Supplemental Figure 1

DNase treatment does not significantly impact survival following IAV infection.

Aged (18-22 months of age) and young (2-4 months of age) C57BL/6 mice were intranasally inoculated with PR8 strain influenza virus. DNase was administered from day 6-12 post-infection as described in Methods. There was no significant alteration in survival in either aged (a, P = 0.54, n= 8-10/group) or young (b, p = 0.97, n = 8-10/group) mice as compared to controls.

Supplemental Figure 2

Aging leads to increase in increase in IL-1β, TNF-α, IL-17 but not CXCl5 following IAV infection.

Aged (18-22 months of age) and young (2-4 months of age) C57BL/6 mice were intranasally inoculated with PR8 strain influenza virus. BAL was collected before infection and days 3, 6, 9 post infection. IL-1β (a), TNF-α (b), IL-17 (c) and CXCL5 (d) levels in BAL were measured by ELISA. $^{**}P < 0.01$, $^{***}P < 0.001$ (Mann-Whitney test). Data are pooled from of two independent experiments. Data is represented as ±SEM.

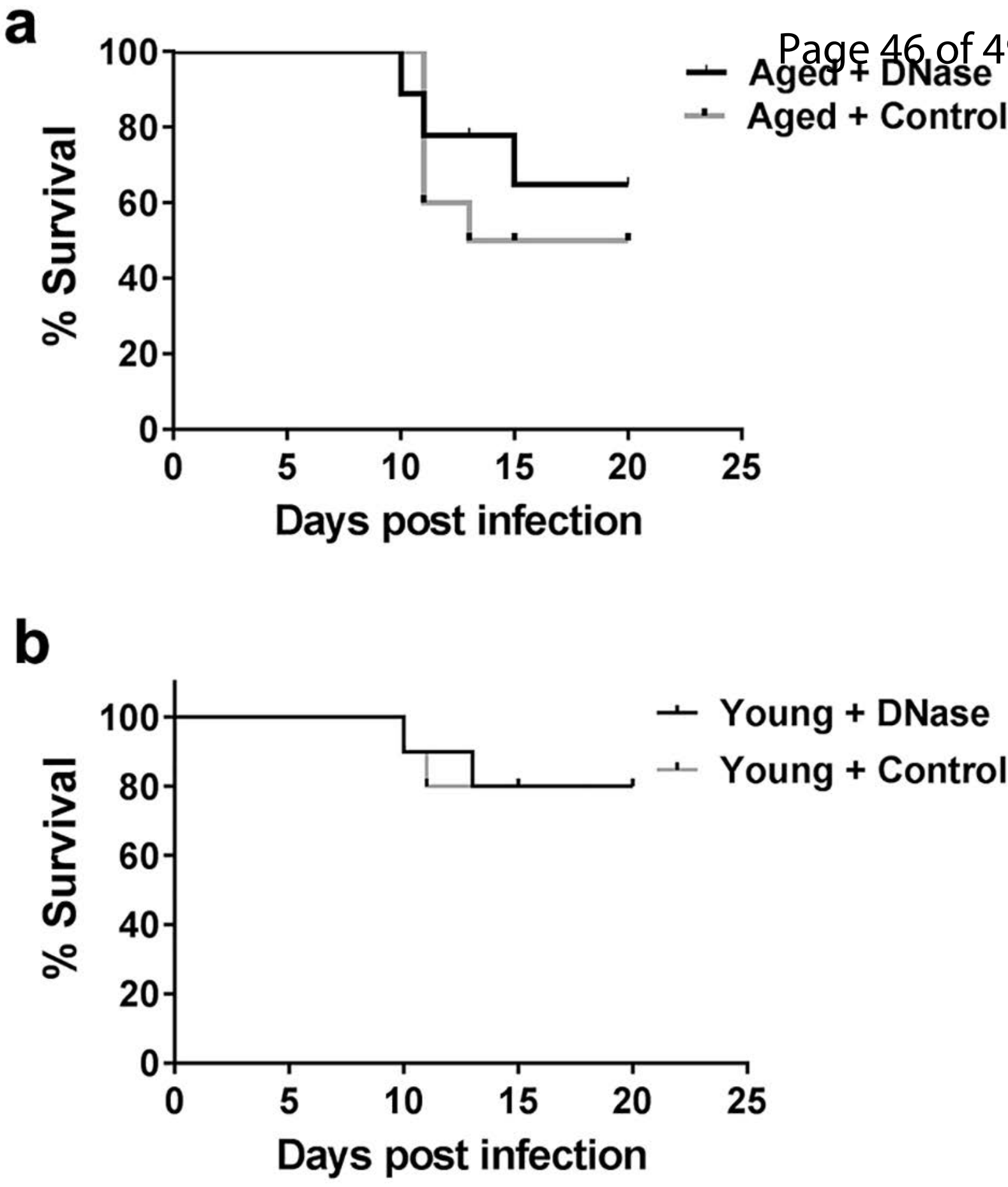
Supplemental Figure 3

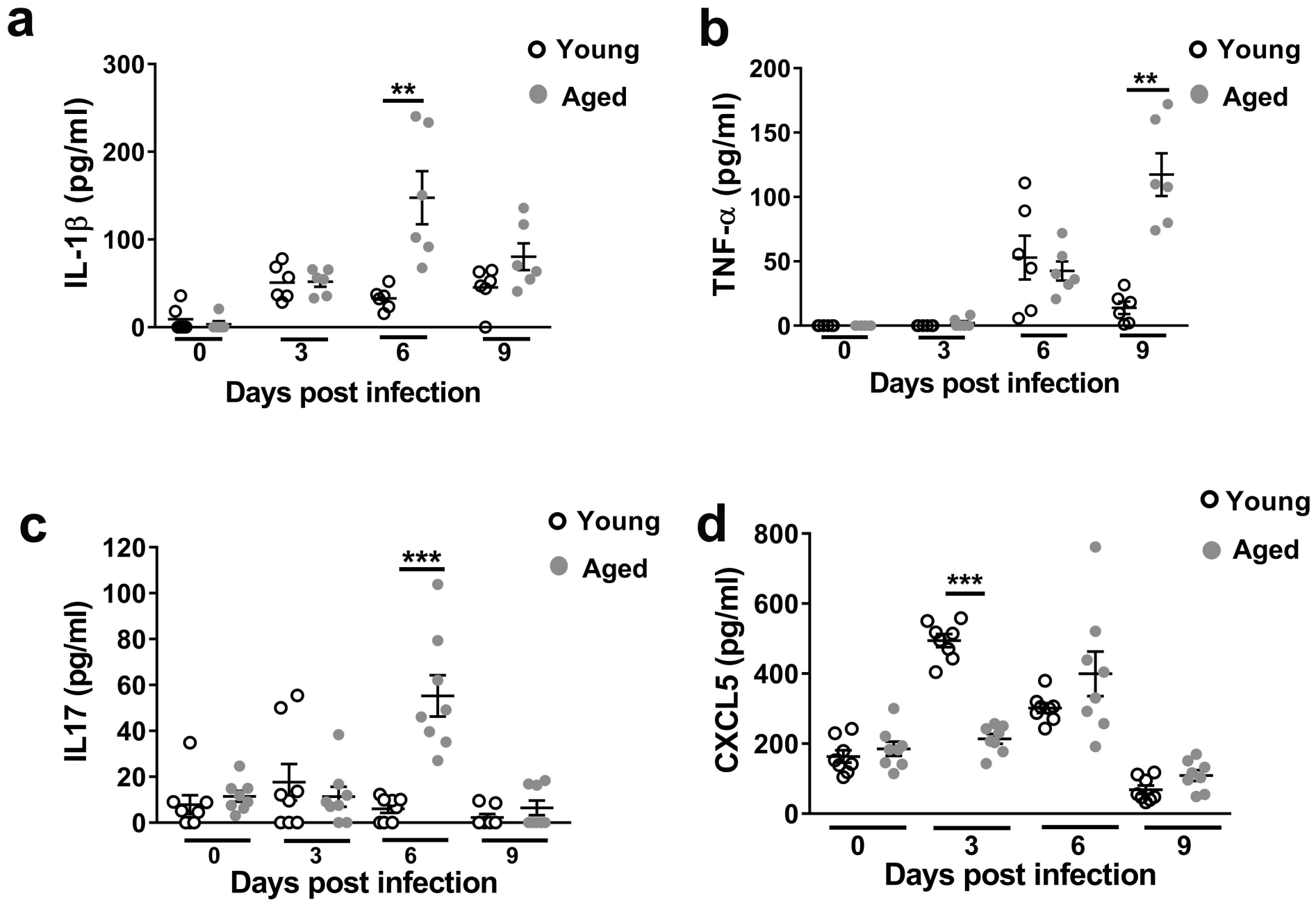
Aging leads to reduced expression of CXCR2 on neutrophils during IAV infection.

