Supplemental figure legends

Supplementary Figure S1- **Evaluation of rubicon antibodies**. Samples from siControl and sirubicon were used. All antibodies detected bands suggested by the manufacturers, and the density of these detected bands was markedly decreased after rubicon knock down, confirming the specificity of the antibodies and existence of different rubicon variants.

Supplementary Figure S2- **Evaluation of rubicon and beclin-1 antibody for immunofluorescent staining.** After transient transfection beclin1 siRNA for 48 hours, beclin-1 expression was significantly decreased as shown in Westernblot and immunofluorescent staining.

Supplementary Figure S3- **Selective EGFR deletion in podocytes had no effect on body weight , fasting glucose and HbA1c in *db/db* mice**. *Egfr^f/f^*; *db/db* (*db/db*) mice and nphs2-Cre; *egfr^f/f^*; *db/db* (*egfr*^podKO^; *db/db*) had similar body weights, fasting blood glucose and HbA1c levels at 8 weeks old and 40 weeks old (8 weeks: n = 11 for *db/db* group and n = 12 for *egfr*^podKO^; *db/db* group; 40 weeks: n = 10 for *db/db* group and n = 12 for *egfr*^podKO^; *db/db* group). Two-tailed unpaired student's t -test.
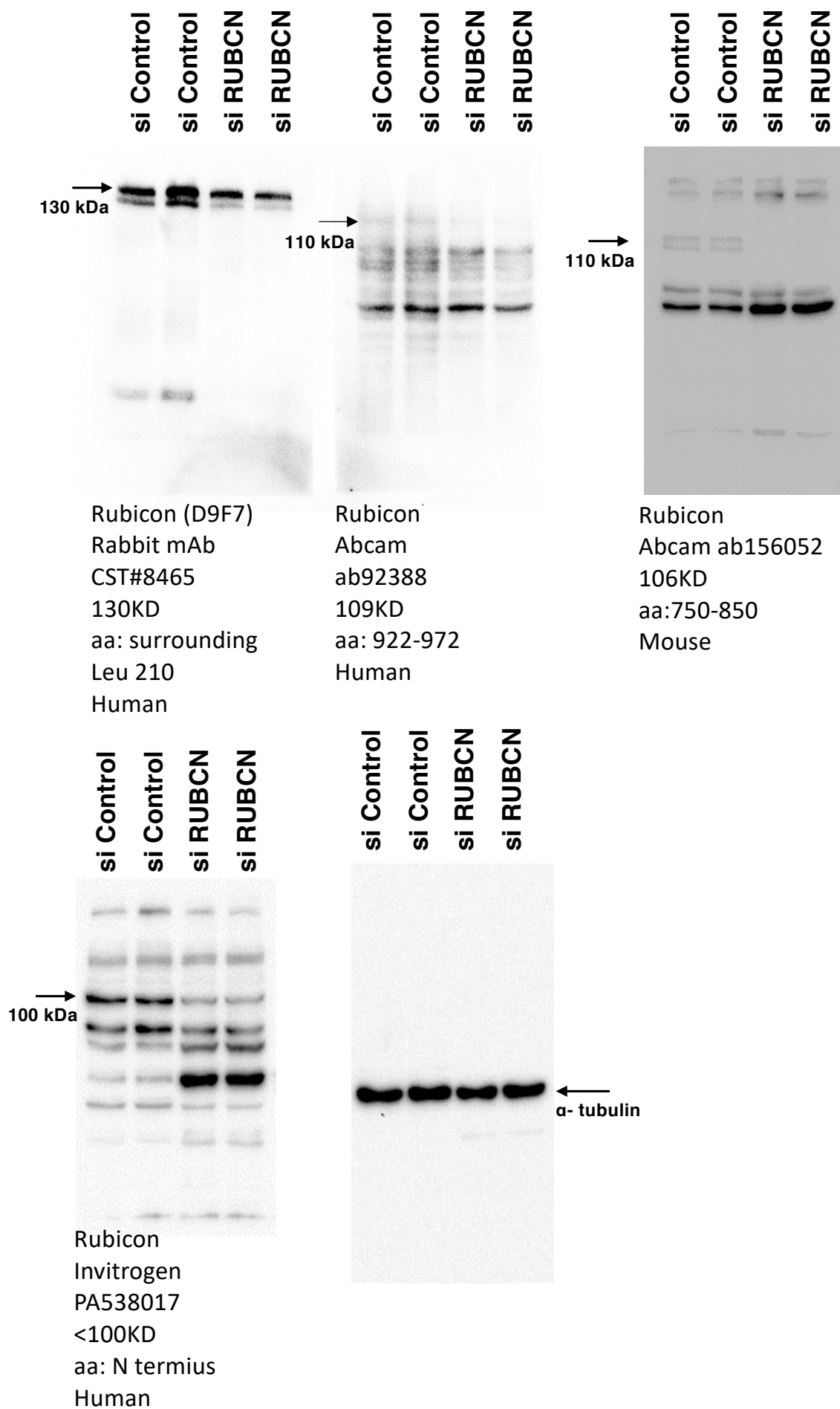
Supplementary Figure S4- Selective EGFR deletion in podocytes had no effect on body weight, fasting glucose and HbA1c in *nos3*^{-/-}; ***db/db* mice**. EGFR[f/f]; *nos3*^{-/-}; *db/db* (*nos3*^{-/-}; *db/db*) and nphs2-Cre; *egfr*[f/f]; *nos3*^{-/-}; *db/db* (*egfr*[podKO]; *nos3*^{-/-}; *db/db*) had similar body weights , fasting blood glucose and HbA1c levels at 8 weeks old and 20 weeks old (8 weeks: n = 19 for *nos3*^{-/-}; *db/db* group and n = 25 for *egfr*[podKO]; *nos3*^{-/-}; *db/db* group; 20 weeks: n = 16 for *nos3*^{-/-}; *db/db* group and *n* = 22 for *egfr*[podKO]; *nos3*^{-/-}; *db/db* group). Two-tailed unpaired student's t -test.

