Supplementary Table 1 Physical and biochemical parameters of mice received STZ by intraperitoneal injection

Variable	Control	DN mice
Body weight (g)	30.2±2.4	23.3±2.5 **a**
Glucose (mmol/l)	5.6±1.1	20.9±3.4 **a**
Heart rate (beats/min)	478.4±41.3	483.8±32.6
Relative kidney weight (mg/g body weight)	10.33±0.93	17.54±2.17 **a**
UACR(mg/mmol)	3.64±0.76	15.42±3.44 **a**

Abbreviations: STZ, streptozotocin; DN mice, diabetic nephropathy mice; UACR, Urinary albumin/creatinine ratio, **a**: P<0.05 vs. Control.

Supplementary Table 2 Clinical data in the kidney from diabetic subjects with or without DN.

Patient	Age at Biopsy(yr)	Sex	CRP (mg/dl)	DM (years)	Scr (μmol/L)	Serum Urea (mmol/L)	UPE/24h (g)	eGFR (ml/min/1.73m	HbA1c (%)	Macrovascular Disease	AD	Renal Disease Other

								2)				**than DN**
Normal controls												
1	59	M	1.78	NA	59	3.58	<0.15	**115**	NA	N	N	N
2	48	F	<0.35	NA	68	4.67	<0.15	**103**	NA	N	N	N
3	63	M	2.78	NA	57	4.35	<0.15	**96**	NA	N	N	N
4	59	M	1.21	NA	60	5.11	<0.15	**127**	NA	N	N	N
5	71	F	3.76	NA	96	7.08	<0.15	**81**	NA	N	N	N
mean±SD	60±8.3		2.38±1.12		68±16.2	4.96±1.31		**104.4±17.6**				
DN group												
1	46	M	NA	1	187	9.78	3.21	**51**	10.3	N	N	N
2	63	M	NA	5	197	17.21	4.65	**31**	6.9	IHD	N	N
3	76	F	NA	10	231	15.7	6.17	**26**	8.3	N	N	N
4	79	F	NA	11	279	22.1	5.97	**21**	9.4	PVD	N	N
5	51	M	NA	2	142	16.87	3.14	**47**	NA	N	N	N

			207.2				
mean±S		5.8±4.5	±51.2	16.33±4.4	4.63±1.4	**35.2±**	
D	63±14.6	5	a	1	5	**13.2 b**	8.7±1.5

SCr, serum creatinine; UPE, urinary protein excretion; eGFR, estimated GFR; CRP, C-reactive protein; HbA1c, hemoglobin A1c; AD, autoimmune disease; F, female; M, male; DN, diabetic nephropathy; IHD, ischemic heart disease; N, not present; PVD, peripheral vascular disease;NA, not applicable/not available.**a**: P<0.05 vs. Control/Normal control. **b**: P<0.01 vs. control/Normal control.

Supplementary Table 3 Physical and biochemical parameters of mice received AAV by tail vein injection

Variable	**Control(Scramble)**	**DN(Scramble)**	**Control(shRNA-β-arrestin-2)**	**DN(shRNA-β-arrestin-2)**
Body weight (g)	31.1±1.3	22.4±0.5 **a**	30.4±1.9	23.5±1.5 **a**
Glucose (mmol/l)	6.4±0.9	22.2±5 **b**	6.2±0.8	23.6±4.7 **b**
Heart rate (beats/min)	466.9±15.3	473.0±15.6	470.9±18.1	468.3±23.6
Relative kidney weight (mg/g body weight)	10.43±0.84	18.73±1.20 **a**	10.40±0.86	15.63±1.55 **ad**

Abbreviations: AAV, adeno-associated virus; DN, diabetic nephropathy; **a**: P<0.05 vs. Scramble/control. **b**: P<0.01 vs. Scramble/control. d: P<0.05 vs. Scramble/STZ-induced DN mice.

