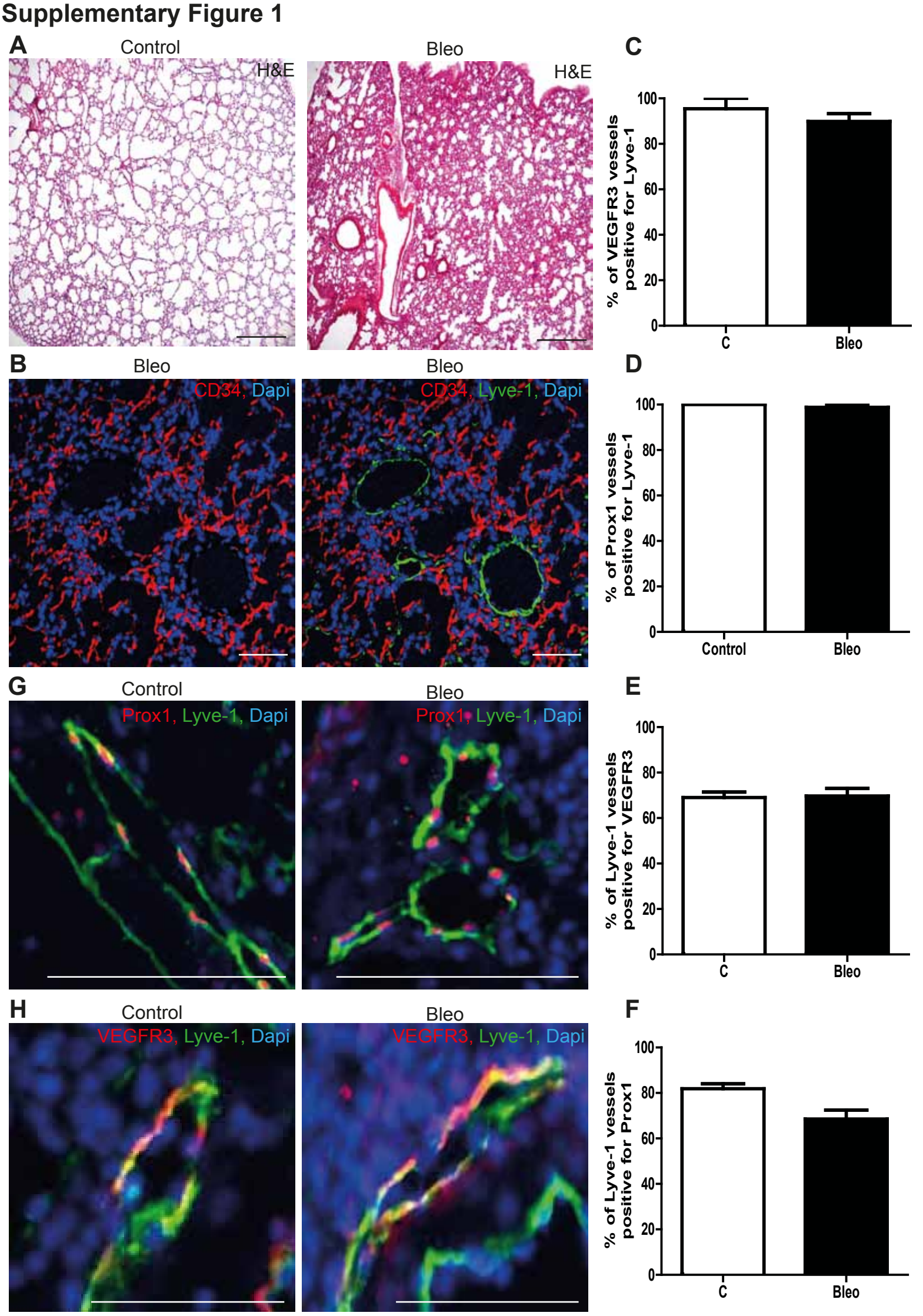
