Supplemental Material

Supplemental Figure Legends

Supplemental Figure 1. The renal function and expression of CCKBR in Ang II-treated WT mice.

WT ($CCKBR^{+/+}$) mice were subcutaneously infused, via osmotic minipump, with saline vehicle (control, 2.64 μL/d) or Ang II (1.44 mg/kg body weight/d) for 28 d. The kidney samples were collected at the end of 28 d.

A and B: Serum creatinine (A) and urinary albumin (B) were quantified. n=6; $^{*}p<0.05$ vs. Vehicle treatment.

C:Representative images of H&E staining and quantitative analysis of renal tubular injury, n=6. $^{*}p<0.05$ vs. Vehicle treatment.

D: Representative images of Masson's trichrome staining and quantitative analysis of renal interstitial fibrosis. n=6. $^{*}p<0.05$ vs. Vehicle treatment.

E and F: The mRNA (E) and protein (F) expressions of CCKBR were quantified in the kidneys from mice after 28 d of saline vehicle (control, 2.64 μL/d) or Ang II (1.44 mg/kg body weight/d) infusion, n=6. $^{*}p<0.05$ vs. Vehicle-treatment.

G: Immunofluorescence analysis of CCKBR (red) and DAPI (blue) in the kidneys of mice after 28 d of saline vehicle (control, 2.64 μL/d) or Ang II (1.44 mg/kg body weight/d) infusion. Representative images are on the left and bar graphs are on the right, n=6. $^{*}p<0.05$ vs. Vehicle treatment.

Supplemental Figure 2. The effects of CCKBR deficiency on urinary albumin, systolic blood pressure, and number of glomeruli with Ang II treatment.

WT ($CCKBR^{+/+}$) and $CCKBR^{-/-}$ mice were subcutaneouslyinfused, via osmotic minipump, with saline vehicle (control, 2.64 μL/d) or Ang II (1.44 mg/kg body weight/d) for 28 d.

A: Representative images of urinary albumin bands stained by Coomassie brilliant blue with BSA as the standard (1, 2, 10 μg) and quantitative analysis of urinary albumin, n=6. $^{*}p<0.05$ vs. WT+Vehicle treatment, $^{\#}p<0.05$ vs. WT+Ang II treatment.

B: Systolic blood pressure (SBP) of WT and $CCKBR^{-/-}$ mice infused with Ang II (1.44 mg/kg body weight/d) at 0, 3, 7,14, 21, 28 d of infusion, n=6. $^{*}p<0.05$ vs. WT+Ang II treatment.

C: H&E staining of the kidneys of WT and $CCKBR^{-/-}$ mice infused with Ang II (performed 10×); the glomeruli are marked with arrows. The bar graphs show the quantitative analysis of number of glomeruli per area (10×, 20 serial sections of the entire kidney), n=3. p=not significant.

Supplemental Figure 3. The effects of CCKBR deficiency with Ang II+Gastrin-treatment.

A: Representative images of H&E staining and quantitative analysis of renal tubular injury, n=6. $^{*}p<0.05$ vs. WT+Ang II+Gastrin treatment.

B: Representative images of Masson's trichrome staining and quantitative analysis of renal interstitial fibrosis, n=6. $^{*}p<0.05$ vs. WT+Ang II+Gastrin treatment.

C: Representative images of Sirius Red staining and quantitative analysis of Sirius Red-positive areas showing the renal collagen deposition, n=6. $^{*}p<0.05$ vs. WT+Ang II+Gastrin treatment.

Supplemental Figure 4. The effect of gastrin on the systolic blood pressure in mice infused with Ang II and mice with UUO.

A: Systolic blood pressure (SBP) was measured by tail-cuff method, in WT ($CCKBR^{+/+}$) mice subcutaneously infused, via osmotic minipump, with saline vehicle (control, 2.64 μL/d), Ang II (1.44 mg/kg body weight/d), and/or gastrin (120 μg/kg body weight/d) at 0, 3, 7, 14, 21, 28 d of infusion, n=6. $^{*}p<0.05$ vs. others, $^{\#}p<0.05$ vs. Ang II treatment.