Supplementary Table 4 Primer pairs of target genes used for real time RT-PCR in this study

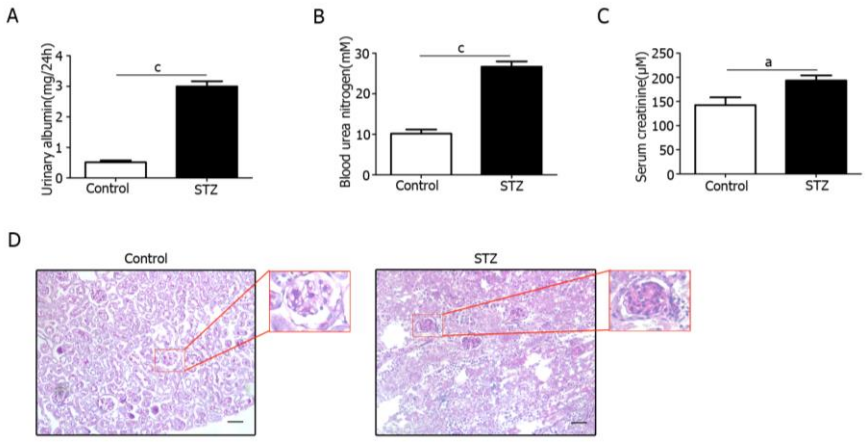
Genes	Accession No.	Forward	Reverse
Mus β-arrestin1	NM_178220.3	ACCTTTGAGATCCCGCCAAA	CAGGGGCATACTGAACCTTC
Mus β-arrestin2	NM_001271360.1	GGAGTAGACTTTGAGATTCGAGC	CTTTCTGATGATAAGCCGCACA
Mus β-actin	NM_007393.3	GGCTGTATTCCCCTCCATCG	CCAGTTGGTAACAATGCCATGT

Supplementary Table 5 Antibodies used in this study

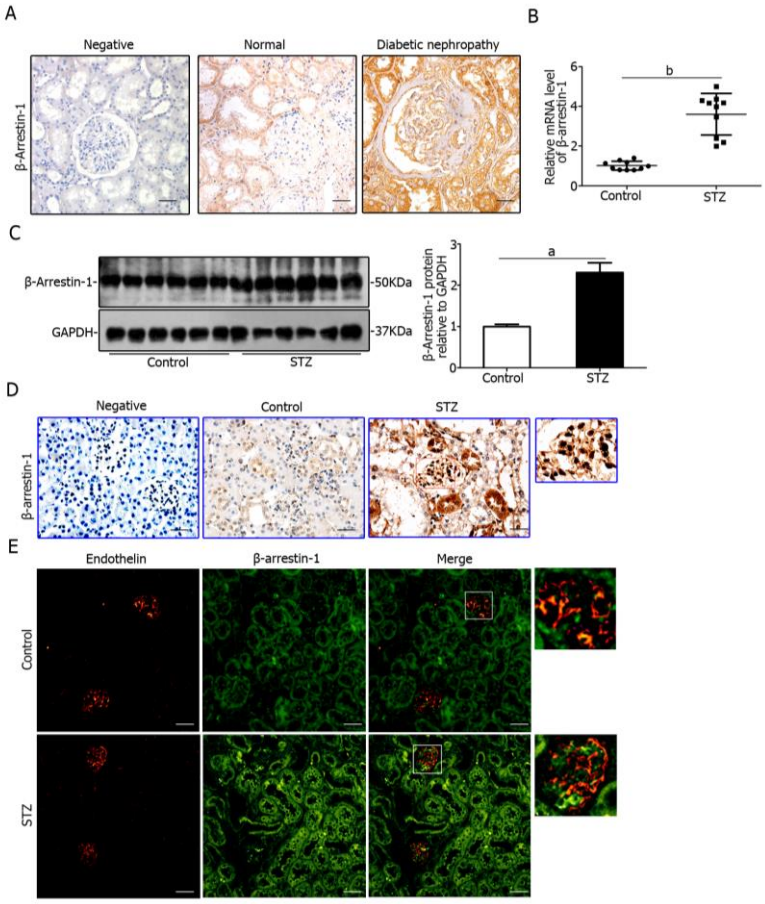
Primary antibodies	**Host**	**Dilution and supplier**	**Product ID**	**Application**
β-arrestin1	Rabbit	1:1000(1:100 for IHC, IF); Bioworld Technology, Louis Park, MN	BS2213	WB, IF, IHC

