

Supplementary material

Supplementary video legends

Supplementary Video 1: Intravital microscopy was used to visualize leukocyte adhesion to untreated mouse cremaster muscle post-capillary venules. Video is representative of experiments performed in three separate mice.

Supplementary Video 2: Intravital microscopy was used to visualize leukocyte adhesion to IL-17-injected mouse cremaster muscle post-capillary venules. 1 ug of IL-17 recombinant cytokine was administered intra-scrotally 2 h prior to intravital microscopy recording. Video is representative of experiments performed in three separate mice.

Supplementary Video 3: Intravital microscopy was used to visualize leukocyte adhesion to TNF α -injected mouse cremaster muscle post-capillary venules. 0.5 ug of TNF α -recombinant cytokine was administered intra-scrotally 2 h prior to intravital microscopy recording. Video is representative of experiments performed in three separate mice.

Supplementary Table I. Primer sequences and amplicon length in qPCR-assays

Primer name	Sequence (5'-3')	Length
IL-8	Forward- GTGCAGTTTTGCCAAGGAGT Reverse- CTCTGCACCCAGTTTTCTT	177
IL-17RA	Forward- GCCCTGCCACTCTCTCCCGA Reverse- TCATGCACTGGGCCCCTCTG	129
IL-17RC	Forward- ATGCCTGTGCCCTGGTTCTTG Reverse- CTGAACCCTGAGCCTTTCCTCG	150
VCAM-1	Forward- ATGAGATGATCTCCTTTCAGTAAGTCTATC Reverse- GGAAGCCGATCACAGTCAA	88
E-Selectin	Forward- TGAAGCTCCCACTGAGTCAA Reverse- GGTGCTAATGTCAGGAGGGAGA	77
ICAM-1	Forward- AAGATCTCGAGTGACAGTCACTGATT Reverse- CCCGAGCTCAAGTGTCTAAAGG	74
PECAM-1	Forward- CTCCAGCCAACCTTACCATC Reverse- ATGACCTCAAACCTGGGCATC	208
vWf	Forward- CGTAGCGATCTCCAATTCCAA Reverse- TCTGTGGATTTCAGTGGATGCA	84
FLAP	Forward- AGAGCACCCCTGGCTACATA Reverse- GAGATGGTGGTGGAGATCGT	144
S100A9	Forward- TCATCAACACCTTCCACCAA Reverse- TCTTTTCGCACCAGCTCTTT	88
IL-1RI	Forward- GCTGGTCAGGGGACTTTACA Reverse- TGGCATGTGGTACCTGACAT	77
IRF2	Forward-: AGTCCCATCTGGACAGCAAC Reverse- AGTCGTTTCGCTTCTGCAT	170
COL1A2	Forward- TCCAAAGGAGAGAGCGGTAA Reverse- CAGATCCAGCTTCCCCATTA	112
Casp1	Forward- GCTTTCTGCTCTTCCACACC Reverse- AAAAACAGAGCCCATTGTGG	63
GAPDH	Forward- AGCAATGCCTCCTGCACCACC Reverse- CCGGAGGGGCCATCCACAGTC	137

Supplementary Table II. Subject demographics

Groups*	Controls	Mild asthma	Severe asthma
Sex (M:F)	07:08	07:08	07:07
Age (y)	36 (19- 81)	30 (21-66)	42 (26-56)
Disease duration (y)	N/A	16 (7-21)	26(15-38)
FEV1 (L)	4 (0,3)	4 (0,2)	2 (0,2)
FEV1 % predicted	100 (3,6)	93 (3,1)	65 (5,2)
FVC (L)	4 (0,3)	5 (0,3)	3 (0,2)
FVC % predicted	102 (2,8)	98 (2,8)	79 (3,2)
FEV1/FVC (%)	81 (1,7)	82 (2,5)	70 (5,3)
Atopy (M:F)	N/A	07:08	04:04

M. Male, F. Female

* Age and disease duration presented as medians (range). All other data presented as means ± (SEM).

