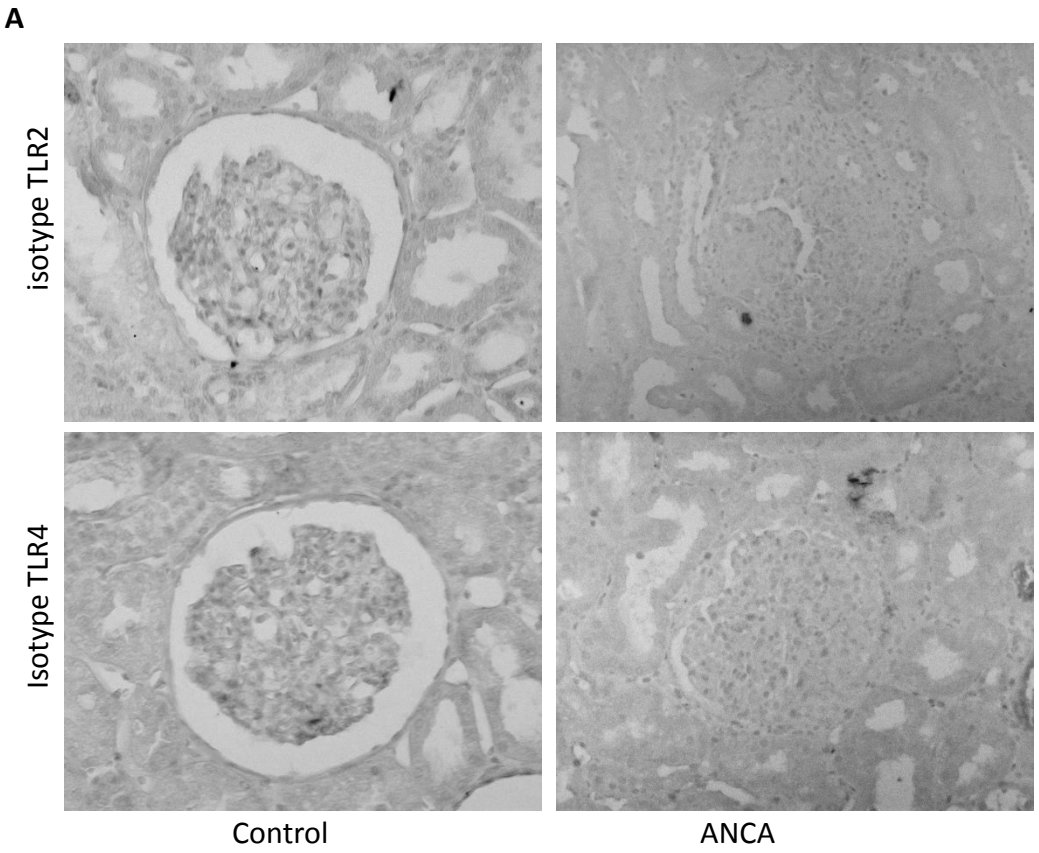


Figure S1
Isotype staining were negative in both control and ANCA kidney sections