β-arrestin2	Rabbit	1:1000 (1:50 for IHC, IF); ProteinTech Group, Chicago, IL	10171-1-AP	WB, IF, IHC
Endothelin	Mouse	1:100 for IF; Invitrogen, Thermo Fisher Scientific	MA3-005	IF
Goat anti-Mouse Alexa Fluor 633	Goat	1:200; Invitrogen, Thermo Fisher Scientific	A-21050	IF
Goat anti-Rabbit Alexa Fluor 488	Goat	1:200; Invitrogen, Thermo Fisher Scientific	A32790	IF
Bcl-2	Rabbit	1:1000; ProteinTech Group, Chicago, IL	12789-1-AP	WB
Bax	Rabbit	1:1000; ProteinTech Group, Chicago, IL	60267-1-lg	WB
β-Actin	Mouse	1:5000; ProteinTech Group, Chicago, IL	66009-1-Ig	WB
Bip	Rabbit	1:1000; Cell Signaling, Danvers, MA	3177	WB
Chop	Mouse	1:1000; Cell Signaling, Danvers, MA	2895	WB
e-NOS	Rabbit	1:1000; Cell Signaling, Danvers, MA	32027	WB
GAPDH	Mouse	1:4000; ProteinTech Group, Chicago, IL	60004-1-Ig	WB
ZO-1	Rabbit	1:1000; Cell Signaling, Danvers, MA	8193	WB
Occludin	Rabbit	1:1000; Cell Signaling, Danvers, MA	91131	WB
ATF4	Mouse	1:1000; Cell Signaling, Danvers, MA	97038	WB
ATF6	Rabbit	1:1000; Abcam	Ab134561	WB
p-PERK	Rabbit	Affinity Biosciences LTD.	DF7576	WB
Histone H3	Rabbit	1:3000; Abcam	Ab1791	WB

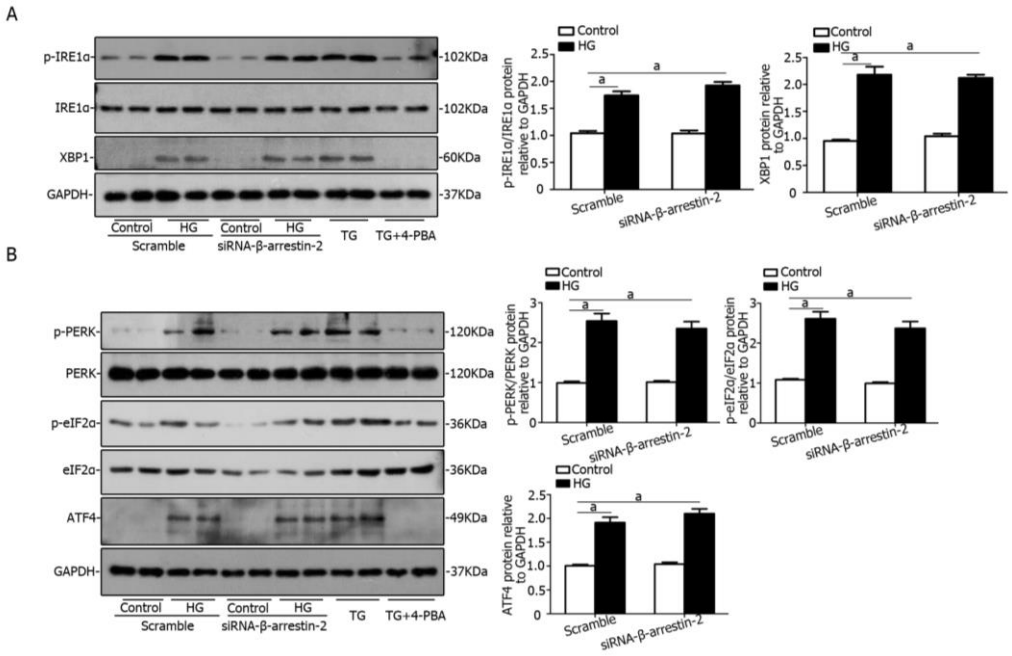
PERK	Rabbit	Affinity Biosciences LTD.	AF5304	WB
IRE1	Rabbit	Affinity Biosciences LTD.	AF7651	WB
p-IRE1 (Ser724)	Rabbit	Affinity Biosciences LTD.	AF7150	WB
XBP1	Mouse	1:1000; Cell Signaling, Danvers, MA	27901	WB
eIF2α	Rabbit	Affinity Biosciences LTD.	AF6087	WB
p-eIF2α	Rabbit	Affinity Biosciences LTD.	AF3087	WB



