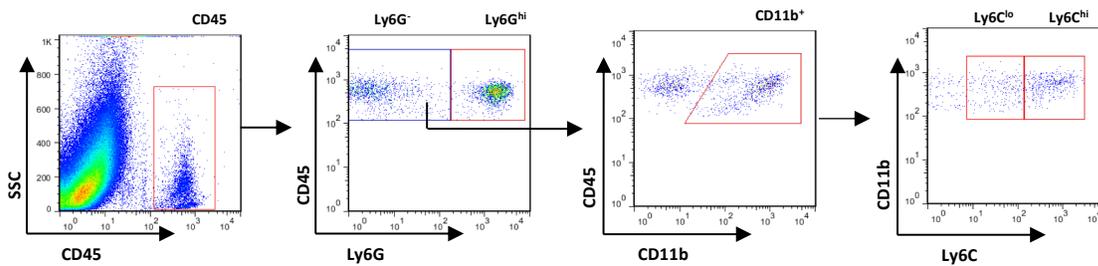
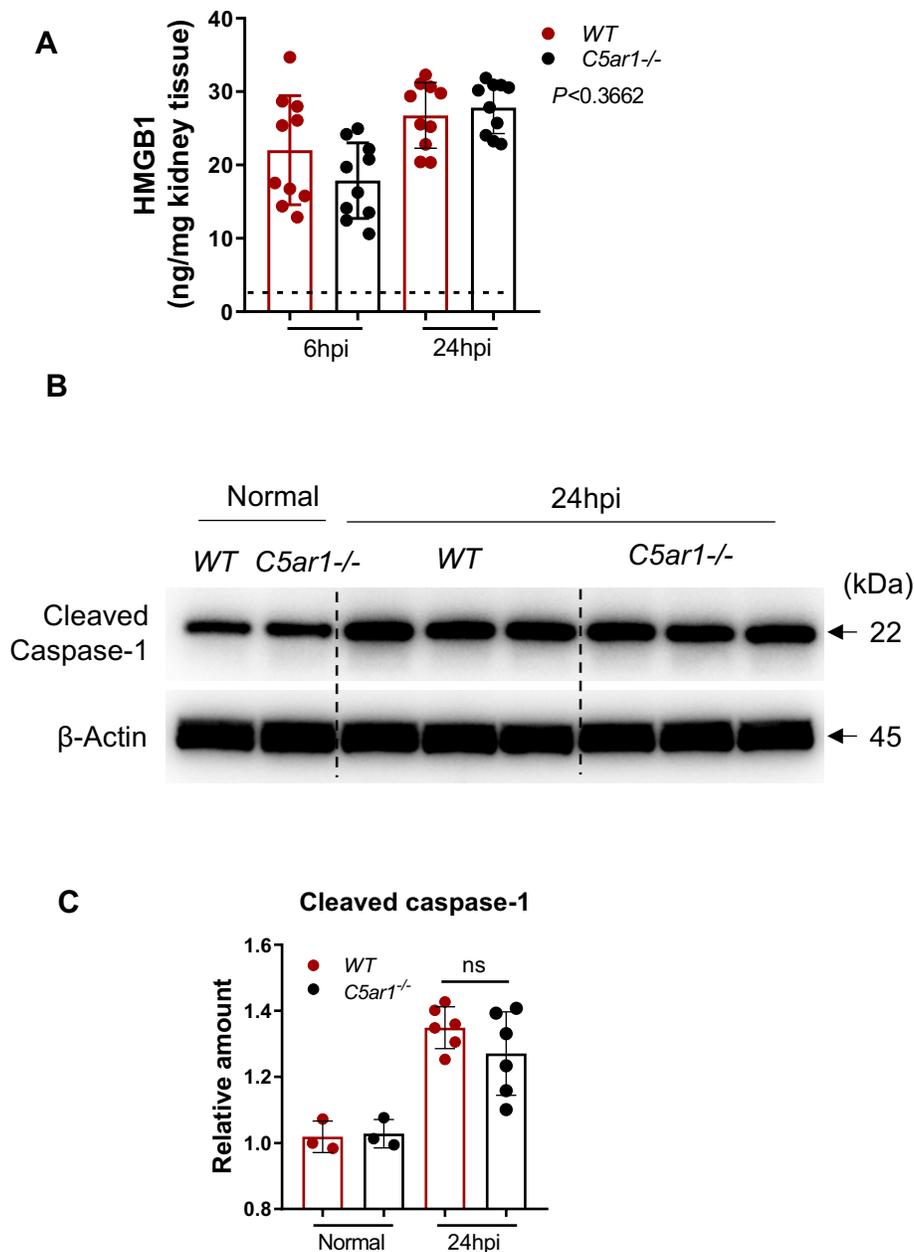


