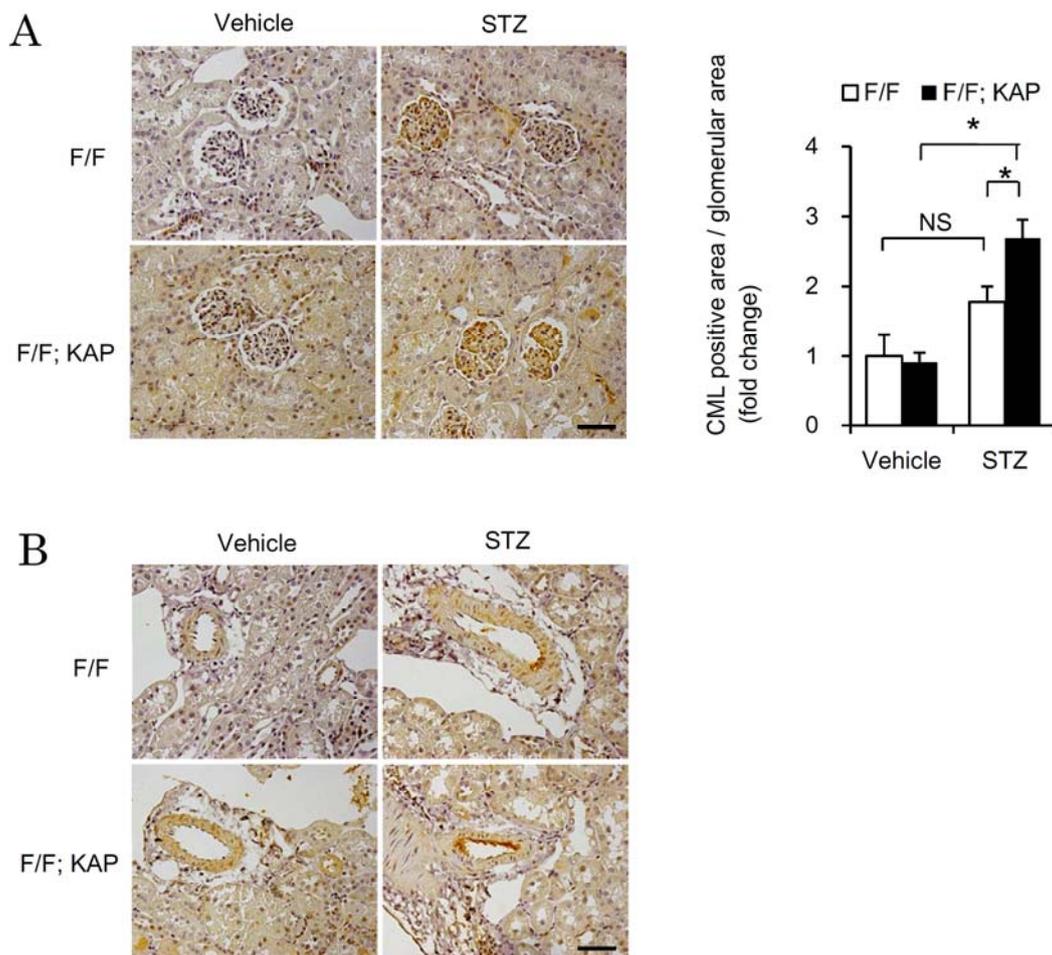


SUPPLEMENTARY DATA

Supplementary Figure 1.

Accumulation of AGEs in glomerulus and vascular vessels in the kidney of diabetic autophagy-deficient mice.

A and B: Representative images of immunohistochemical staining for CML in glomerulus (A) and intrarenal vascular vessels (B) of vehicle- or STZ-treated *Atg5^{F/F}:KAP* or control (*Atg5^{F/F}*) mice. Quantitative analysis for CML-positive area in glomerulus is performed in at least 10 high power fields (x 400) in each mouse. The mean value of vehicle-treated control mice is adjusted to “1” as a reference. F/F, *Atg5^{F/F}* mice; F/F:KAP, *Atg5^{F/F}:KAP* mice. Bars: 50 μ m. Data are means \pm SE (n = 6, vehicle-treated mice; n = 10, STZ-treated mice); * $p < 0.05$; NS, not significant.