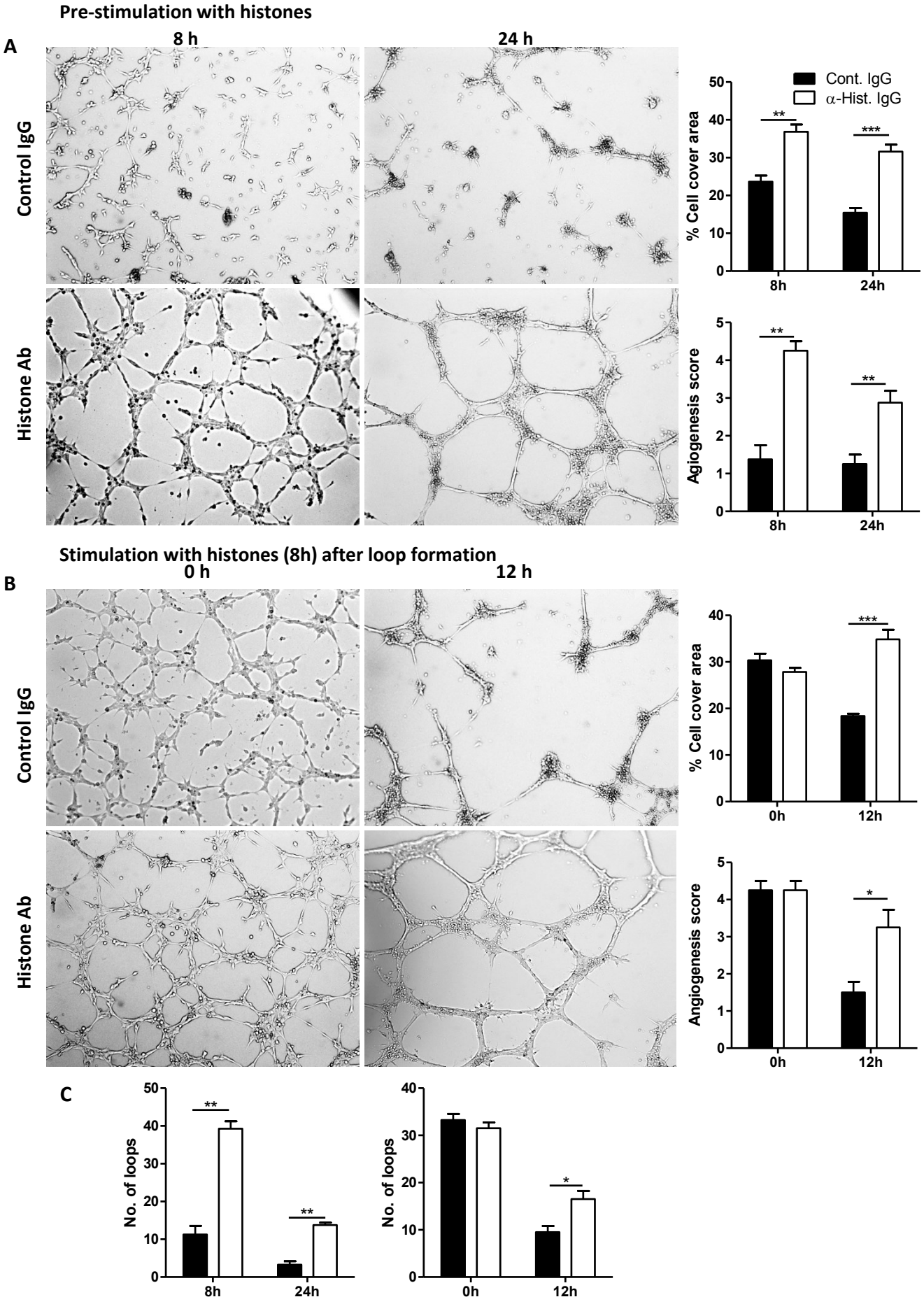


Figure S2

Histones and endothelial cell microtubes in vitro. A. Murine glomerular endothelial cells were seeded into a matrigel matrix for angiogenesis experiments as described in methods. Histones +/- anti-histone IgG were added either during (A) or 8h after (B) microtube formation to test histone toxicity on microtube forming (A) or destruction (B), respectively. Quantitative assessment included the number of living cells and network formation. Data represent the mean of these endpoints ± SEM of three independent experiments at the indicated time points. * p<0.05, ** p<0.01, ***p<0.001 versus control IgG.

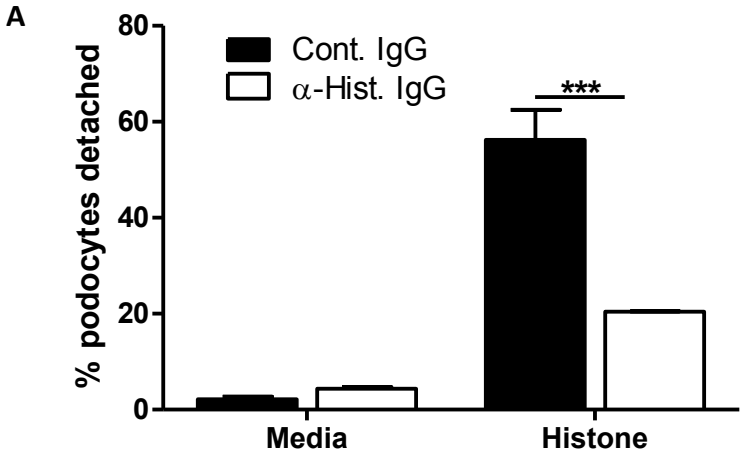


Figure S3
Anti-histone IgG and podocyte detachment in vitro. Murine podocytes were exposed to histones with or without anti-histone IgG. Data show the mean percentage ± SEM of podocytes that detached from the culture dish within 24 hours. * $p<0.05$, *** $p<0.001$ versus control.