**sFigure 1. Flow cytometry gating strategy**



Renal inflammatory cell infiltration was analysed in infected WT and *C5ar2<sup>-/-</sup>* mice at 24hpi by flow cytometry. Stepwise gating strategy used in flow cytometric analysis of leukocytes (CD45<sup>+</sup>), neutrophil (CD45<sup>+</sup>Ly6G<sup>+</sup>), monocyte/macrophage (Ly6G<sup>-</sup>CD11b<sup>+</sup>) and Ly6C<sup>hi</sup> and Ly6C<sup>lo</sup> monocyte/macrophage in kidney tissues.

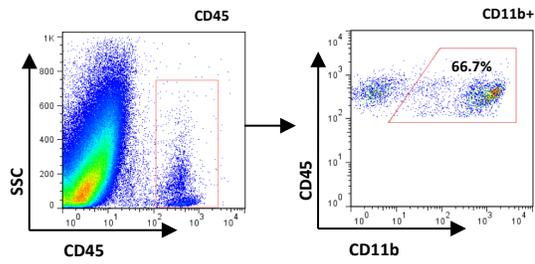
**sFigure 2. Intrarenal HMGB1 and cleaved caspase-1 in *WT* and *C5ar1*<sup>-/-</sup> mice following inoculation with UPEC**



(A) Intrarenal protein levels of HMGB1 in *WT* and *C5ar1*<sup>-/-</sup> mice at 6hpi and 24hpi, determined by ELISA. Data were analyzed by Two-way ANOVA with multiple comparisons test (n=10 mice/group). (B) Representative Western blots showing cleaved caspase-1 and  $\beta$ -actin in kidney tissue lysates from *WT* and *C5ar1*<sup>-/-</sup> mice (i.e. uninfected [normal] and infected [24hpi]). (C) Relative amount of cleaved caspase-1, corresponding to the groups of mice in B, quantified as described in Materials and methods. Data were analyzed by Unpaired 2-tailed Student's t test (n= 3-6 mice/group, pooled from two experiments). Error bars represent standard deviation. ns, no significant difference.