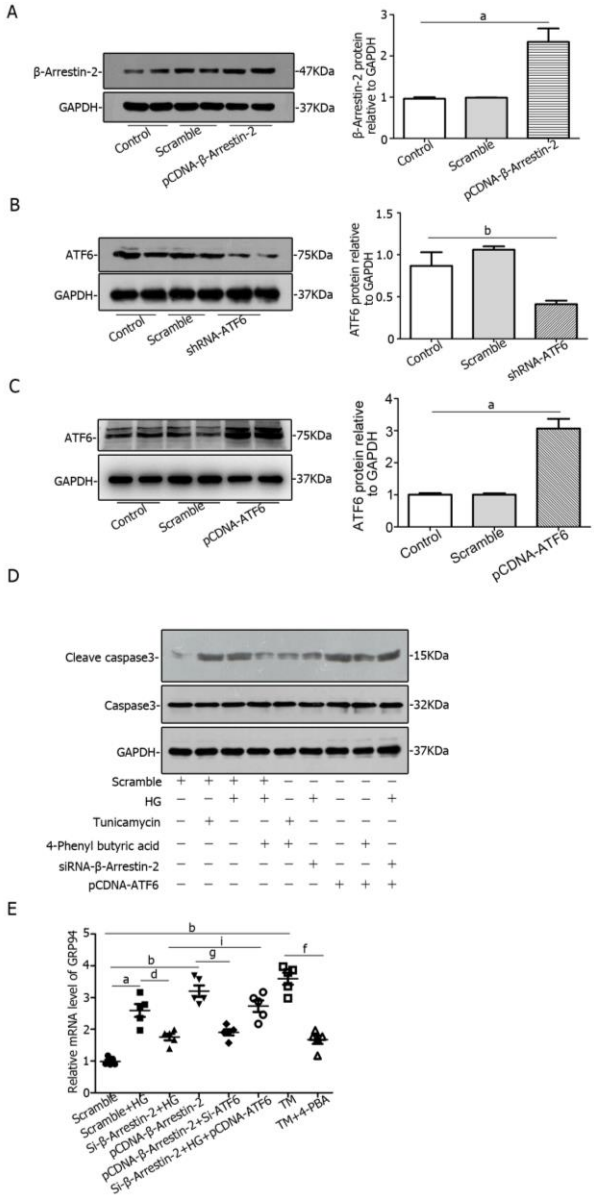
Supplementary Figure 1 The data showed renal injury in the mice model of diabetic nephropathy(DN) induced by streptozotocin (STZ). (A) The urinary albumin in the mice at the 24 hours. (c: P<0.001 vs. control, n=8) **(B)** The blood urea nitrogen in the mice from different groups. (c: P<0.001 vs. control, n=8). **(C)** Serum creatinine in the mice from different groups. (a: P<0.05 vs. control, n=8) **(D)** The images of PeriodicAcid-Schiff stain (PAS) staining in control and STZ-induced DN mice. (black bars=50μm).



Supplementary Figure 2 The expression of β-Arrestin-1 was increased significantly in renal biopsies from diabetic nephropathy patients but not in glomerular endothelial cells (GENC) from mice with diabetic nephropathy.(A) Representative images of IHC staining showing β-arrestin-1 expression in human renal paraffin section from normal people and DN patients. (black bars=20μm) **(B)** Relative mRNA levels of β-arrestin-1 in the renal cortex from STZ-induced diabetic nephropathy mice (mean±SD, b: $P<0.01$ vs. control, n=5). **(C)** Western blot showing the expression of β-arrestin-1 in the renal cortex from STZ-induced DN mice. (mean±SD, a: $P<0.05$ vs. control, n=8) **(D)** Representative images of IHC staining of β-arrestin-1 expression in STZ-induced DN mice. (black bars=20μm) (**E**) Detection of β-arrestin-1 expression in glomerular endothelial cells (GENC) in STZ-induced diabetic nephropathy mouse model by immunofluorescence double labeling: endothelin (red, mark protein in GENC), β-arrestin-1 (green). (white bars=20μm)

