Supplemental Figure Legends

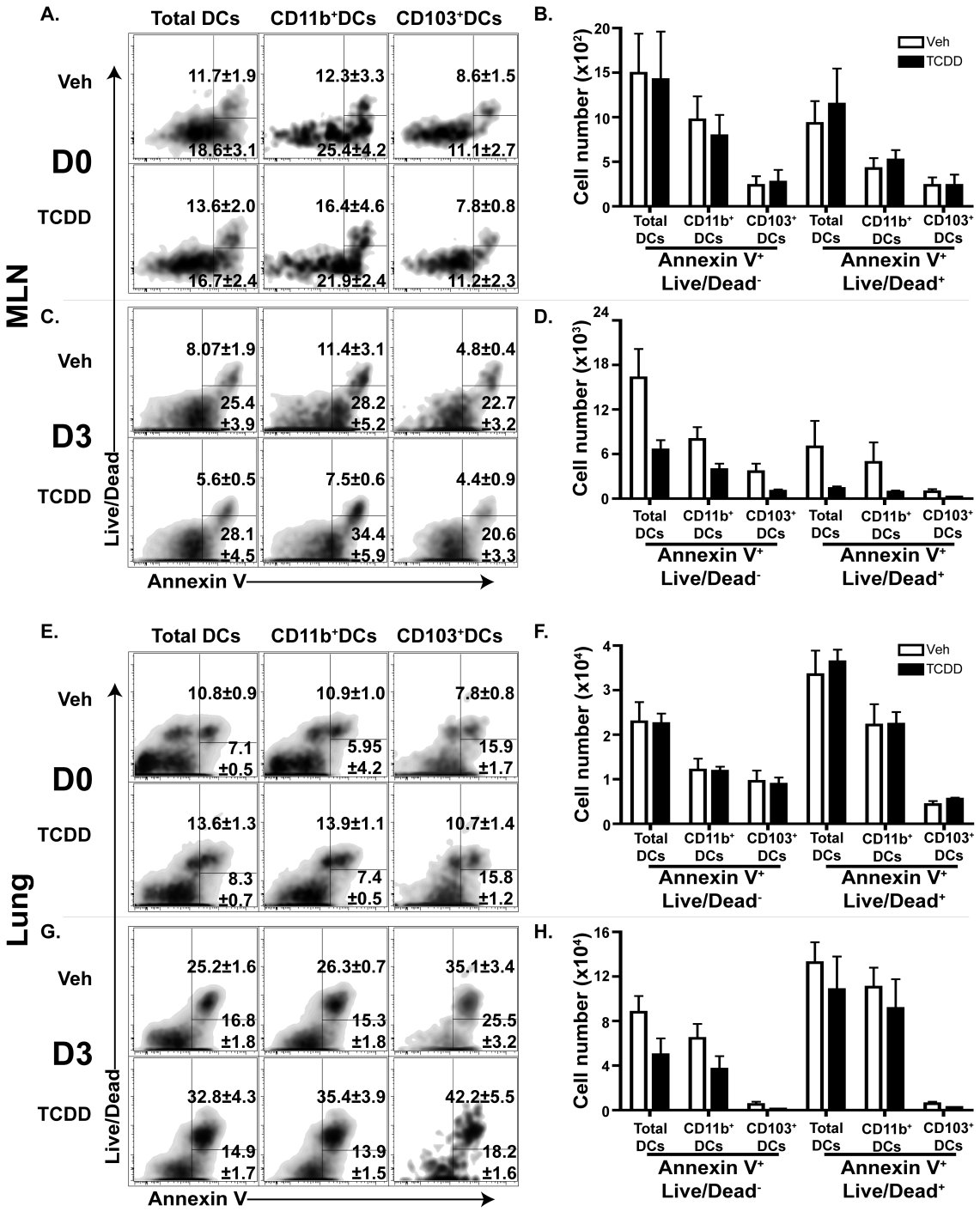
Supplemental Figure 1. Comparison of apoptosis in DCs from MLN and lung. Female B6 mice were dosed orally with peanut oil (vehicle-treated group, Veh, open bars) or 10 μg/kg of TCDD (TCDD-treated group, TCDD, closed bars). One day later, treated mice were either sacrificed (naïve group, D0) or infected (i.n.) with influenza virus. Infected mice were sacrificed 3 days later (D3). (A-B) MLN and (C-D) lung cells were collected and stained for flow cytometric analysis, as described in Fig. 1 and Fig. 4 (but not excluded Live/Dead$^+$ cells) respectively, with the addition of Annexin-V to the staining reagents. (A,C) Representative plots show the percentage of early apoptotis (Annexin V$^+$ Live/Dead$^-$) and late apoptosis (Annexin V$^+$ Live/Dead$^+$) for total DCs and the indicated DC subsets from MLN of naïve (D0) and infected (D3) mice. (B,D) The bar graphs show the number of total DCs, CD11b$^+$DCs and CD103$^+$DCs in the MLN that are Annexin-V positive and Live/Dead negative or Annexin-V and Live/Dead double positive at D0 and D3. (E,G) Representative plots show the percentage of early and late apoptotic DCs from lungs of naïve and infected mice. (F,H) The bar graphs show the number of total DCs, CD11b$^+$DCs and CD103$^+$DCs in the lung that are Annexin-V positive and Live/Dead negative or Annexin-V and Live/Dead double positive at D0 and D3. There were no significant differences in Annexin-V single positive events in any DC subsets from V and T treated, naïve or infected mice (not shown). Data are shown as mean ± SEM (n = 5/group) from one experiment.