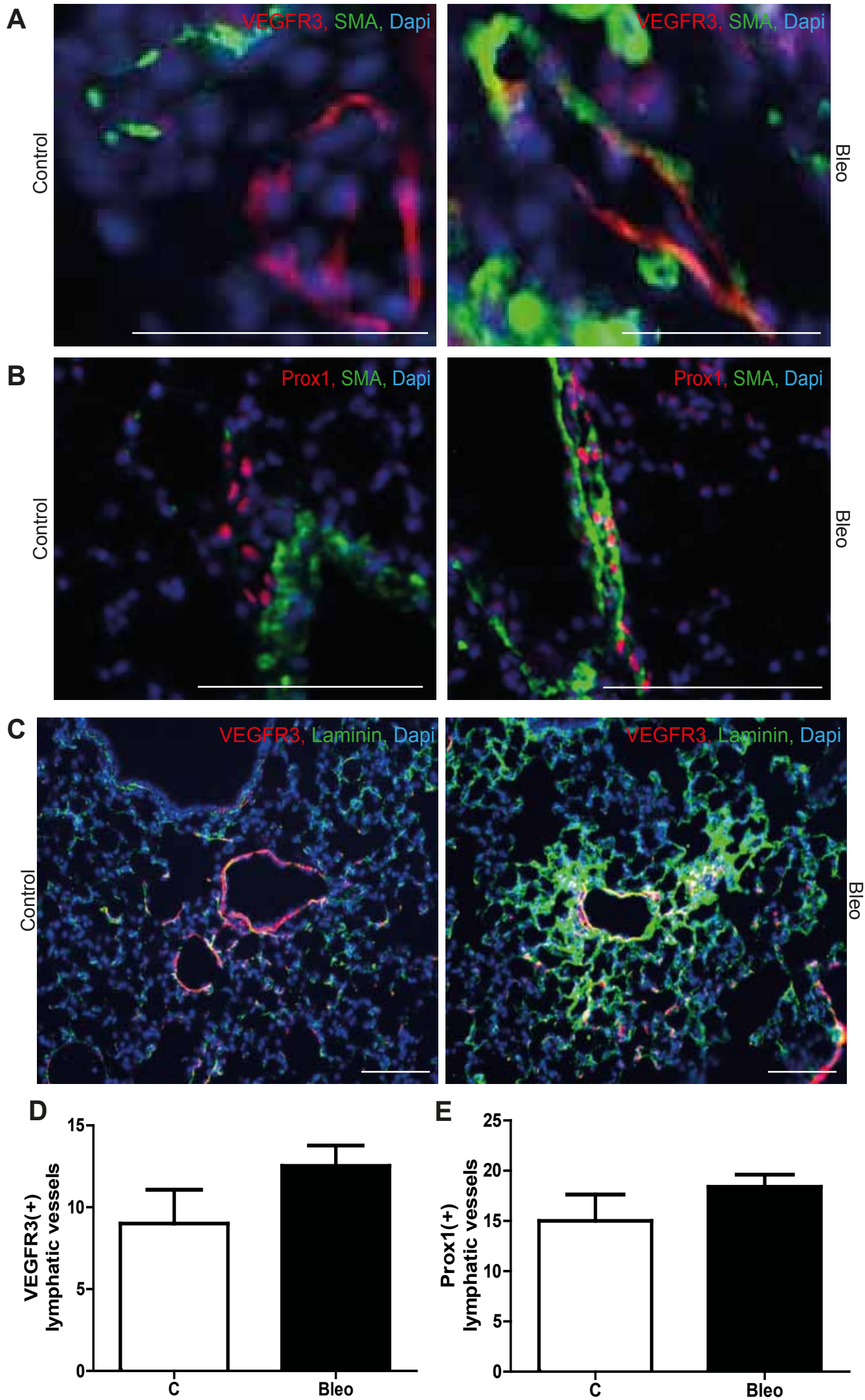