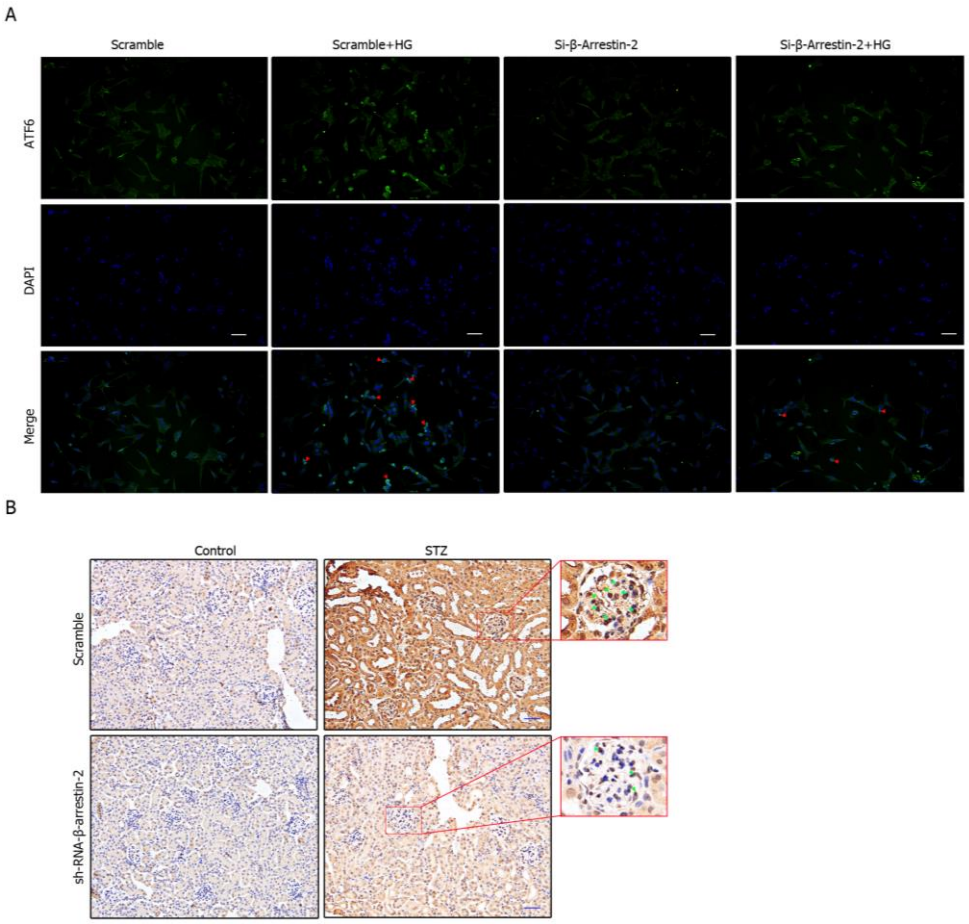


Supplementary Figure 3 There was no effect of silencing of β-arrestin-2 on the PERK and IRE signal pathway of ER stress. (A) Immunoblotting images and summarize data showing the effects of β-arrestin-2 knockdown on the expression of p-IRE1α, IRE1α and XBP1 in GENC under HG condition. (a: P<0.05 vs. Scramble/control, n=6) **(B)** Immunoblotting images and summarize data showing the effects of β-arrestin-2 knockdown on the expression of p-PERK, PERK, p-eIF1α and ATF4 in HG-treated GENC. (a: P<0.05 vs. Scramble/control, n=6)