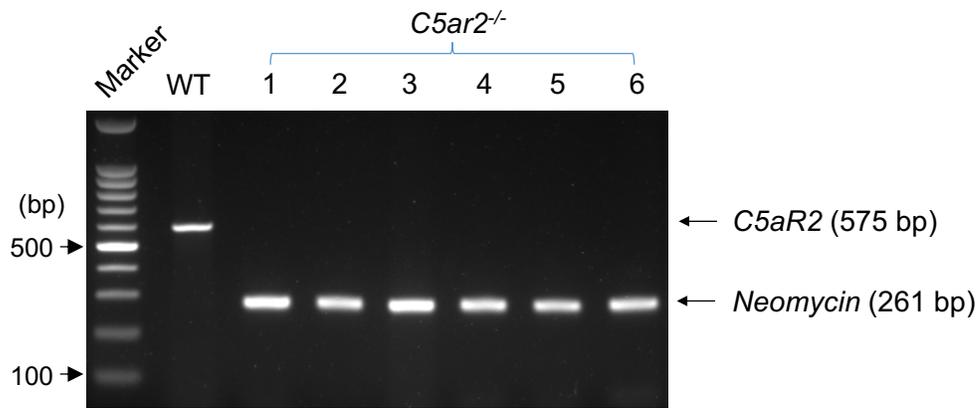


**sFigure 4. Most renal infiltrating leukocytes are CD11b<sup>+</sup> cell**



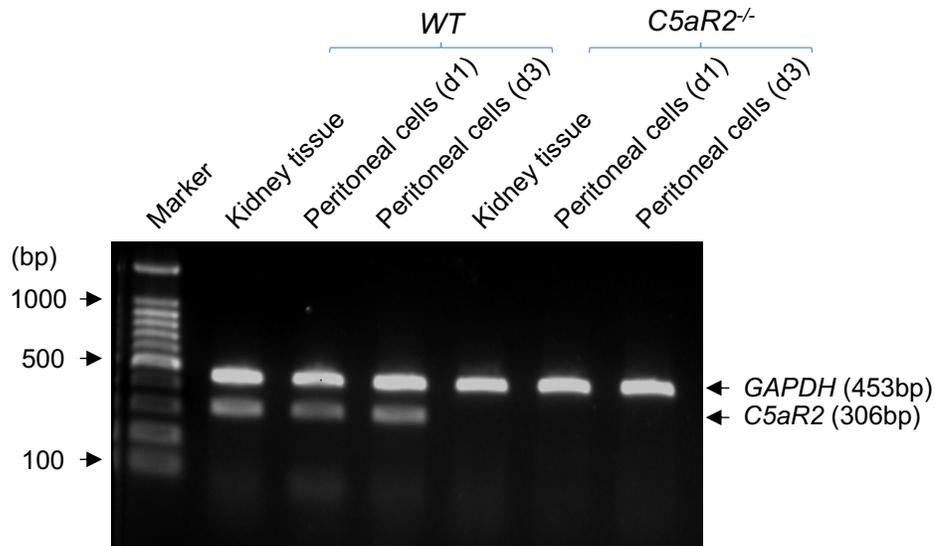
Renal inflammatory cell infiltration was analysed in infected WT mice at 24hpi. Flow cytometry analysis revealed that the majority (>60%) of renal infiltrating leukocytes were CD11b<sup>+</sup> cells following the inoculation in this model.

**sFigure 5. Genotyping of *C5ar2*<sup>-/-</sup> mice**



PCR analysis of tail genomic DNA from WT and *C5ar2*<sup>-/-</sup> mice (n=6 from 2 breeding pairs). PCR was performed using two pairs of primers for *C5aR2* and *neomycin*. The agarose gel shows negative detection of *C5aR2* and positive detection of neomycin in *C5ar2*<sup>-/-</sup> mice, whereas positive detection of *C5aR2* and negative detection of neomycin in WT mice, thus confirming *C5aR2* gene knockout in *C5ar2*<sup>-/-</sup> mice. Marker: 100 bp DNA ladder.

**sFigure 6. Absence of *C5aR2* expression in kidney tissue and peritoneal cells of *C5aR2*<sup>-/-</sup> mice**



RT-PCR analysis of *C5aR2* mRNA in normal kidney tissue and peritoneal cells of WT and *C5aR2*<sup>-/-</sup> mice. Peritoneal cells were prepared from peritoneal exudate cells d1 or d3 after (i.p) injection of 1mL of Thioglycolate. The agarose gel shows positive detection of *C5aR2* expression in kidney tissue and peritoneal cells of WT mice but negative detection in *C5aR2*<sup>-/-</sup> mice. GAPDH, used as an internal control in RT-PCR. Marker: 100 bp DNA ladder. PCR primer sequences are given in Materials and methods.