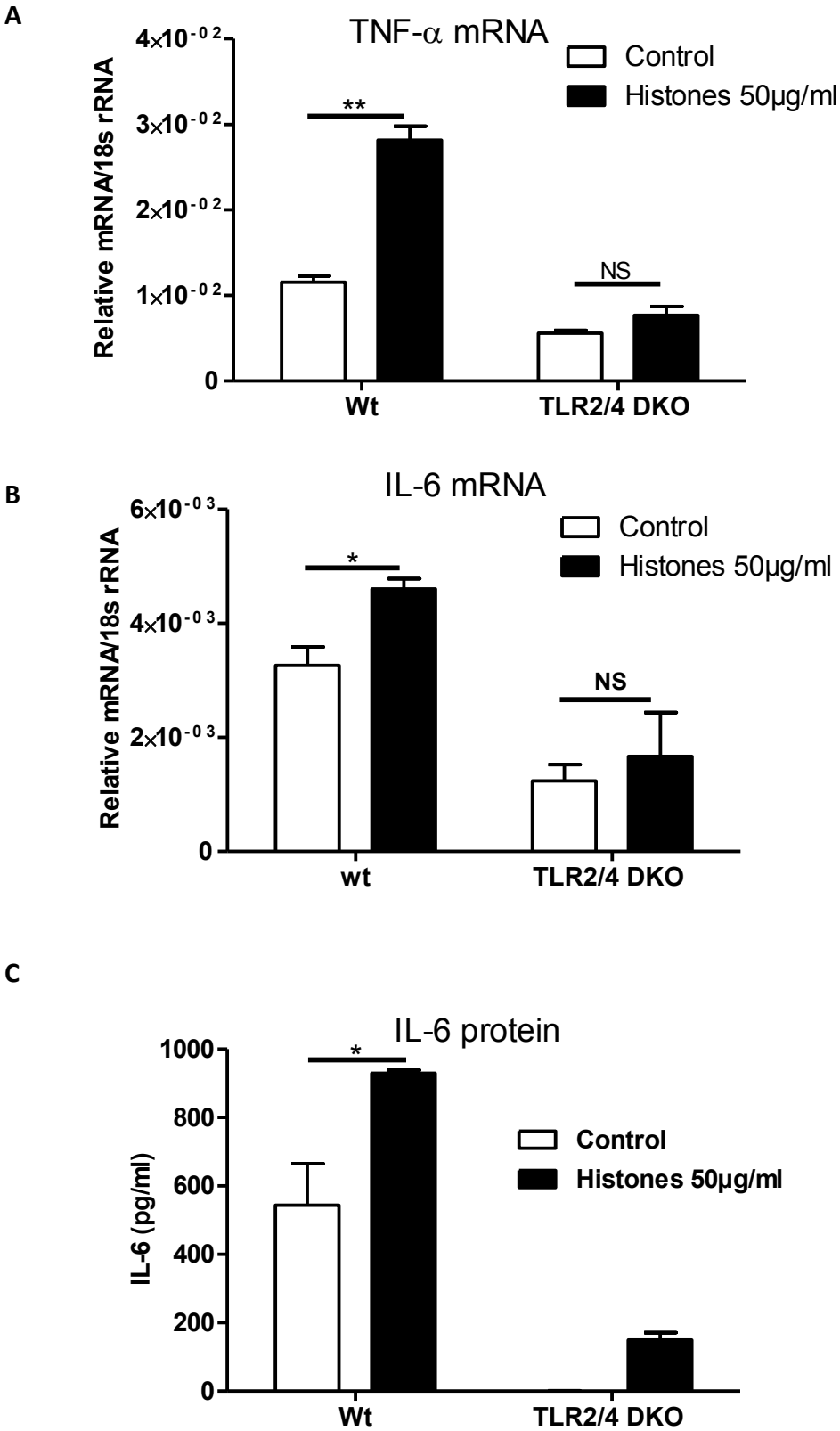
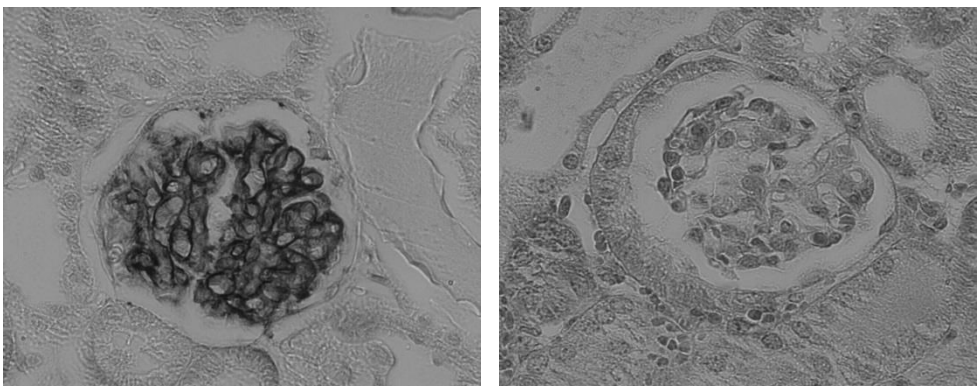