Supplemental Figure 2. Comparison of DC subsets in influenza infected wild-type and global Ahr knockout mice. Female wild-type ($Ahr^{d/d}$) and $Ahr^{-/-}$ mice were treated and infected as in Fig 6. MLN and lung-derived immune cells were stained with mAbs 3 days after infection. DCs in the (A) MLN and (B) lung were analyzed as described in Fig. 1 and Fig. 4. Numbers on the dot plots depict the percentage of cells in the gated region. Bar graphs show the number of indicated population of cells in (A) MLN and (B) lung. Data are shown as mean ± SEM (n=4-5/group) from one experiment.. *$p < 0.05$, ***$p < 0.001$, two-tailed unpaired Student's t-test.

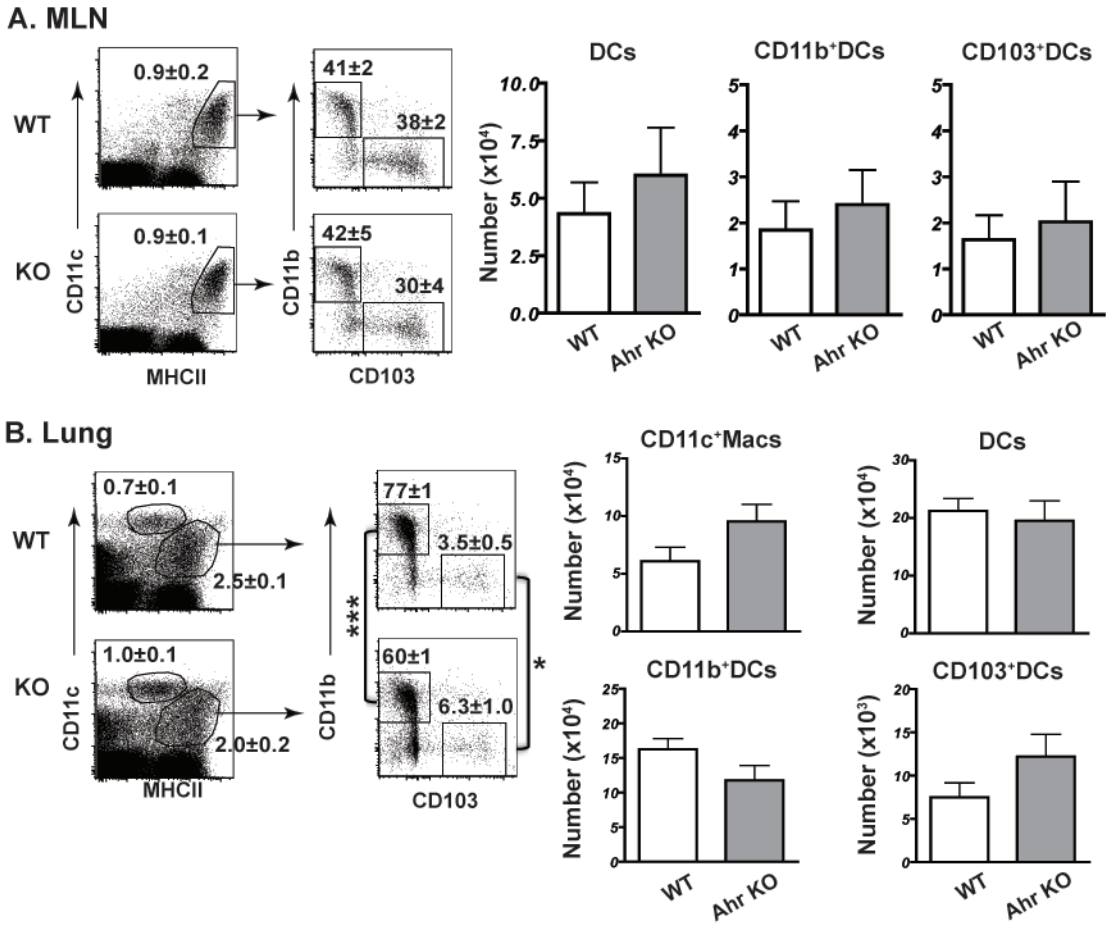
Supplemental Figure 3. *Ahr* gene expression in $CD11c^+$ subsets. (A) $CD11c^+$Macs and DCs were sorted from wild-type mice as described in Fig. 5, and $CD11b^+$ and $CD103^+$DCs were sorted as described in Fig. 3. Purity was ≥95%. Total RNA was extracted, RNA amplification and RT was performed using the Ovation Pico SL WTA v2 kit (NuGEN Technologies, Inc., San Carlos, CA) and *Ahr* gene expression was examined by quantitative real time PCR. (B) Gene expression normalized to *L13 (ΔCT)* is shown for each subpopulation. Total RNA from mouse liver was used as a positive control. Data are shown as mean ± SD (not SEM? We always use SEM) (n= 2 pools, where each pool consisted of cells from at least 12 mice) from one experiment.