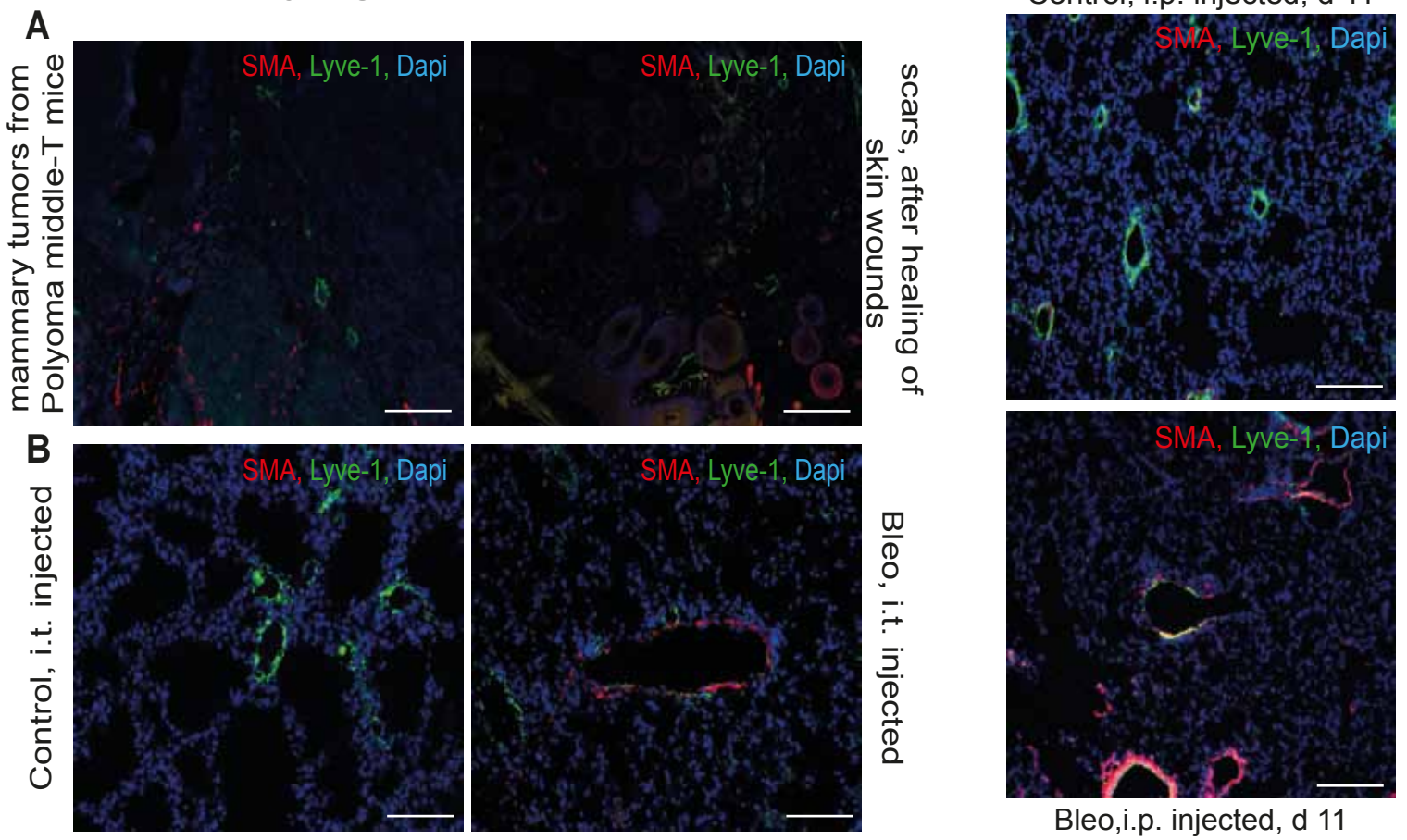
Supplementary Figure 1