Supplementary Figure S5- Selective EGFR deletion in podocytes led to decreased renal T lymphocyte infiltration in *nos3*^{-/-}; ***db/db* mice**. *egfr*[f/f]; *nos3*^{-/-}; *db/db* (*nos3*^{-/-}; *db/db*) and nphs2-Cre; *egfr*[f/f]; *nos3*^{-/-}; *db/db* (*egfr*[podKO]; *nos3*^{-/-}; *db/db*) were sacrificed at 20 weeks old. Podocyte EGFR deletion led to decreases in renal T lymphocyte infiltration, as indicated by CD3 staining, a marker of T lymphocytes. Original magnification: x 400.
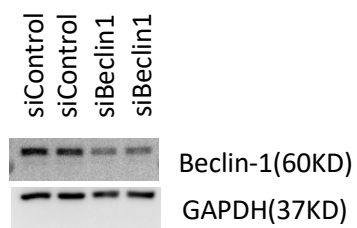
Supplementary Figure S6- Selective EGFR deletion in podocytes led **to decreased renal oxidative stress.** *egfr*[f/f]; *nos3*^{-/-}; *db/db* (*nos3*^{-/-}; *db/db*) and nphs2-Cre; *egfr*[f/f]; *nos3*^{-/-}; *db/db* (*egfr*[podKO]; *nos3*^{-/-}; *db/db*) were sacrificed at 20 weeks old. Podocyte EGFR deletion led to decreased renal oxidative stress as indicated by decreased 4-HNE staining, a marker of oxidative stress. Original magnification: x 400.

Supplementary Figure S7- Selective EGFR deletion in podocytes had no effect on macrophage M2 polarization. *egfr*^f/f^; *nos3*^-/-^; *db/db* (*nos3*^-/-^; *db/db*) and nphs2-Cre; *egfr*^f/f^; *nos3*^-/-^; *db/db* (*egfr*^podKO^; *nos3*^-/-^; *db/db*) were sacrificed at 20 weeks old. Podocyte EGFR deletion had no effect on M2 polarization of macrophages, as indicated by comparable mRNA levels of M2 markers, including mannose receptor (Mrc1, CD206), IL4Rα, arginase 1, YM1 and FIZZ1, in *nos3*^-/-^; *db/db* and *egfr*^podKO^; *nos3*^-/-^; *db/db* mice (n = 5 in each group). Two-tailed unpaired student's t -test.

