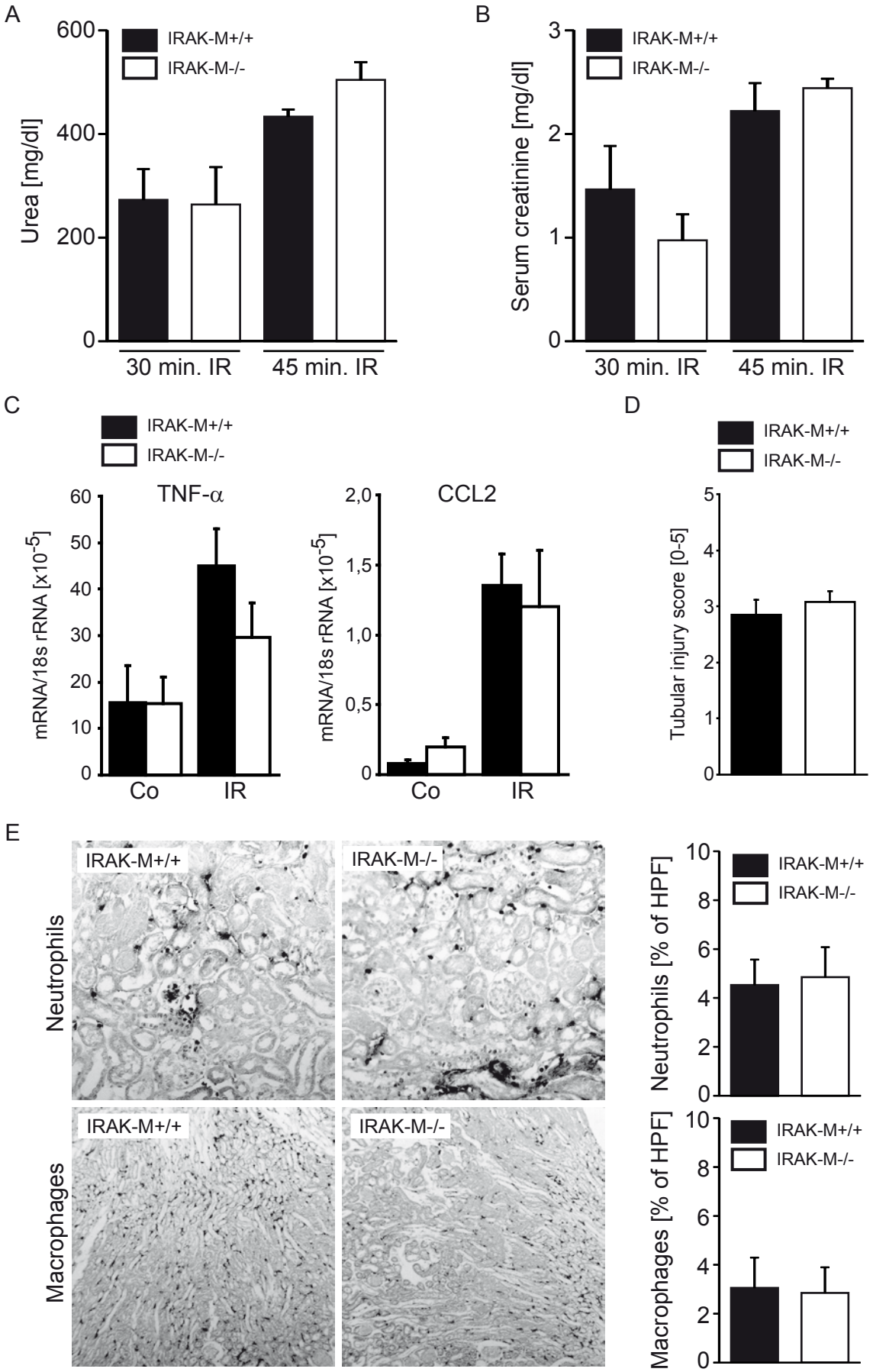
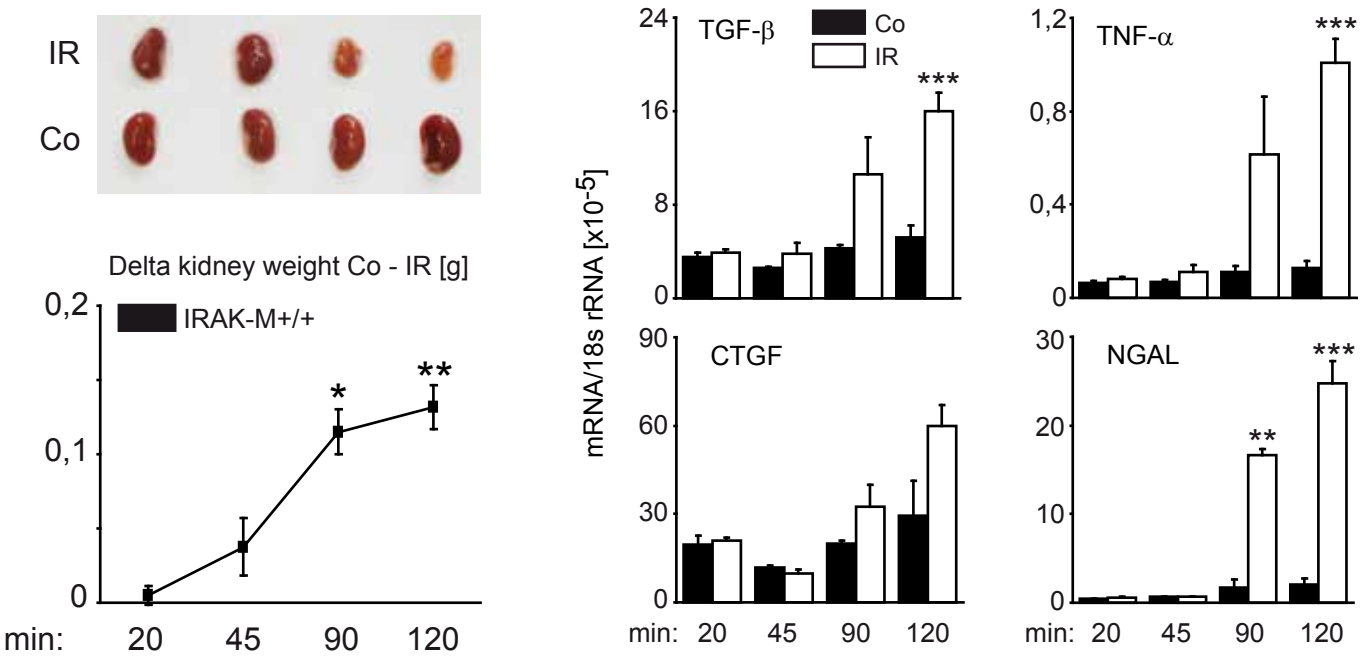
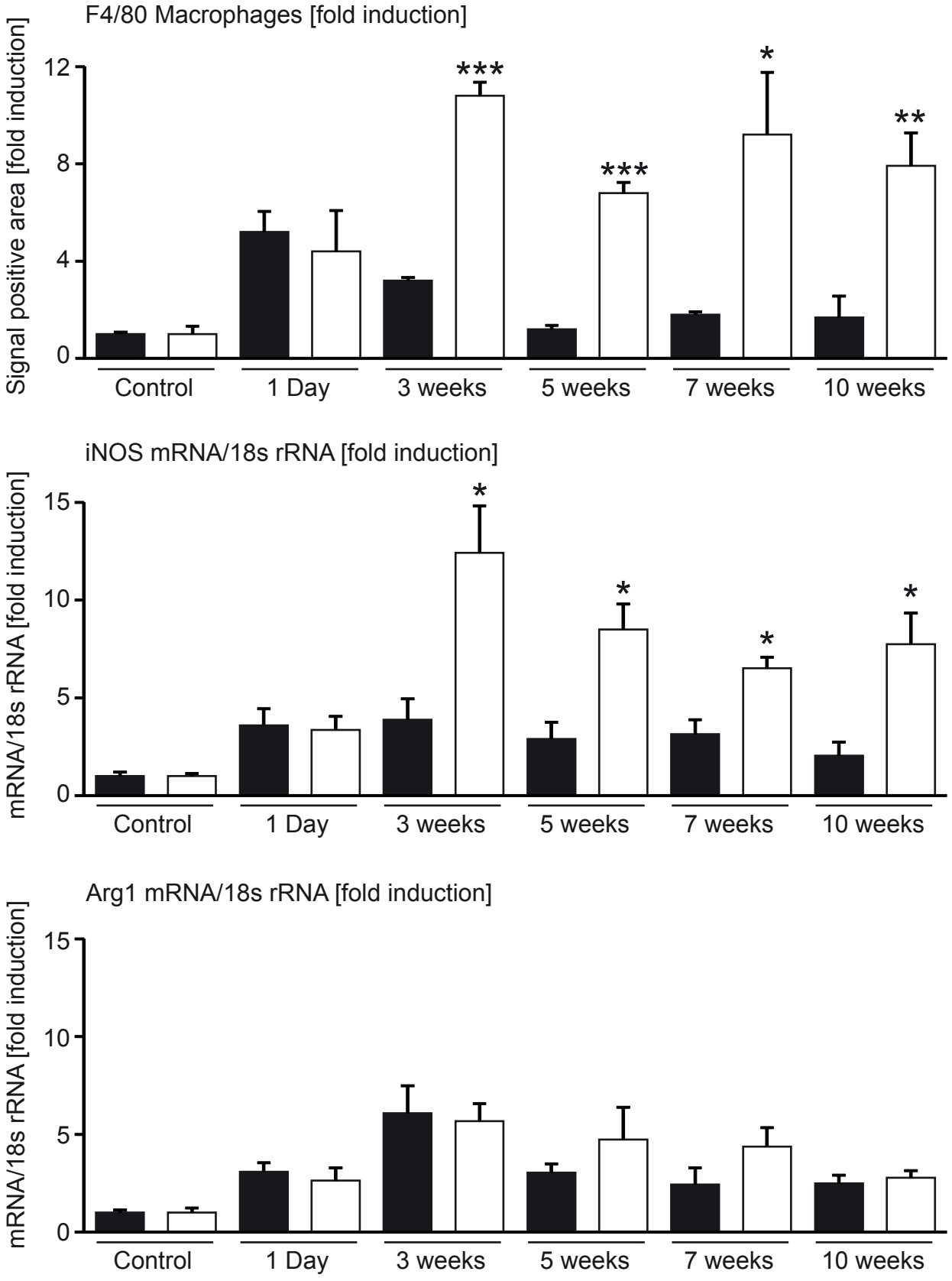


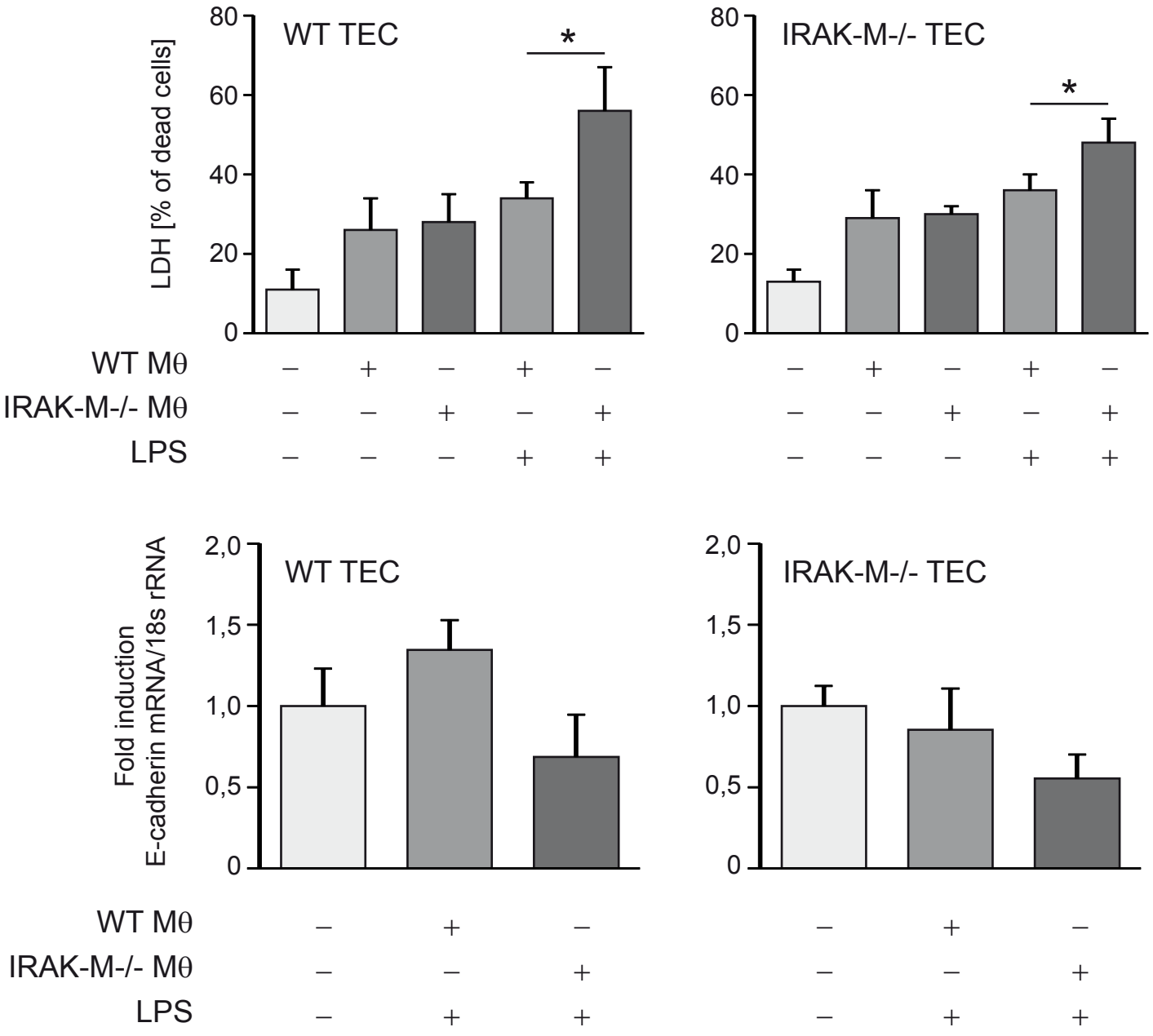
Supplementary figure 1. IRAK-M in acute kidney injury. Wild-type (n=6) and IRAK-M-deficient mice (n=6) underwent bilateral IR for 30 or 45 minutes. Data represent mean serum urea (A) and creatinine (B) levels ± SEM of six mice in each group 24 hours after injury. C: Renal tissue was obtained at 1 day after unilateral renal pedicle clamping by IRAK-M or wild type mice; mRNA expression levels were determined by real-time RT-PCR. Data are expressed as mean of the ratio versus the respective 18s rRNA level ± SEM. D: From the same experiment ischemic kidney section were stained for PAS (not shown). The tubular injury score was determined in post-ischemic (IR) and contralateral kidneys (Co) as described in methods. Data