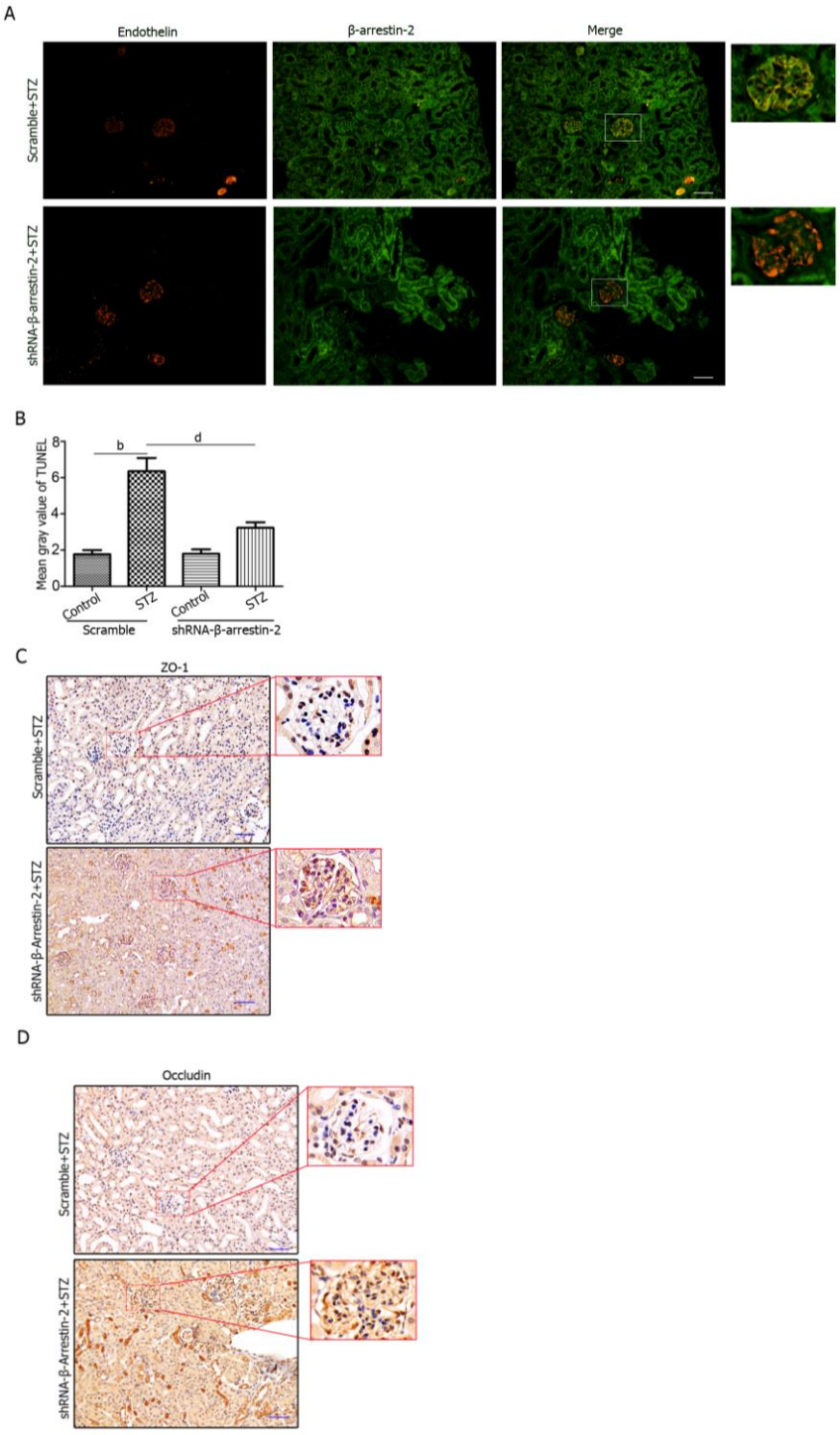
Supplementary Figure 4 (A) Representative western blotting and summarized data showing the overexpression of β-arrestin-2 by pCDNA-β-arrestin-2 plasmids transfection. (a: *P*<0.05 vs. control, n=6) **(B)** Images and summarized data showing the efficiency of silencing ATF6 with shRNA-ATF6 by western blotting. (b: *P*<0.01 vs. control, n=6) **(C)** Representative western blotting and summarized data showing that overexpression of ATF6 by pCDNA-ATF6 plasmids transfection. (a: P<0.05 vs. scramble, n=6) **(D)** Representative western blotting showing that the expression of cleaved caspases-3 and the whole caspase-3 under various stimuli. **(E)** Relative mRNA levels of GRP94 which is one of the target genes

regulated by ATF6 in GENC with various stimuli (mean±SD). (a: P<0.05 vs. Scramble/control, b: P<0.01 vs. Scramble/control, d: P<0.05 vs. Scramble/HG treated group, f: P<0.05 vs. TM treated group, g: P<0.05 vs. pCDNA-β-arrestin-2 treated group, i: P<0.05 vs. Si-β-arrestin-2 /HG treated group, n=6)



Supplementary Figure 5 (A) The image of immunofluorence (IF) of GENC showing activating transcription factor 6 translocated into nucleus under high glucose which was inhibitted by silencing of β-arrestin-2. Red arrows means ATF6 translocated into the nucleus. (white bar=50μm). **(B)** The image of immunohistochemical (IHC) showing the expression of ATF6 in kidney from different groups. Green arrows means ATF6 translocated into the nucleus.

(blue bars=50μm).



Supplementary Figure 6 (A) The image of immunofluorence (IF) showing the expression of β-arrestin-2 in glomerular endothelial cells (GENC) from different groups. (white bar=50μm) **(B)** The quantifications of the TUNEL staining from different groups. (b: $P<0.01$,vs. Scramble/control, d: $P<0.05$,vs. Scramble/STZ, n=6). **(C)** Representative images of immunohistochemical (IHC) staining showing ZO-1 expression in mice renal from different groups. (blue bars=50μm) **(D)** Representative images of immunohistochemical (IHC) staining showing occludin expression in mice renal from different groups. (blue bars=50μm).