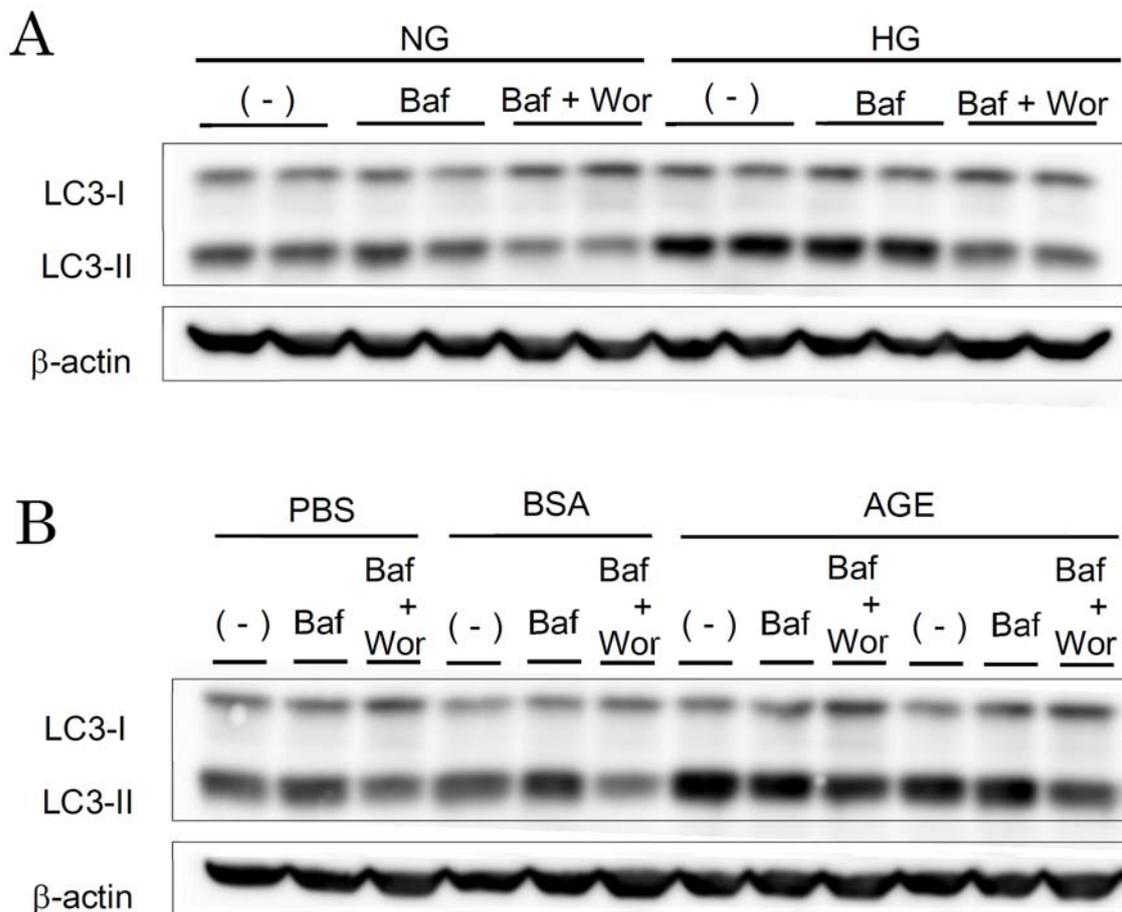


SUPPLEMENTARY DATA

Supplementary Figure 2.

Autophagosome formation is not blocked by high glucose or AGEs.

A and B: Western blot analysis for LC3 using the lysates of *Atg5* (+) PTECs which had been exposed to high- and normal-glucose medium for 72 hours (A) and vehicle (PBS), BSA, or AGE-BSA for 24 hours. To investigate which step in autophagy flux is disturbed by high glucose or AGEs, bafilomycin (100 nM) with or without wortmannin (200 nM) is administered to the medium 60 minutes before harvest. NG, normal glucose; HG, high glucose; Baf, Bafilomycin; Wor, Wortmannin

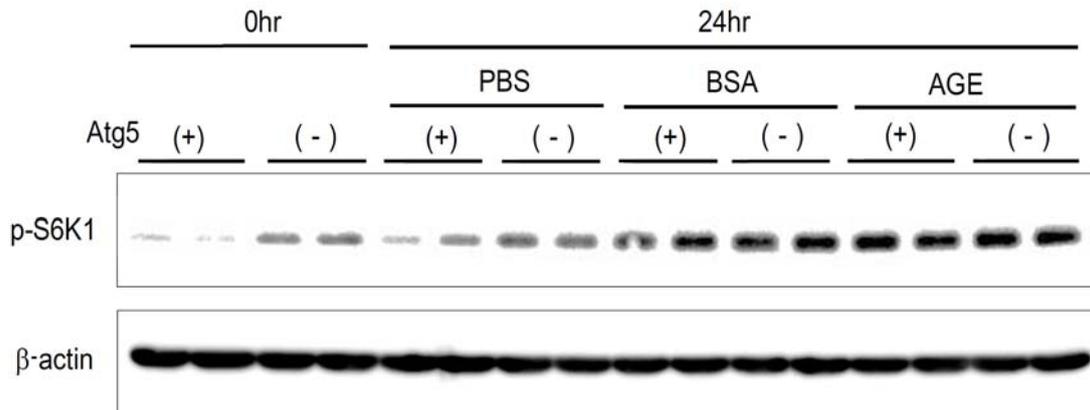


SUPPLEMENTARY DATA

Supplementary Figure 3.

Effects of AGEs on mTOR pathway in the *Atg5* (+) and *Atg5* (-) PTECs .

Western blot analysis for p-S6K1 is performed using the lysates of *Atg5* (+) or *Atg5* (-) PTECs, which had been exposed to vehicle (PBS), BSA, or AGE-BSA for 24 hours. *Atg5* (+), *Atg5* (+) PTECs; *Atg5* (-), *Atg5* (-) PTECs



SUPPLEMENTARY DATA

Supplementary Figure 4.

Formation of SQSTM1/p62-positive dots in autophagy-competent and -deficient diabetic kidney.

Representative images of immunohistochemical staining for p62 using the kidneys of vehicle- or STZ-treated *Atg5^{F/F}:KAP* or control mice (original magnification; x 1000). Quantification of the number of SQSTM1/p62-positive dots is performed in at least 10 high power fields (x 400) in each mouse (right panel). F/F, *Atg5^{F/F}* mice; F/F:KAP, *Atg5^{F/F}:KAP* mice. Bars: 20 μ m; data are means \pm S.E. (n = 6, vehicle-treated mice; n = 10, STZ-treated mice); * $p < 0.05$; NS, not significant.