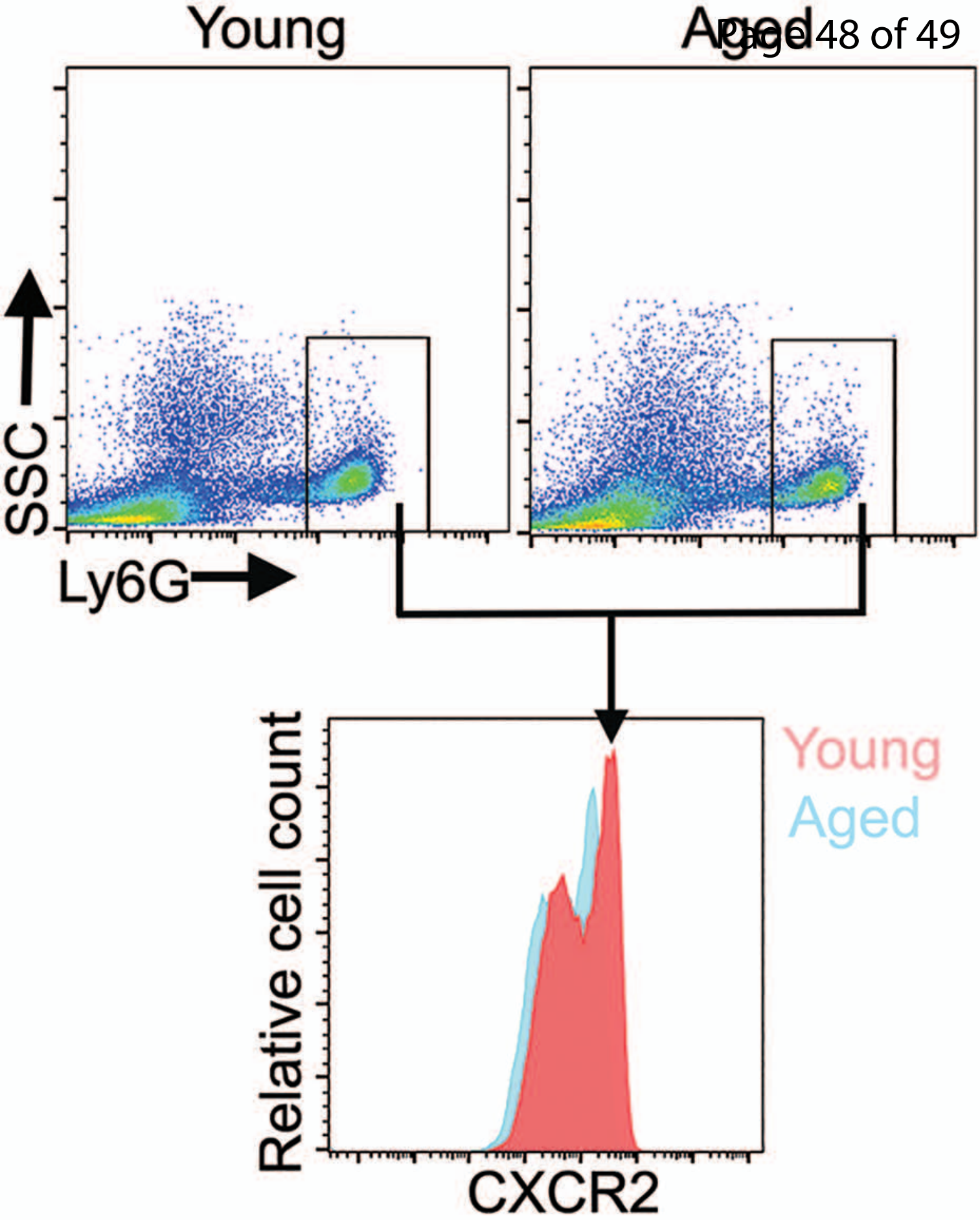
Neutrophils from the bone marrow of young and aged non-infected mice were stained with anti-Ly6G antibody and CXCR2 expression was measured by fluorescent antibody staining coupled to flow cytometry. Representative gating on neutrophil population with histogram showing median fluorescence intensity of CXCR2 expression on neutrophils is shown.