Supplementary Figure S8- Podocyte EGFR deficiency led to increased autophagy in podocytes in *nos3*^-/-^ *db/db* mice. Both *nos3*^-/-^; *db/db* (*nos3*^-/-^; *db/db*) mice and *nos3*^-/-^; *db/db* (*Egfr*^podKO^; *nos3*^-/-^; *db/db*) mice were sacrificed at 20 weeks old. Representative kidney sections were stained and indicated that podocyte EGFR deletion decreased SQSTM1 and protein expression and increased LC3b

si Control
si Control
si RUBCN
si RUBCN

130 kDa →

Rubicon (D9F7)
Rabbit mAb
CST#8465
130KD
aa: surrounding
Leu 210
Human

si Control
si Control
si RUBCN
si RUBCN

110 kDa →

Rubicon
Abcam
ab92388
109KD
aa: 922-972
Human

si Control
si Control
si RUBCN
si RUBCN

110 kDa →

Rubicon
Abcam ab156052
106KD
aa:750-850
Mouse

si Control
si Control
si RUBCN
si RUBCN

100 kDa →

Rubicon
Invitrogen
PA538017
<100KD
aa: N termius
Human

si Control
si Control
si RUBCN
si RUBCN

← α- tubulin

Supplemental Figure S1

siControl
siControl
siBeclin1
siBeclin1

Beclin-1(60KD)

GAPDH(37KD)

SignalSilence® Beclin-1 siRNA I #6222
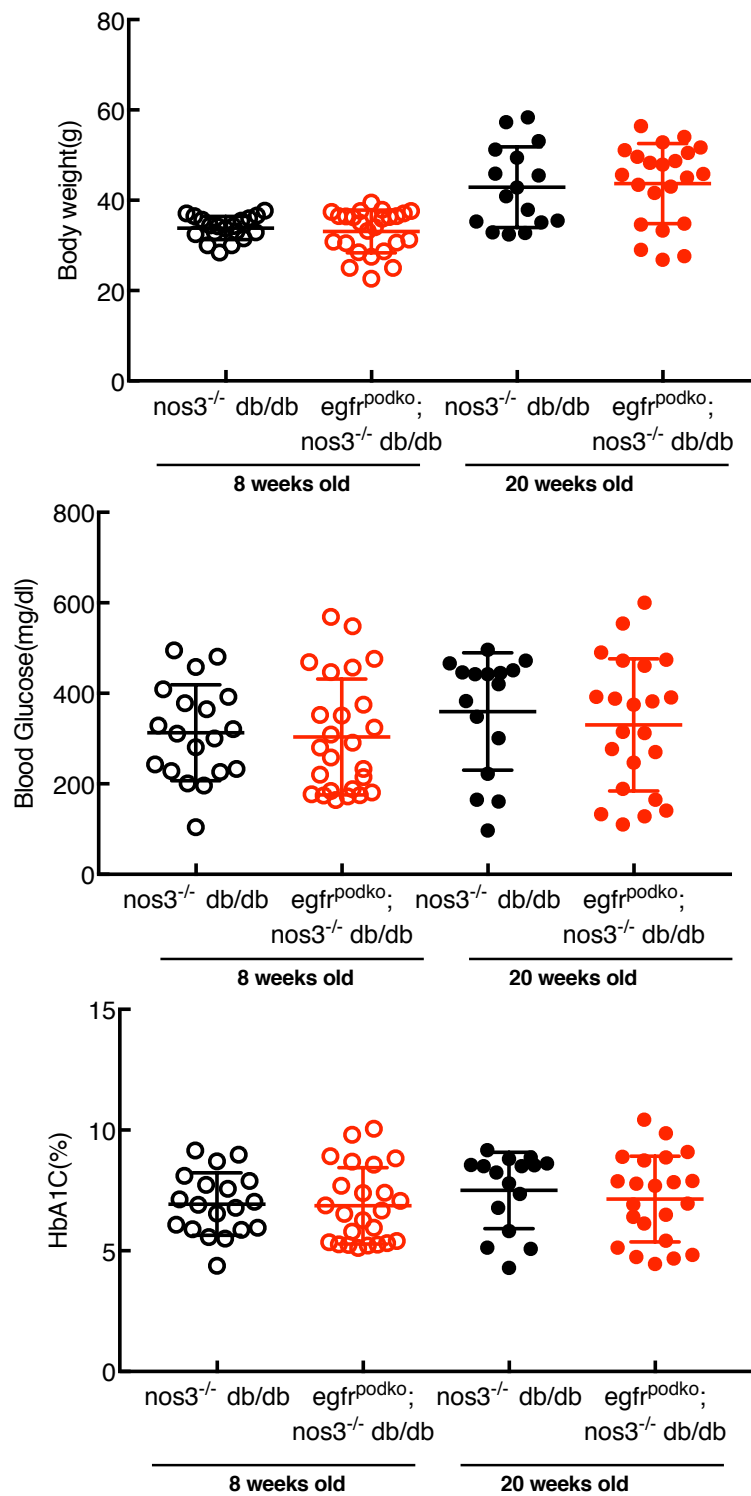Tranfection 10nM siControl or siBeclin1 48 hour.
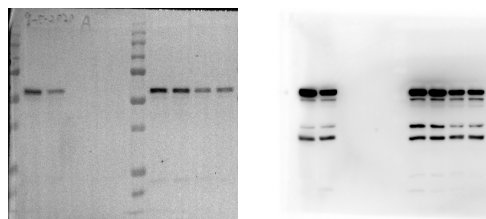
sFigure To confirm Belin-1 IF staining was specific
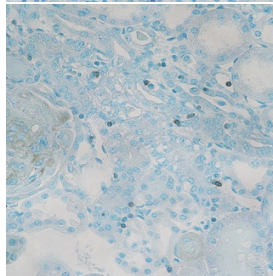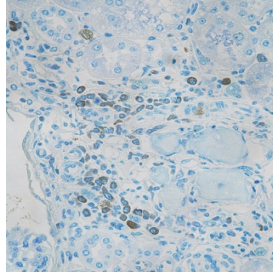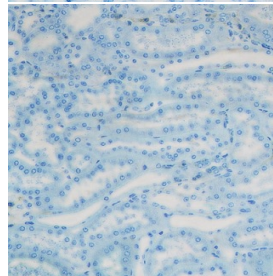signaling but not background. Do beclin1 Knockdown
experiment.