are means ± SEM. E: Neutrophils and macrophages (original magnification x100) were quantified by digital morphometry and presented as percentage of hpf. Data are means ± SEM.



Supplementary figure 2. Ischemia time determines AKI outcomes. Ischemic kidneys from wild type mice (n= 5-10) were obtained at 5 weeks after IRI. Kidney weight loss of ischemic kidney (IR) was c ompared with contralateral kidney (Co) after various times of renal pedicle clamping (20, 45, 90 or 120 minutes). Data on the left represent mean ratio of the delta kidney weight (Control minus ischemic kidney) ± SEM from at least five mice; * $p<0.05$, ** $p<0.01$ versus 20 min ischemia time. Total RNA was extracted from IR kidneys (white bars) or contralateral (co) kidneys (black bars) of wild-type mice at various length of ischemia time as indicated. mRNA expression levels were determined for the indicated targets by real-time RT-PCR. Data are expressed as mean of the ratio versus the respective 18s rRNA level ± SEM; * $p<0.05$, $p<0.01$, *** $p<0.001$ versus control kidneys of the same ischemia time.

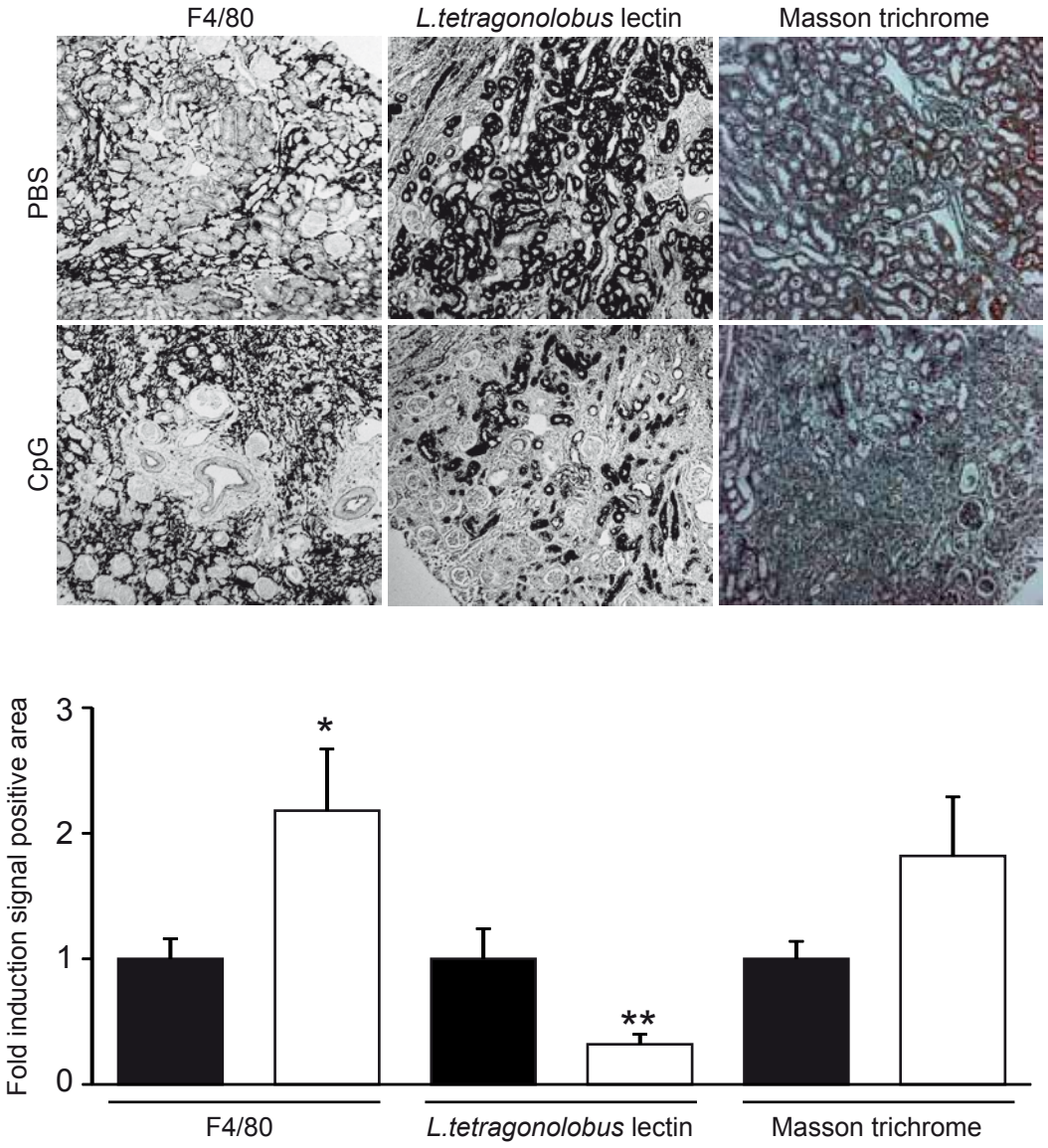


Supplementary figure 3. Lack of IRAK-M and macrophage numbers and phenotype in post-ischemic kidneys. Ischemic kidneys from wild type mice (n= 5-10) were obtained at 1 day or 3, 5, 7, and 10 weeks after IRI. Kidneys were stained for F4/80+ renal mononuclear phagocytes and quantified by original magnification of x50. The quantitative analysis were performed using image software and are expressed as means ± SEM of positivity per high power field; * $p<0.05$, $p<0.01$, *** $p<0.001$ versus wild-type mice of the same time point. Data are presented as fold induction compared to control kidney. Total RNA was extracted from wild type kidneys (black