B: Schematic diagram of the experimental design. The mice with UUO were subcutaneously infused, via osmotic minipump with saline vehicle (control,2.64 μL/d) or gastrin (120 μg/kg body weight/d) for 7 d. The kidneys were harvested 7 d after gastrin infusion.

C: Systolic blood pressure (SBP, measured by tail-cuff method) in UUO mice infused with saline vehicle control or gastrin, n=6. p=not significant.

Supplemental Figure 5. The gating strategy for *in vivo* efferocytosis assay and the effect of gastrin on efferocytosis-associated genes *in vitro*.

A: These figures show flow cytometry electronic gating strategy used to identify peritoneal macrophages that had engulfed BCECF AM-labeled apoptotic cells *in vivo*.

A1 shows the cells with normal morphology. A2 shows CD11b-positive-cells. A3 shows CD11b-positive cells with normal morphology. A4 shows F4/80-positive cells in CD11b-positive cells; these cells were considered as peritoneal macrophages. A5 shows FITC-positive cells in peritoneal macrophages. Because the apoptotic Jurkat cells were FITC-positive, the FITC-positive cells in peritoneal macrophages were considered as the macrophages that engulfed the apoptotic Jurkat cells.

B: RAW264.7 cells were incubated with gastrin (10^{-7} mol/L) or saline vehicle (control, 25μL) for 24 h, after which time the mRNA expression of efferocytosis-associated genes was measured, n=5. $^{*}p<0.05$ vs. Vehicle treatment.

Supplemental Figure 6. The effect of gastrin treatment on the mRNA expression of PPAR-α and the effect of sh-PPAR-α on efferocytosis.

A: RPT cells were incubated with gastrin (10^{-7} mol/L) or saline vehicle (control, 25 μL) for 24 h, after which time PPAR-α mRNA was quantified by qRT-PCR, n=5. $^{*}p<0.05$ vs. Vehicle treatment.

B: The mRNA expression of PPAR-α was quantified in the kidneys from different groups of mice; all mice were WT ($CCKBR^{+/+}$), unless indicated as $CCKBR^{-/-}$. $^{*}p<0.05$ vs. Vehicle treatment, $^{\#}p<0.05$ vs. WT+Ang II treatment (WT=red open bar vs $CCKBR^{-/-}$=blue open bar), $^{\&}p<0.05$ vs. UUO treatment.

C:The flow cytometry analysis shows the percentage of $CD11b^{+}$ (FITC) F4/80^{+} (PE) macrophages (kidney-resident macrophages) in isolated kidney cells.

D: Kidney-resident macrophages were incubated with saline vehicle (control, 25 μL),

CI988 (10^{-7} mol/L), gastrin (10^{-7} mol/L), or gastrin (10^{-7} mol/L)+CI988 (10^{-7} mol/L) for 24 h, prior to measuring PPAR-α mRNA, n=5. $^{*}p<0.05$ vs. others, $^{\#}p<0.05$ vs. Gastrin treatment.

EandF: RAW264.7(E) and RPT (F)cells were infected with lenti-shPPAR-α or control lentiviral particles (scramble shRNA transfected with lentiviral particles) for 72 h, after which time PPAR-α mRNA was quantified by qRT-PCR. The knockout efficiency was at least 70%, n=5. $^{*}p<0.05$ vs. Vehicle treatment.

G:Representative fluorescent images showing engulfment of apoptotic Jurkat cells (BCECF AM) by kidney-resident macrophages (Dil) (left panel). Kidney-resident macrophages were infected with lentivirus expressing PPAR-α shRNA or control (scramble) shRNA, and then treated with saline vehicle (control, 20 μL) or gastrin (10^{-7} mol/L) for 24 h prior to measuring efferocytosis (phagocytosis) (right graph), n=4. $^{*}p<0.05$ vs. Vehicle treatment; $^{\#}p<0.05$ vs. Gastrin treatment.

Supplemental Figure 7. Original Western Blot images.

A: Refer to Figure 4C.

B: Refer to Figure 4D.

C: Refer to Figure 4E.

D: Refer to Figure 4F.

E: Refer to Figure 4G.

F: Refer to Figure 4H.

G: Refer to Figure 5C.

H: Refer to Figure 5E.

I: Refer to Figure 6G.

J and K: Refer to Figure 7C.