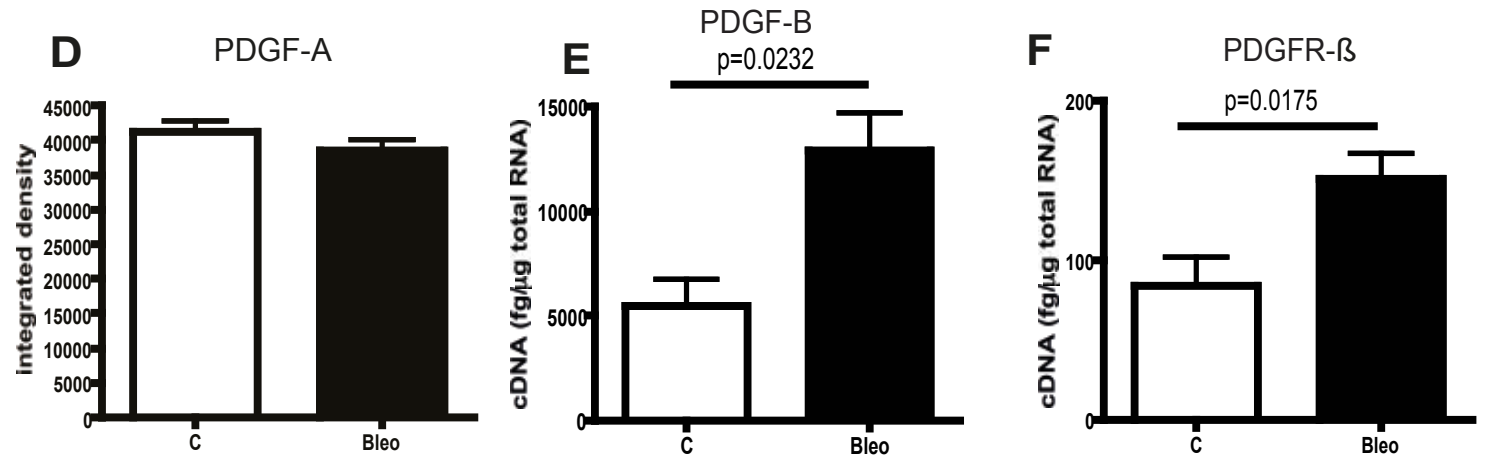


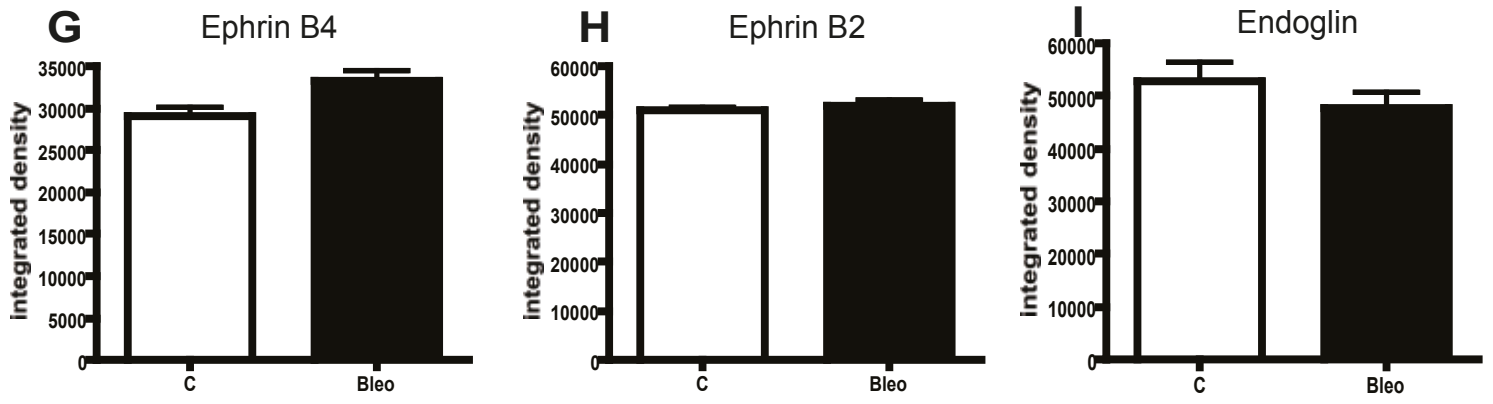
Supplementary Figure 2

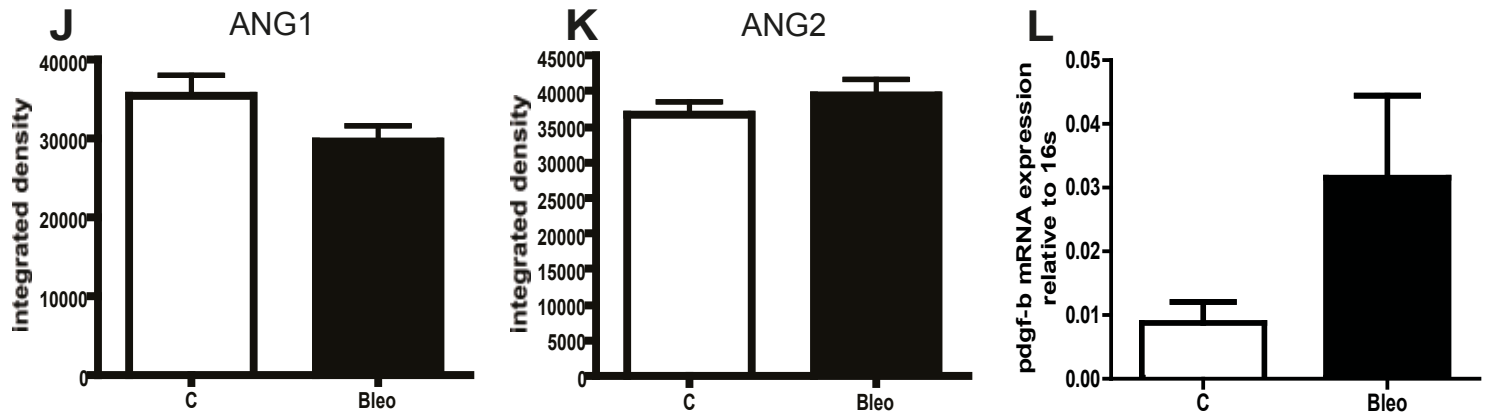


Supplementary Figure 3

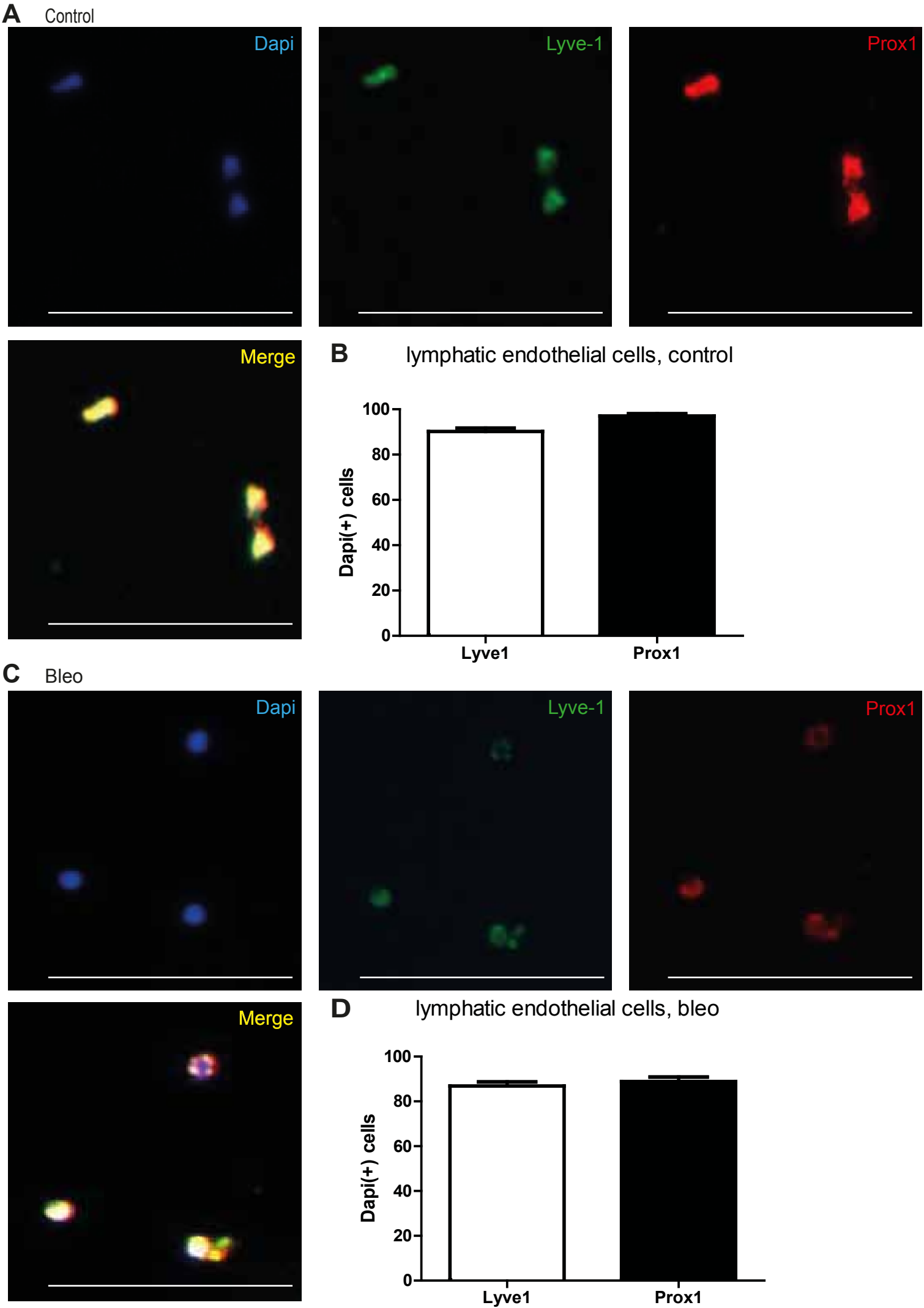




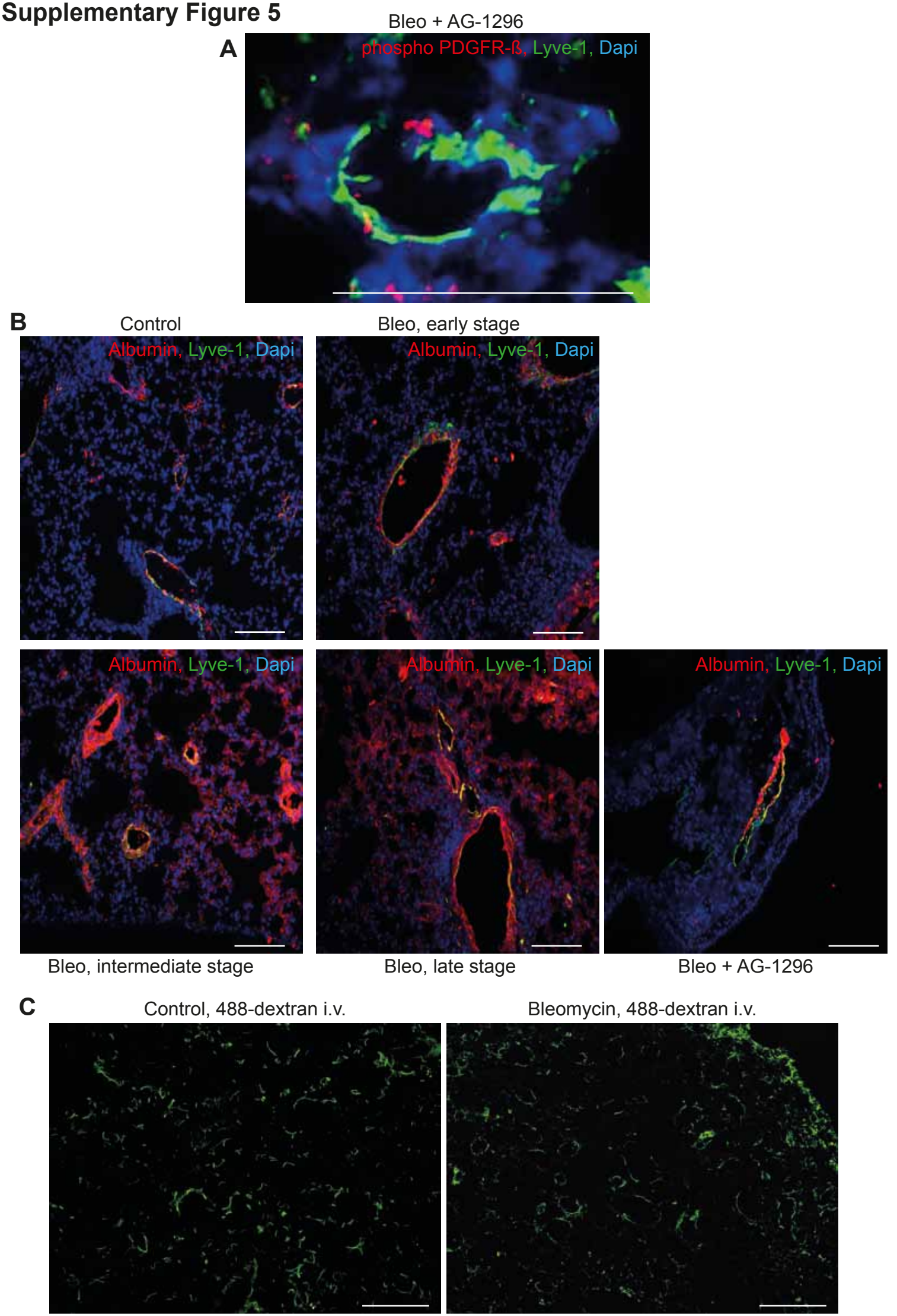




Supplementary Figure 4



Supplementary Figure 5



Supplementary Figure 1

(A) Hematoxylin and Eosin histology on lung tissue of mice treated with bleomycin (Bleo) or with PBS (Control) at day 28. Scale bars equal 500 µm.