Supplemental Figure 4. Gating strategy for flow cytometric data

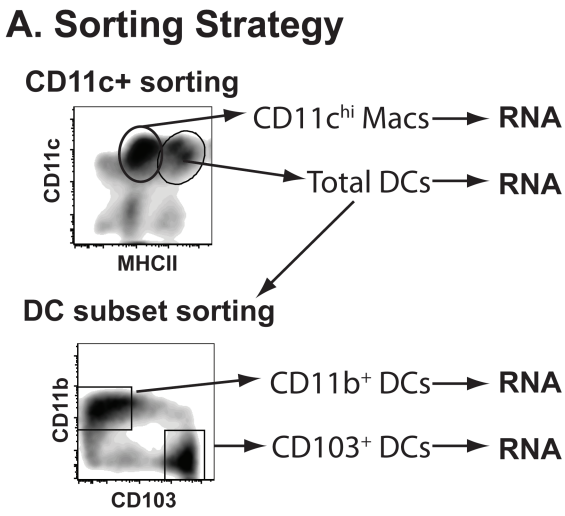
sFigure 1

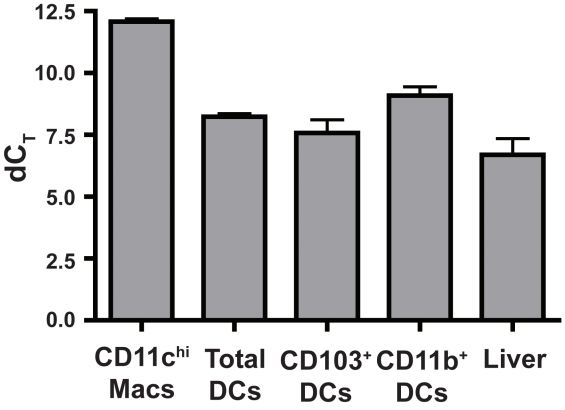

A.
Total DCs
CD11b+DCs
CD103+DCs
MLN
Veh
D0
TCDD
11.7±1.9
12.3±3.3
8.6±1.5
18.6±3.1
25.4±4.2
11.1±2.7
13.6±2.0
16.4±4.6
7.8±0.8
16.7±2.4
21.9±2.4
11.2±2.3
B.
Cell number (x10²)
Veh
TCDD
Total DCs
CD11b+ DCs
CD103+ DCs
Annexin V+ Live/Dead-
Annexin V+ Live/Dead+
C.
Veh
D3
TCDD
8.07±1.9
11.4±3.1
4.8±0.4
25.4 ±3.9
28.2 ±5.2
22.7 ±3.2
5.6±0.5
7.5±0.6
4.4±0.9
28.1 ±4.5
34.4 ±5.9
20.6 ±3.3
Live/Dead
Annexin V
D.
Cell number (x10³)
E.
Total DCs
CD11b+DCs
CD103+DCs
Lung
Veh
D0
TCDD
10.8±0.9
10.9±1.0
7.8±0.8
7.1 ±0.5
5.95 ±4.2
15.9 ±1.7
13.6±1.3
13.9±1.1
10.7±1.4
8.3 ±0.7
7.4 ±0.5
15.8 ±1.2
F.
Cell number (x10⁴)
Veh
TCDD
G.
Veh
D3
TCDD
25.2±1.6
26.3±0.7
35.1±3.4
16.8 ±1.8
15.3 ±1.8
25.5 ±3.2
32.8±4.3
35.4±3.9
42.2±5.5
14.9 ±1.7
13.9 ±1.5
18.2 ±1.6
Live/Dead
Annexin V
H.
Cell number (x10⁴)

sFigure 2


A. MLN
WT
KO
0.9±0.2
0.9±0.1
41±2
38±2
42±5
30±4
CD11c
MHCII
CD11b
CD103
DCs
CD11b+DCs
CD103+DCs
Number (x10⁴)
WT
Ahr KO
B. Lung
0.7±0.1
2.5±0.1
1.0±0.1
2.0±0.2
77±1
3.5±0.5
60±1
6.3±1.0
***
*
CD11c+Macs
DCs
CD11b+DCs
CD103+DCs
Number (x10³)

sFigure 3


A. Sorting Strategy
CD11c+ sorting
CD11c
MHCII
CD11c^hi Macs
RNA
Total DCs
RNA
DC subset sorting
CD11b
CD103
CD11b+ DCs
RNA
CD103+ DCs
RNA


B. Ahr expression
12.5
10.0
7.5
5.0
2.5
0.0
dC_T
CD11c^hi Macs
Total DCs
CD103+ DCs
CD11b+ DCs
Liver

sFigure 4

Gating Strategy