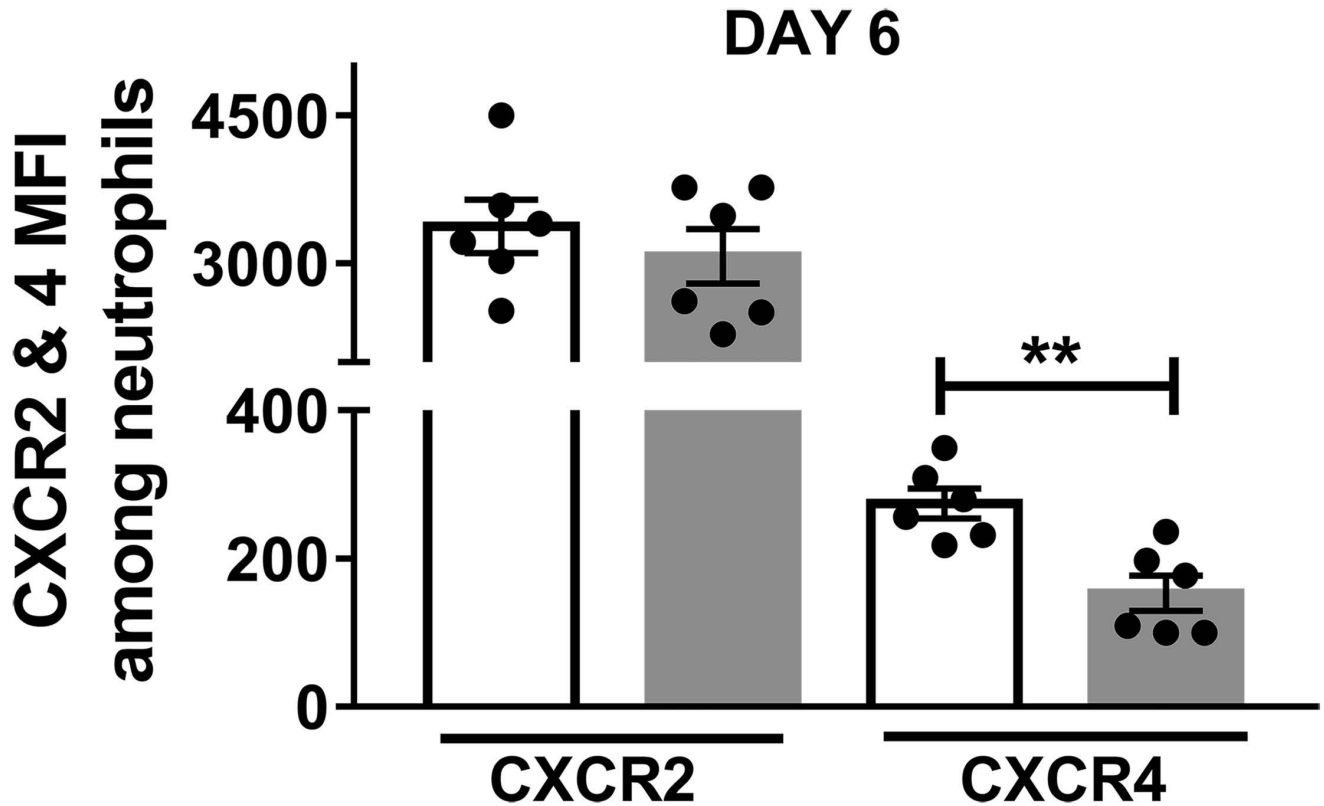
Supplemental Figure 4

Surface expression of CXCR2 and CXCR4 on bone marrow neutrophils on day 6 p.i. ** $P < 0.01$ (Mann-Whitney test) Data representative of one of two independent experiments, which yielded similar results. Data are expressed as mean ± SEM.


a
100
80
60
40
20
0
% Survival
0
5
10
15
20
25
Days post infection
Aged + DNase
Aged + Control
b
100
80
60
40
20
0
% Survival
0
5
10
15
20
25
Days post infection
Young + DNase
Young + Control


a
Young
Aged
IL-1β (pg/ml)
300
200
100
0
**
0
3
6
9
Days post infection
b
Young
Aged
TNF-α (pg/ml)
200
150
100
50
0
**
0
3
6
9
Days post infection
c
Young
Aged
IL17 (pg/ml)
120
100
80
60
40
20
0
***
0
3
6
9
Days post infection
d
Young
Aged
CXCL5 (pg/ml)
800
600
400
200
0
***
0
3
6
9
Days post infection


Young
Aged
SSC
Ly6G
Relative cell count
CXCR2
Young
Aged


DAY 6
CXCR2 & 4 MFI among neutrophils
4500
3000
400
200
0
**
CXCR2
CXCR4