(B) Immunodetection of blood vessels (CD34) and simultaneous detection of blood vessels (CD34) and lymphatic vessels (Lyve-1) on lungs of bleomycin treated mice at day 28. Scale bars equal 100 µm.

(C) Quantitative analysis of the amount of VEGFR3-positive lymphatic vessels which are positive for Lyve-1 on lung tissue of bleomycin- and PBS-treated mice at day 28 (controls n= 3, Bleo n=6, Bleo + AG-1296 n=6).

(D) Quantitative analysis of the amount of Prox1-positive lymphatic vessels which are positive for Lyve-1 on lung tissue of Bleo and control mice at day 28 (controls n= 3, Bleo n=6, Bleo + AG-1296 n=6).

(E) Quantitative analysis of the amount of Lyve-1-positive vessels which are positive for VEGFR3 on lung tissue of bleomycin-treated and control mice at day 28 (controls n= 3, Bleo n=6, Bleo + AG-1296 n=6).

(F) Quantitative analysis of the amount of Lyve-1-positive vessels which are positive for Prox1 on lungs of bleomycin- and PBS-treated mice at day 28 (controls n= 3, Bleo n=6, Bleo + AG-1296 n=6). Error bars show s.e.m.

(G) Double staining of lymphatic vessels with the markers Prox1 and Lyve-1 on lung tissue of healthy PBS-treated (100 µl twice a week i.p.) and fibrotic bleomycin-treated (10 mg/kg body weight twice a week i.p.) mice at day 28. Scale bars equal 100 µm.

(H) Simultaneous immunodetection of lymphatic vessels with VEGFR3 and Lyve-1 on tissue of control and fibrotic lungs at day 28. Scale bars equal 100 µm.