sicontrol

siBeclin1

Beclin-1/DAPI

sicontrol

siRubicon

Rubicon/DAPI

Supplemental Figure S2
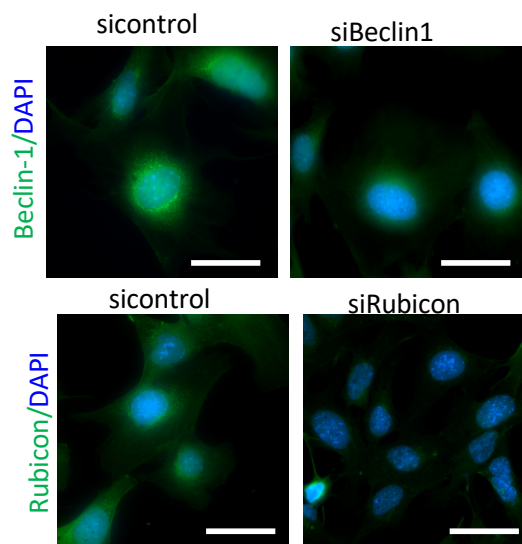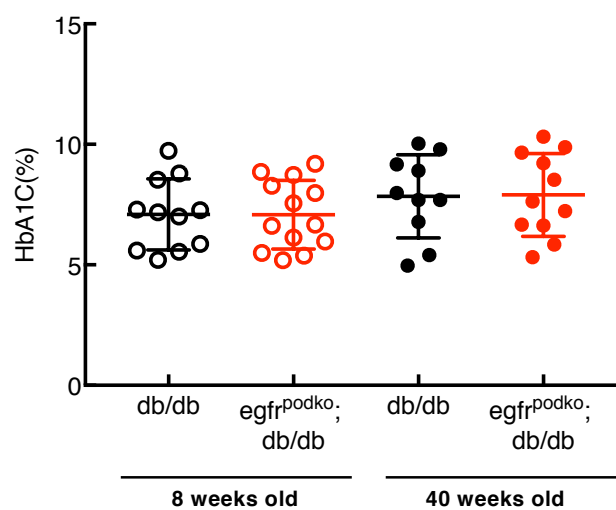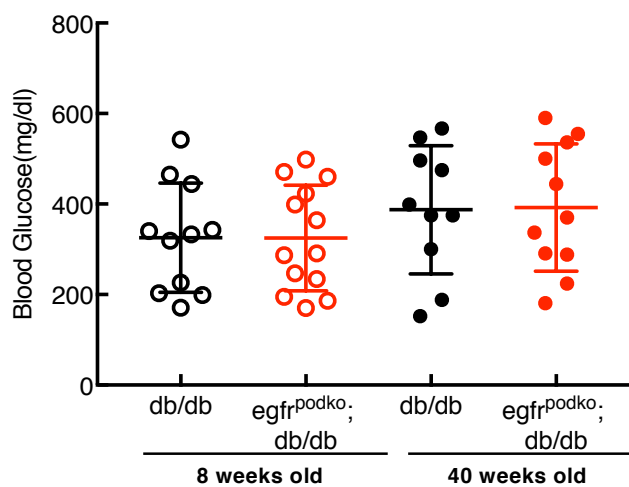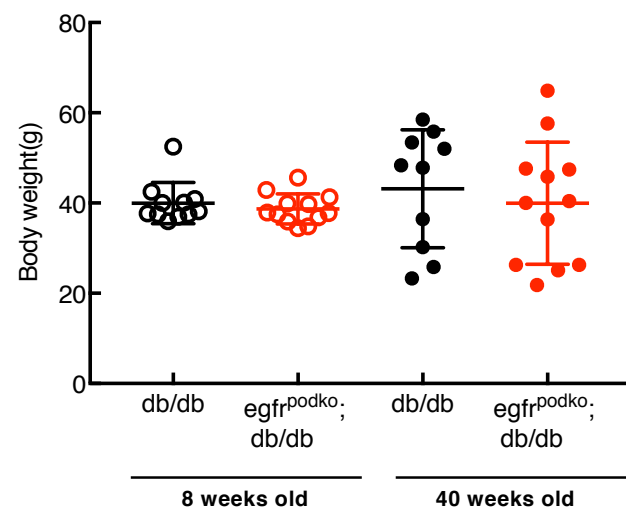
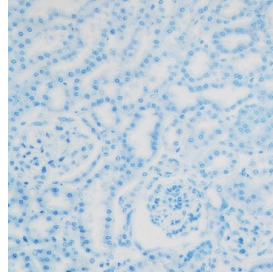**Supplemental Figure S3**