Figure S4

Cytokine induction in glomeruli ex vivo exposed to histones. Glomeruli were isolated from wild type mice and *Tlr2-/4* double knockout mice and exposed to histones. 12 hours later the mRNA levels of TNF-α (A) and IL-6 (B) were determined by real-time RT-PCR. Data show the mRNA expression level corrected for the housekeeper 18srRNA ± SEM each done in triplicate. IL-6 protein release was also determined in cell culture supernatants (C). * p<0.05, **p<0.01, n.s. = not significant.

A



Sheep IgG Mouse IgG

B

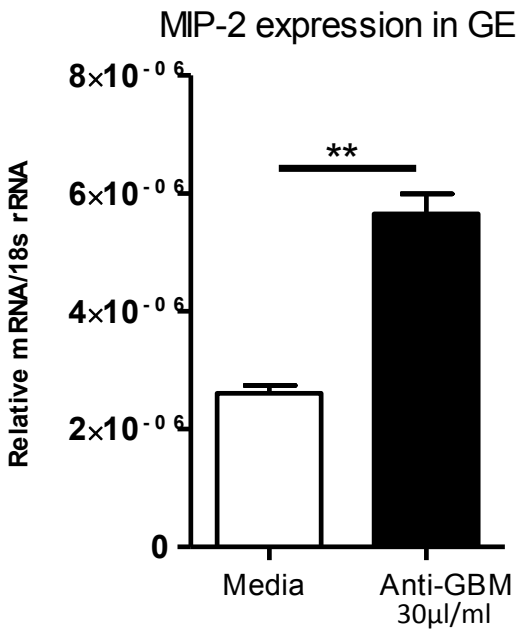
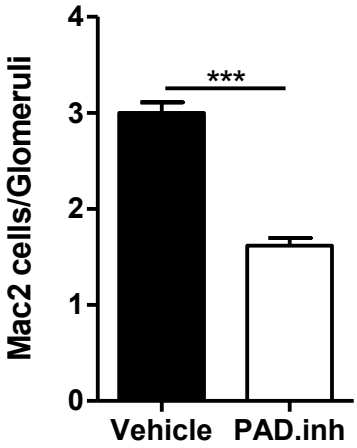


Figure S5

MIP2/CXCXL2 mRNA expression in glomerular endothelial cells (GEnC). Exposure to glomerular basement membrane (GBM) antiserum induces the mRNA expression levels of MIP2/CXCL2. Data are means ± SEM from three independent experiments.