Supplementary Figure 2

(A) Double staining of lymphatic vessels (VEGFR3) and mural cells (SMA) on lung tissue of healthy PBS-treated and fibrotic bleomycin-treated mice at day 28. Scale bars equal 100 µm.

(B) Simultaneous immunodetection of lymphatic vessels with the marker Prox1 and mural cells (SMA) on tissue of control and fibrotic lungs at day 28. Scale bars equal 100 µm.

(C) Immunodetection of basement membrane (Laminin) and lymphatic vessels (VEGFR3) on lung sections of bleomycin-treated and PBS-treated mice at day 28. Scale bars equal 100 µm.

(D) Quantitative analysis of VEGFR3-positive lymphatic vessels of fibrotic and healthy lungs at day 28 (control n=15, Bleo n=29).

(E) Quantitative analysis of Prox1-positive lymphatic vessels of fibrotic and healthy lungs at day 28 (control n=5, Bleo n=6). Error bars show s.e.m.

Supplementary Figure 3

(A) Simultaneous detection of mural cells (SMA) and lymphatic vessels (Lyve-1) on tissue of mammary tumors from polyoma middle-T mice and on murine cutaneous wounds. Scale bars equal 100 µm.

(B) Double staining of SMA and Lyve-1 on lung tissue at day 14 after intratracheal (i.t.) injection of bleomycin or PBS, respectively. Scale bars equal 100 µm.

(C) Immunodetection of mural cells (SMA) and lymphatic vessels (Lyve-1) in fibrotic lungs of mice treated with bleomycin (Bleo) and in healthy control tissue of mice treated with PBS at day 11. Scale bars equal 100 µm.

(D-K) Gene expression and quantitative analysis of pdgf-a, pdgf-b, pdgfr-β, ephrin b4, ephrin b2, angiopoietin 1 (ANG1), angiopoietin 2 (ANG2) and endoglin in fibrotic lungs from bleomycin-treated mice and in healthy controls from mice treated with PBS at day 28 (Bleo n=10, control n=8).

(L) Quantitative analysis of the pdgfr-β expression in isolated lymphatic endothelial cells of control mice at day 28. Lymphatic endothelial cells were either cultured with bleomycin-supplemented Medium (Bleo) (10 µg/ml Medium, 12 h) or Medium without bleomycin (control) (12 h). Error bars show s.e.m.

Supplementary Figure 4:

(A) Double-staining for Lyve-1 and Prox1 on isolated lymphatic endothelial cells (LEC) of healthy control (100 µl PBS twice a week i.p.) mice at day 28 (n=6). Scale bars equal 100 µm.

(C) Quantitative analysis of Lyve-1- and Prox1-positive LECs of control mice standardized to Dapi-positive cells.

(B) Simultaneous immunodetection of Lyve-1 and Prox1 on isolated LECs of bleomycin-treated (Bleo) (10 mg/kg body weight twice a week i.p.) mice at day 28 (n=4). Scale bars equal 100 µm.

(D) Quantitative analysis of Lyve-1- and Prox1-positive LECs of bleo mice standardized to Dapi-positive cells. Error bars show s.e.m.

Supplementary Figure 5:

(A) Double immunodetection of phospho-PDGFR-β and Lyve-1 on lung tissue of mice that were treated with bleomycin+AG-1296 at day 28. Scale bars equal 100 µm.

(B) Simultaneous immunodetection of albumin and Lyve-1 on lungs of mice treated with bleomycin (Bleo), bleomycin+AG-1296 (Bleo+AG-1296) or in healthy control lungs of PBS-treated mice, respectively, showing tissue of early, intermediate and late stage fibrosis at day 28. Scale bars equal 100 µm.

(C) Fluorescence microscopy of 488-dextran (50 µl injected intravenously (i.v.) at day 28) in lung tissue of mice treated with bleomycin (Bleo) and in healthy control lungs of PBS-treated mice, respectively. Scale bars equal 100 µm.