Supplementary figure legends

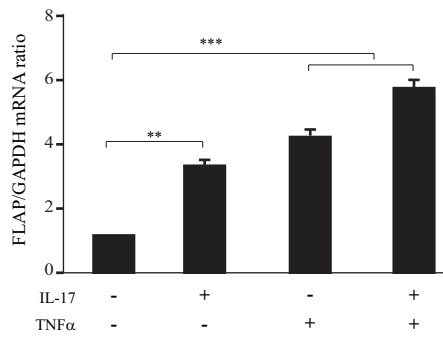
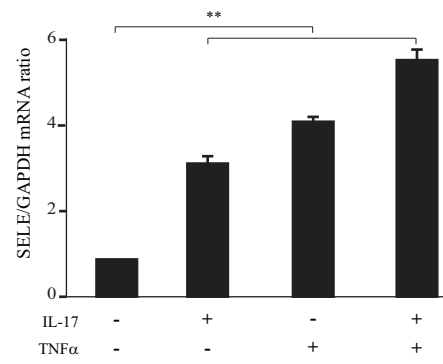
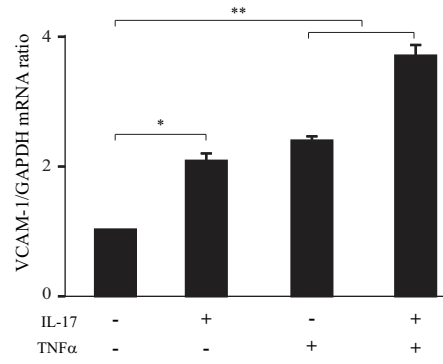
Supplementary Figure 1. IL-17 and TNF α -induction of VCAM-1, E-selectin and FLAP in LMVEC. LMVEC were left untreated (–) or exposed to 20 ng/mL IL-17 or 10 ng/ml TNF α for 2 h. After stimulation, transcript levels of VCAM-1, E-selectin (A), and FLAP were determined by qPCR.

Supplementary Figure 2. IL-17 induces p38 MAPK-dependent expression of adhesion molecules in HUVECs. HUVEC were pre-incubated for 1 h with vehicle (DMSO) (–), 0.1 μ M BIRB0796 (BIRB), 5 μ M SB 203580 (SB) as indicated. The cells were then left untreated (–) or exposed to 20 ng/mL IL-17 for 2 h. After stimulation, transcript levels of VCAM-1 and E-selectin were determined by qPCR.

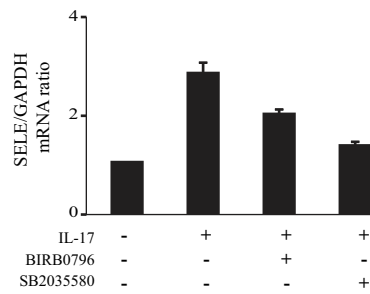
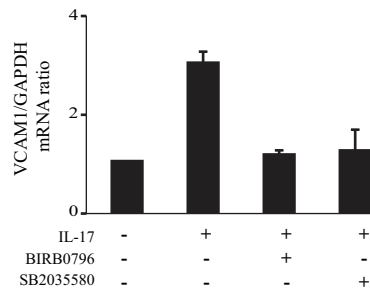
Supplementary Figure 3. IL-17 induces VCAM-1 expression at the surface of LMVEC. Flow cytometric analysis of VCAM-1 surface expression on untreated and IL-17 (20ng/ml) or IL-4 (1ng/ml) + TNF α (10ng/ml)-treated HUVEC for 12h. The HUVEC population (86%) was gated in the dot plot using cell size (FSC) vs. cell granularity (SSC) (left graph). Histograms show isotype control, secondary antibody only and not stained cells (NS) (center graph) whereas untreated or treated-HUVEC are displayed right.

Supplementary Figure 4. Transcripts up-regulated in IL-17-stimulated LMVEC, whose expression is dependent on the p38 MAPK pathway. LMVEC were pre-incubated for 1 h with DMSO as vehicle (–), 0.1 μ M BIRB0796 (BIRB), 5 μ M SB 203580 (SB) as indicated. The cells were then left untreated (–) or exposed to 20 ng/mL IL-17 for 2 h. After stimulation, the transcript levels of S100A9 (**A**), IL-1R1 (**B**), IRF2 (**C**), COL1A2 (**D**) and CASP1 (**E**) were determined by qPCR.

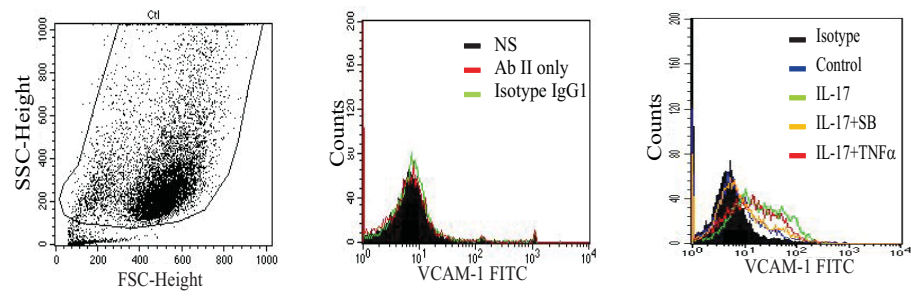
Supplementary Fig 1



Supplementary Figure 2



Supplementary Fig 3



Supplementary Fig 4