A



B

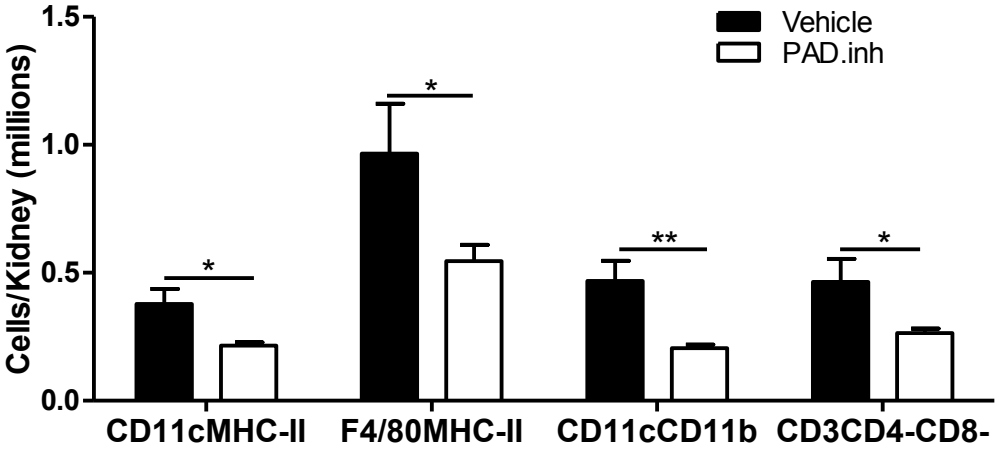


Figure S6 . (A): The model of antiserum-induced GN using PAD inhibitor showed reduced number of Macrophages per glomeruli. (B): immune cells FACS in kidney suspension. Data represent mean ± SEM from five to six mice of each group. * $p<0.05$, ** $p<0.01$, *** $p<0.001$ versus vehicle.