Supplemental Figure S4

Nos3-/- db/db   *Egfr*^{podKO} *;Nos3-/- db/db*

CD3, marker of lymphocytes, x 400.

Supplemental Figure S5

Nos3-/- db/db   ***Egfr*^{podKO} ; *Nos3-/- db/db***
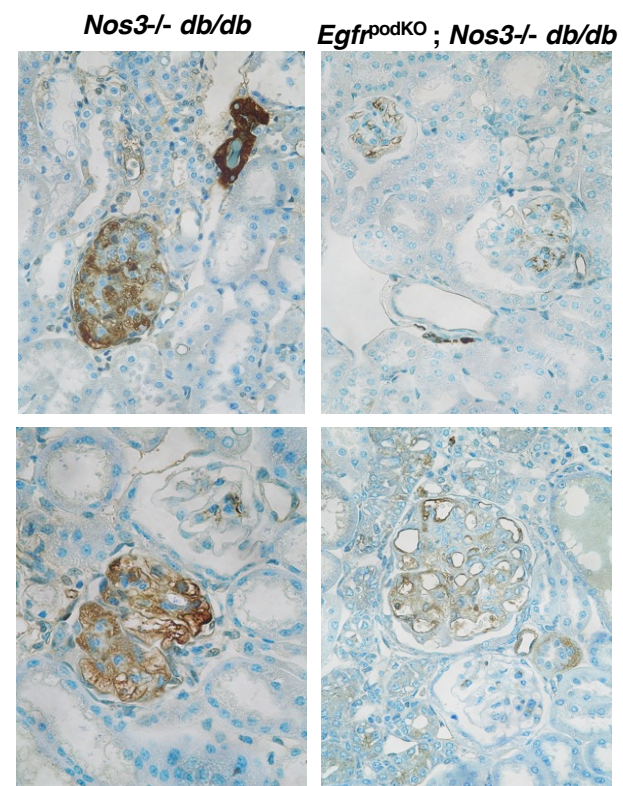
4-HNE staining, marker of oxidative stress, x 400.

Supplemental Figure S6

Supplemental Figure S7

nos3⁻/⁻ **nos3⁻/⁻ db/db** **egfr ᵖᵒᵈᵏᵒ; nos3⁻/⁻ db/db**

P62/DAPI

LC3b/DAPI

Supplemental Figure S8