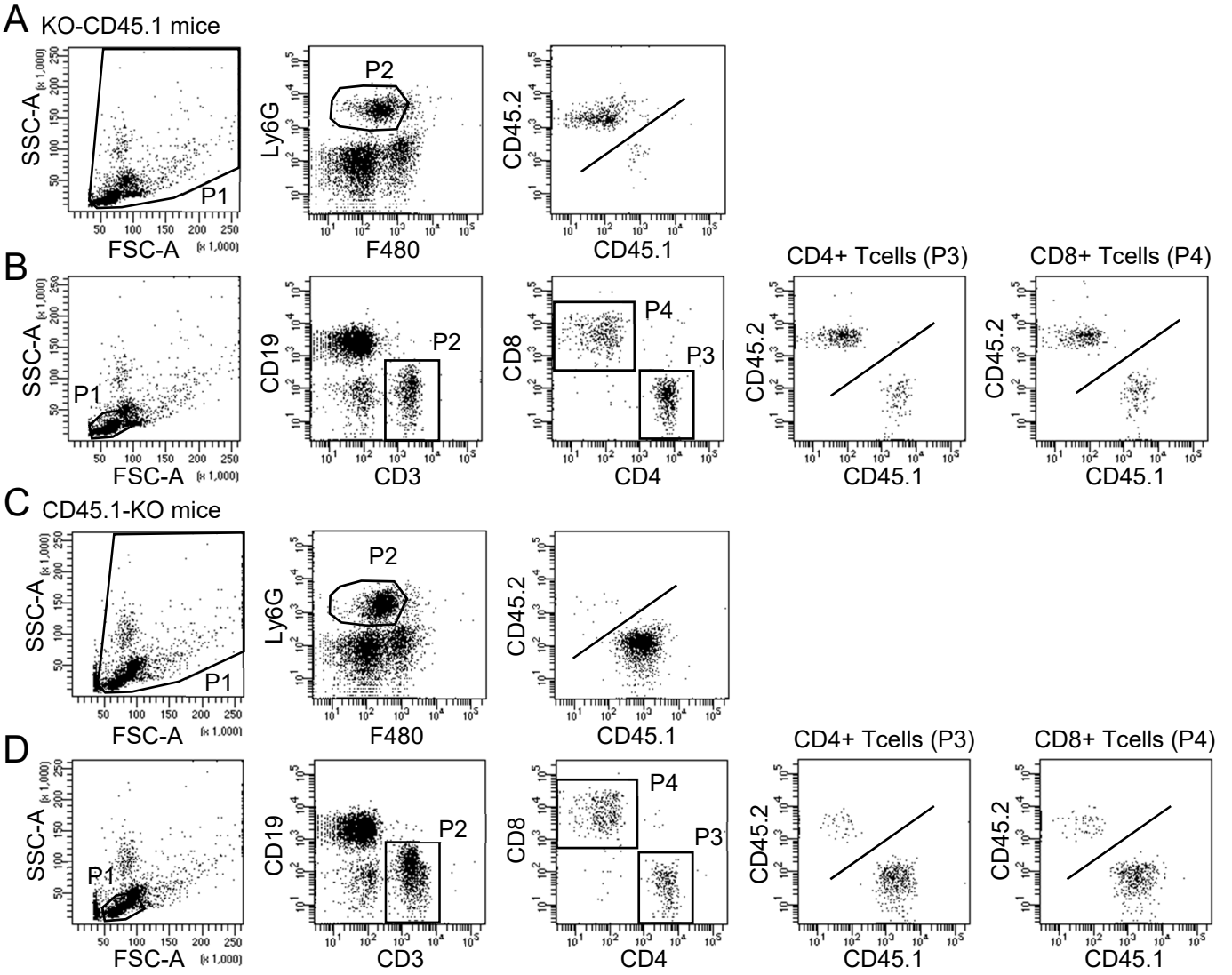
Supplemental Figure S3: Elastica-van-Gieson stained lung sections of BLM treated WT and *IL17af*$^{-/-}$ mice. WT and *IL17af*$^{-/-}$ mice were treated with either saline (CL) or BLM for various time points. The Figure shows Elastica-van-Gieson stained lung tissue sections of WT and *IL17af*$^{-/-}$ mice at day 8 post-BLM. Panels are at x 10 original magnification (scale bars, 100 μm), while the two right panels are taken at x 40 original magnification (scale bars, 20 μm).