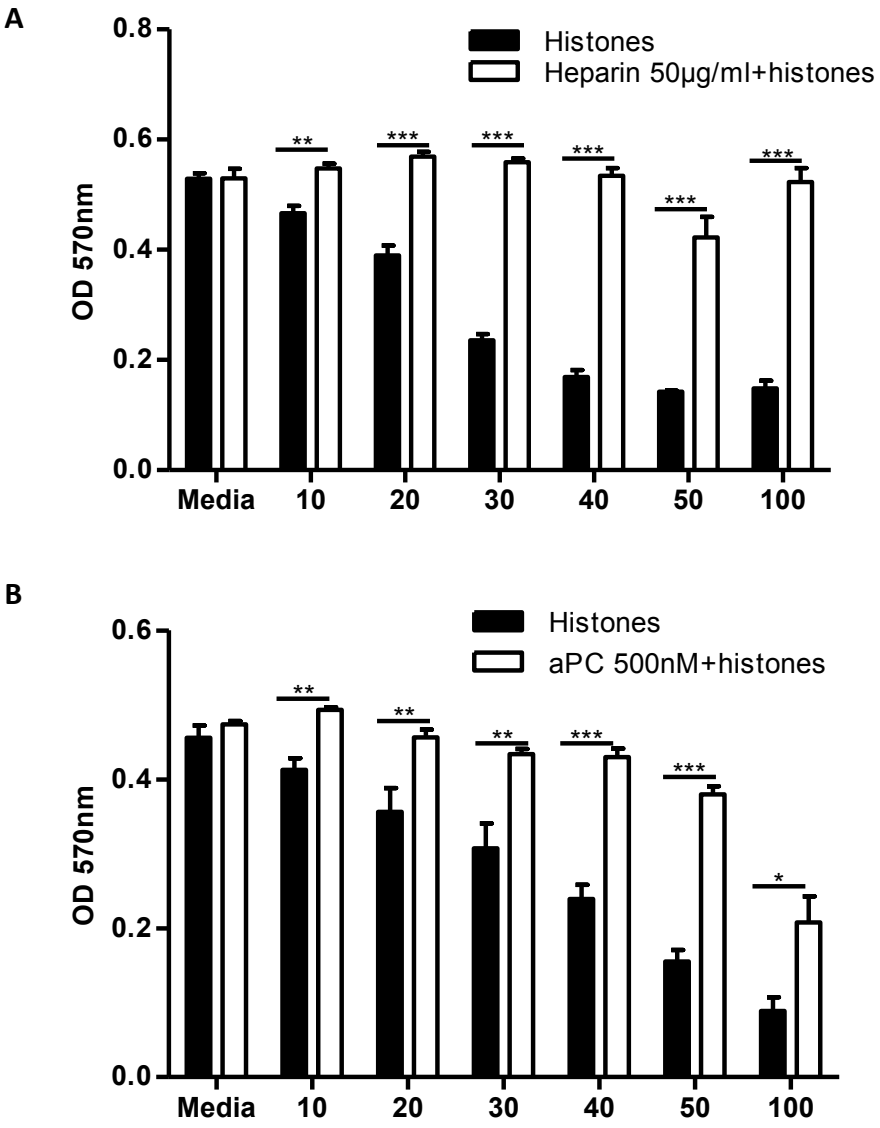


Figure S7

Heparin and activated protein C (aPC) block histone toxicity on glomerular endothelial cells. Glomerular endothelial cells were exposed to increasing doses of histones with or without heparin (A) or aPC (B) as indicated. Data represent mean OD ± SEM of three MTT assay experiments measured at a wavelength of 492 nm.