bars) or IRAK-M-deficient kidneys (white bars) at various time after ischemia injury as indicated. mRNA expression levels were determined for the indicated targets by real-time RT-PCR. Data are expressed as mean of the ratio versus the respective 18s rRNA level ± SEM; * $p<0.05$, $p<0.01$, *** $p<0.001$ versus control kidneys of the same ischemia time.



Supplementary figure 4. In vitro studies with renal macrophages and tubular epithelial cells. Renal tubular epithelial cells were prepared from IRAK-M+/+ and IRAK-M-/- mice and cultured with CD11b+ myeloid cells prepared from IRAK-M+/+ and IRAK-M-/- untreated or prestimulated with LPS as indicated. Cell culture supernatants were harvested and LDH assay was performed. Data are expressed as mean ± SEM; * $p<0.05$ versus wild type TECs. The expression levels of E-cadherin were determined by real-time RT-PCR. Data are expressed as mean of the ratio versus the respective 18s rRNA level ± SEM.

A



Supplementary figure 5. Treatment of post-ischemic AKI with CpG-DNA. A: Wild type mice underwent unilateral renal pedicle clamping as described in methods. Groups of mice (n=8) received vehicle control or 40µg CpG-DNA i.p. given on every alternate day starting 5 days after reperfusion. A: Renal tissue was obtained at 5 weeks after unilateral renal pedicle clamping and stained for renal macrophages, tubular cells (with lotus tetragonolobus lectin) and Masson trichrome for fibrosis. Representative images are shown for both genotypes and each time point at an original magnification of x50. The quantitative analysis were performed using image software as described in methods and are expressed as fold induction means ± SEM of positivity per high power field; ** $p<0.01$ versus wild-type mice of the same time point.