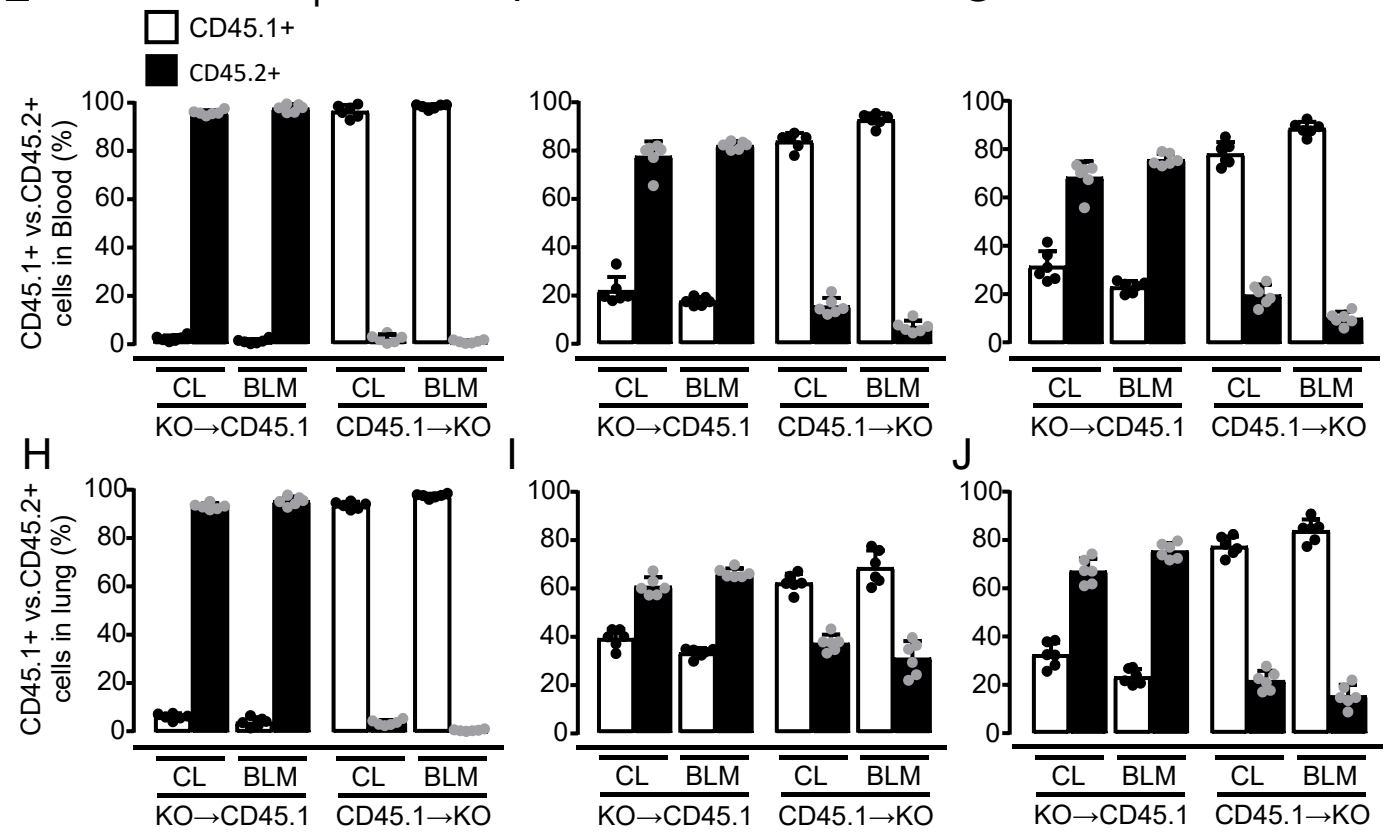

A
WT
IL17af-/-
Neutrophils in blood (%)
100
80
60
40
20
0
CL
6
8
10
days post BLM
B
B cells in BAL (x10^4)
10
8
6
4
2
0
CL
6
8
10
days post BLM
C
B cells in lung (x10^5)
15
10
5
0
CL
6
8
10
days post BLM