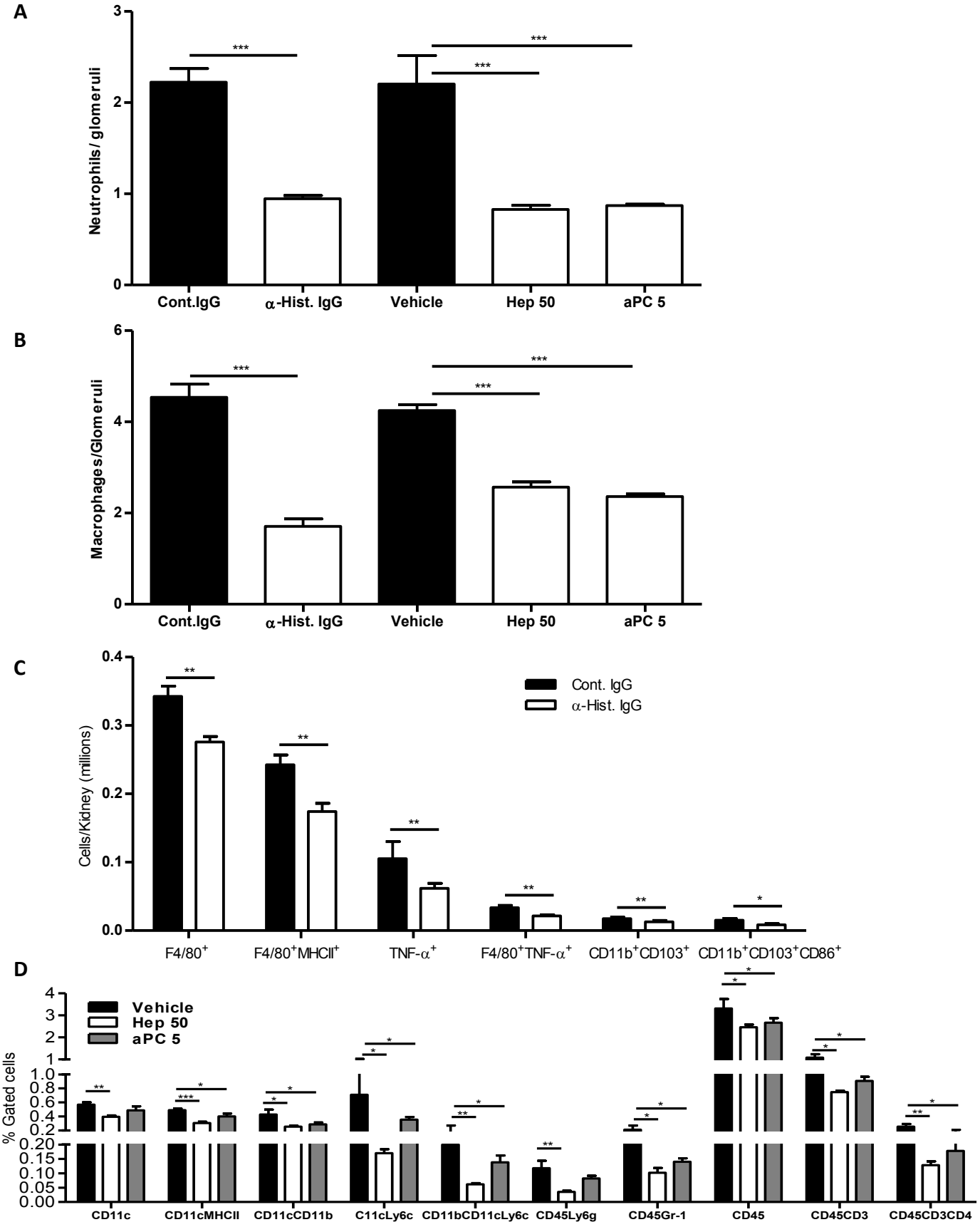


Figure S8

Therapeutic histone blockade and renal leukocytes. Renal sections were obtained at day 7 of the experiment and stained for GR-1 (neutrophils, A) and Mac2 (macrophages, B). Data represent mean glomerular cell counts ± SEM of 5-6 mice in each group. C and D: Flow cytometry data for various leukocyte subsets as indicated. * p<0.05, ** p<0.01, ***p<0.001 versus control IgG or vehicle, respectively.

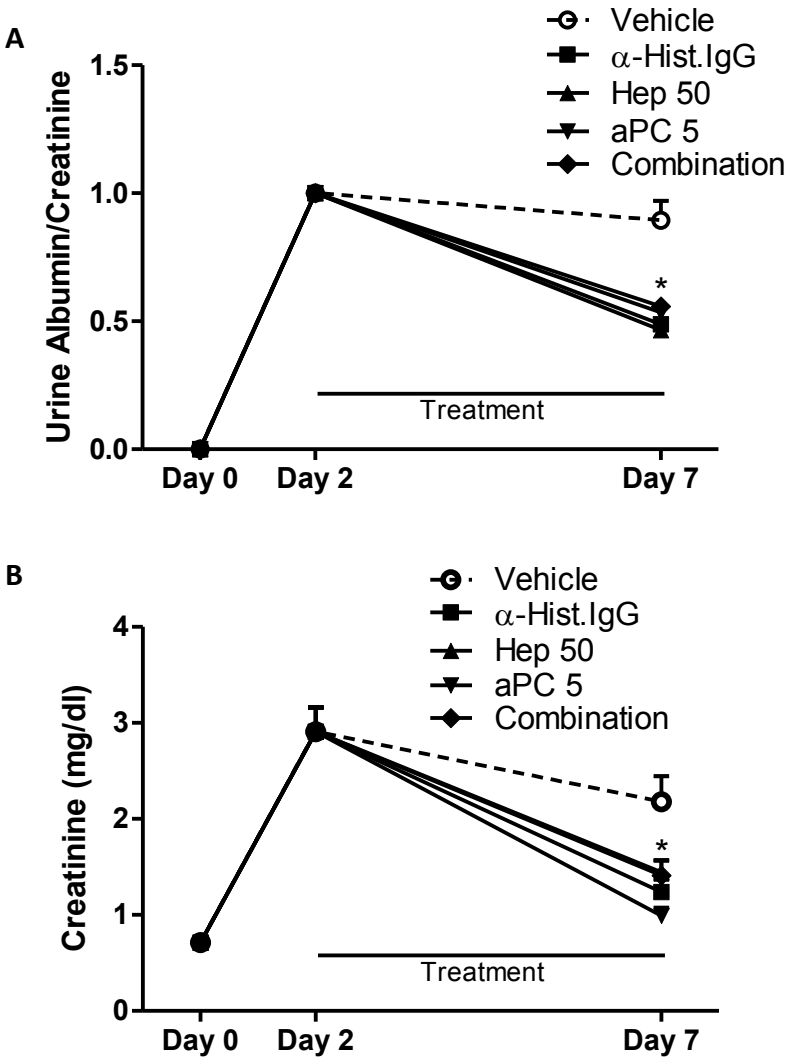


Figure S9. (A): Effect of different treatments and combination of anti-histone IgG, heparin and aPC on proteinurea on day 7 of anti-GBM induced glomerulonephritis and (B) creatinine levels, combination treatment did not show any additive effect