

SUPPLEMENTARY DATA

Supplementary figure 1. Kidney Klotho expression is decreased in AKI induced by cisplatin (CDDP). Mice were injected with a single dose of CDDP or vehicle (control) and killed at 72 h. Total kidney Klotho mRNA levels were measured by qRT-PCR. Mean \pm SEM of 5 animals per group. * p <0.05 vs control.

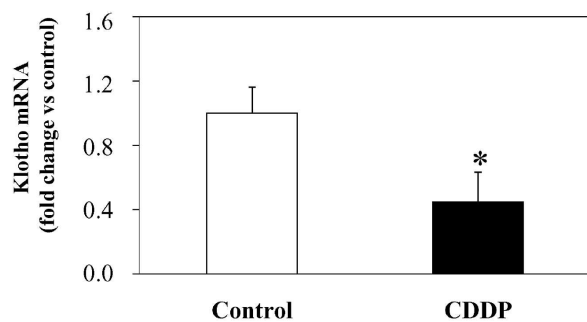
Supplementary figure 2. TWEAK absence improves renal function and reduces tubular injury during AKI. AKI was induced by a folic acid overdose in healthy WT or TWEAK KO mice. **A, B**) Renal function was assessed at 72 h measuring plasma creatinine (**A**) and BUN (**B**). Mean (\pm SEM) of 5 animals per group. * p <0.005 vs healthy WT mice # p <0.04 vs AKI WT mice. **C**) Histological tubular injury was assessed by an AKI score which had a maximum value of 21. Mean \pm SEM of 5 animals per group. * p <0.005 vs healthy WT mice # p <0.03 vs AKI WT mice.

Supplementary figure 3. I κ B α down-regulation by siRNA reduces Klotho expression in tubular cells. **A**) I κ B α protein level is efficiently down-regulated by a specific siRNA. Tubular cells were transfected with a siRNA oligonucleotide targeting I κ B α or scrambled control and I κ B α expression was assessed 48 hours after transfection by Western blot. **B**) Klotho mRNA expression in tubular cells transfected 48 h earlier were quantified by qRT-PCR. I κ B α binds RelA and keeps it inactive in the cytoplasm. I κ B α siRNA targeting, a maneuver facilitating RelA nuclear translocation and NF κ B activation, reduces Klotho mRNA in unstimulated cells. Mean \pm SD of three independent experiments. * p <0.02 vs scrambled control.

Supplementary figure 4. The HDAC inhibitor valproic acid prevents TWEAK- and TNF α -induced downregulation of Klotho mRNA in cultured tubular cells. **A**) The histone deacetylase inhibitor valproic acid (VPA) rescues TWEAK or TNF α -induced repression of Klotho at 3h in MCT cells. Cells were pre-stimulated with VPA (3 mM) 1 hour prior to cytokines. Mean \pm SD of three independent experiments. * p <0.005 vs control. # p <0.05 vs cytokine alone.

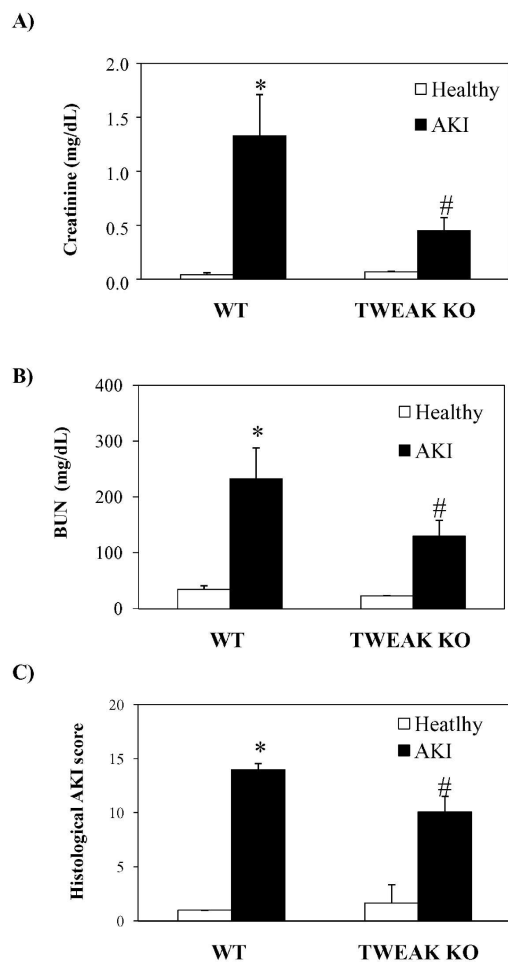
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Supplementary Figure 1



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Supplementary Figure 2

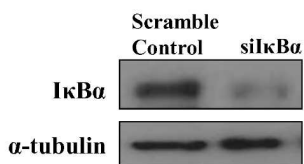


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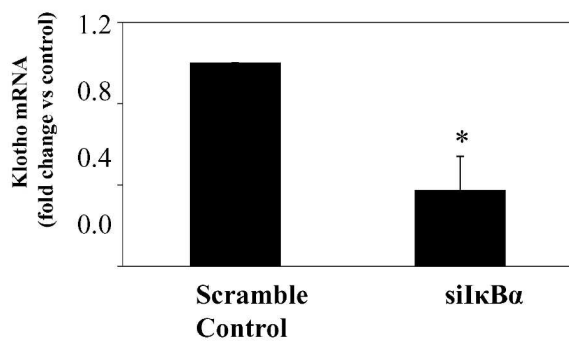
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Supplementary Figure 3

A)

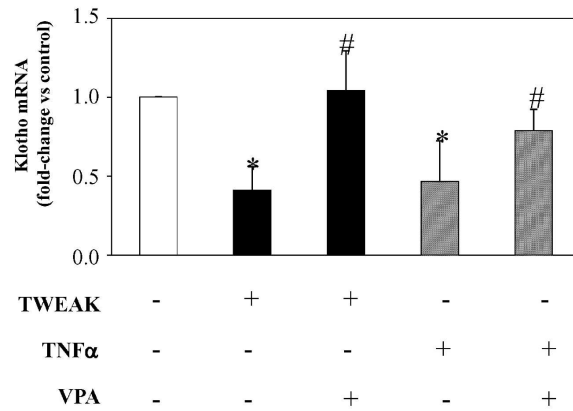


B)



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Supplementary Figure 4



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Table S1. Primers for ChIP (chromatin immunoprecipitation) real-time RT- PCR.

NAME	SEQUENCE
Region 1s	CCCTTGATCTCCCCAAATTC
Region 1a	AGCAGGGCTCCTAGATCACA
Region 2s	CCCTTGATCTCCCCAAATTC
Region 2a	AGCAGGGCTCCTAGATCACA