Supplemental Figure S4: Characterization of peripheral blood neutrophils and B cells in lungs of BLM treated WT and *IL17af*$^{-/-}$ mice. WT (white bars) and *IL17af*$^{-/-}$ mice (grey bars) were treated with either saline (CL) or BLM for 6, 8 and 10 days, as indicated. (A) Percentages of neutrophils in peripheral blood smears of BLM treated WT and *IL17af*$^{-/-}$ mice. (B, C) Immunophenotypic analysis B cells in BALF (B) and lung tissue (C) of BLM treated WT and *IL17af*$^{-/-}$ mice. Data are shown as mean±SD of n= 5-8 mice per group and time point and are representative of two independent experiments. *p≤0.05, **p≤0.01 compared with mice from the control group. $^{+}$p≤0.05, $^{++}$p≤0.01 IL17af-/- relative to WT mice (Mann-Whitney U Test).


A
WT
IL17af-/-
Neutrophils in BAL (x10^4)
B
Neutrophils in lung (x10^5)
C
CD4+ T cells in BAL (x10^4)
D
CD4+ T cells in lung (x10^5)
E
CD8+ T cells in BAL (x10^5)
F
CD8+ T cells in lung (x10^5)
G
Tregs in lung (x10^4)
H
CD3+ TCRγδ+ T-cells in lung (x10^5)
CL
d 1
d 3
d 6
d 8
d 10
days post LPS

Supplemental Figure S5: Characterization of leukocyte subsets in BALF and lung tissue of LPS treated WT and *IL17af*$^{-/-}$ mice. WT (white bars) and *IL17af*$^{-/-}$ mice (grey bars) were treated with either PBS (CL) or LPS for 1, 3, 6, 8 and 10 days, as indicated. Immunophenotypic analysis of neutrophils (A, B), CD4^{pos} T cells (C, E), CD8^{pos} T cells (D, F) in BALF and lung tissue of LPS treated WT and *IL17af*$^{-/-}$ mice. (G, H) Numbers of CD4^{pos}/CD25^{pos}/FoxP3^{pos} Tregs (G) and TCRγδ^{pos} T cells (H) in lung tissue of LPS treated WT and *IL17af*$^{-/-}$ mice. Data are shown as mean ±SD of n= 4-5 mice per group and time point and are representative of two independent experiments. *p≤0.05 compared with mice from the control group. $^{+}$p≤0.05, $^{++}$p≤0.01 *IL17af*$^{-/-}$ relative to WT mice (Mann-Whitney U Test).


A KO-CD45.1 mice
SSC-A
FSC-A
P1
P2
Ly6G
F480
CD45.2
CD45.1
B
CD19
CD3
CD8
CD4
P3
P4
CD4+ Tcells (P3)
CD8+ Tcells (P4)
C CD45.1-KO mice
D


E Neutrophils
F CD4+ T-cells
G CD8+ T-cells
CD45.1+
CD45.2+
CD45.1+ vs.CD45.2+ cells in Blood (%)
CL
BLM
KO→CD45.1
CD45.1→KO
H
I
J
CD45.1+ vs.CD45.2+ cells in lung (%)

Supplemental Figure S6: Immunophenotypic analysis of bone marrow engraftment of different leukocyte subsets in chimeric KO→CD45.1 and CD45.1→KO mice. (A-D) Representative FACS analysis of CD45.1 versus CD45.2 alloantigen expression on neutrophils and T cells in BLM treated chimeric KO→CD45.1 (A, B) and CD45.1→KO (C, D) mice. (A, C) Neutrophils were gated according to their FSC-A/ SSC-A characteristics (P1) and Ly6G cell surface expression (P2), followed by analysis of their CD45.1 versus CD45.2 alloantigen expression. (B, D) T cells were gated according to their FSC-A versus SSC-A characteristics (P1) and their $CD3^{pos}$ immunophenotype (P2), and were then analyzed according to their $CD4^{pos}$ (P3) versus $CD8^{pos}$ (P4) cell surface expression followed by analysis of alloantigenic CD45.1 versus CD45.2 expression. (E-J) Analysis of CD45.1 (white bars) versus CD45.2 (black bars) alloantigen expressing neutrophils (E, H), $CD4^{pos}$ T cells (F, I) and $CD8^{pos}$ T cells (G, J) in blood (E-G) and lung tissue (H-J) of chimeric KO→CD45.1 and CD45.1→KO mice exposed to saline (CL) or BLM. Data are shown as mean ±SD of n= 6 mice per group and time point.