

Sodium butyrate ameliorated diabetic nephropathy-associated tubulointerstitial inflammation by modulating tight junction of renal tubular epithelial cells

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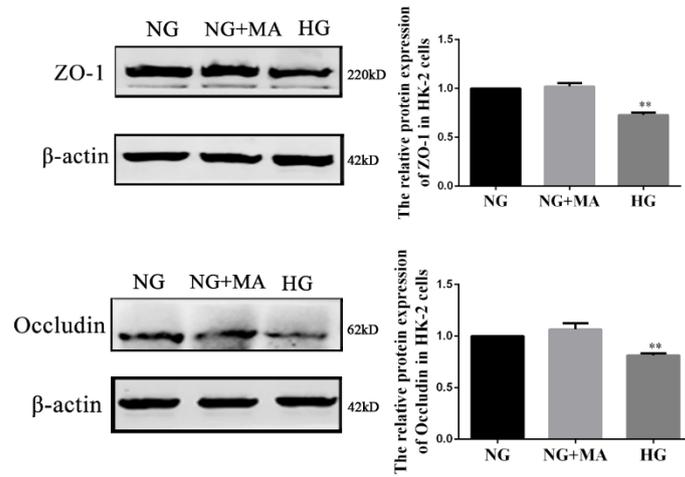


Fig S1. Effects of mannitol on TJ-related proteins of HK-2 cells. NG: cells treated with normal glucose 5.56 mmol/L; NG+MA: cells treated with normal glucose and mannitol 54.44 mmol/L; HG: cells treated with high glucose 60 mmol/L. Each bar represents the mean \pm SEM for groups of three. ** $P < 0.01$, means HG vs NG.

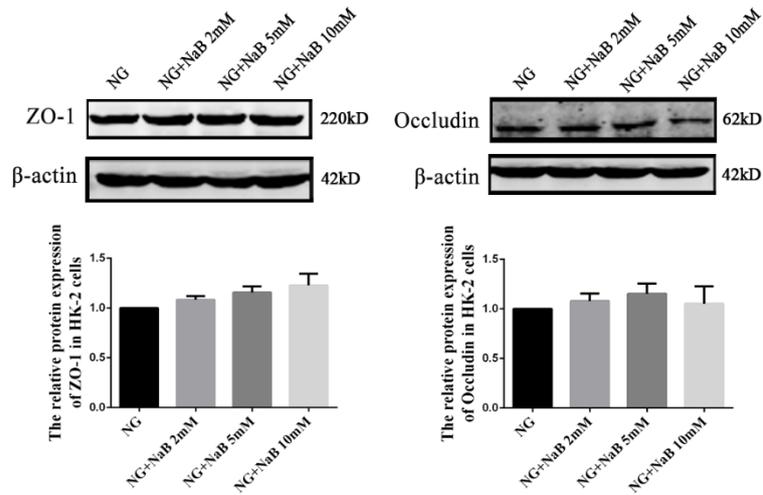


Fig S2. Effects of NaB on TJ-related proteins of NG cultured HK-2 cells. NG: cells treated with normal glucose 5.56 mmol/L; HG: cells treated with high glucose 60 mmol/L. HG+NaB 2 mM: cells treated with 60 mmol/L glucose and 2 mM NaB; HG+NaB 5 mM: cells treated with 60 mmol/L glucose and 5 mM NaB; HG+NaB 10 mM: cells treated with 60 mmol/L glucose and 10 mM NaB. Each bar represents the mean \pm SEM for groups of three.

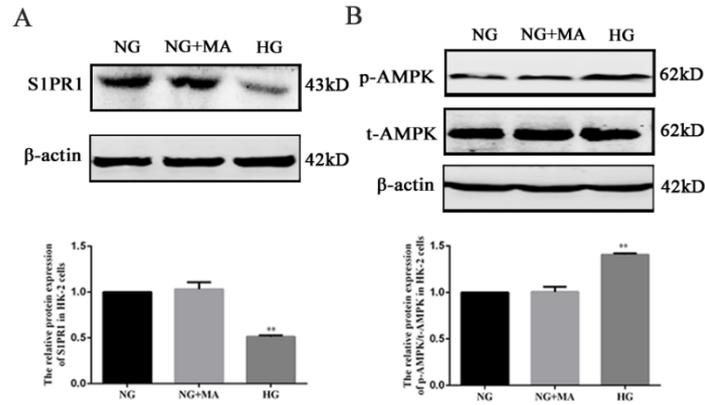


Fig S3. Effects of mannitol on S1PR1/AMPK signaling pathway of HK-2 cells. (A) The relative protein levels of S1PR1 through Western blot analysis in HK-2 cells. (B) The relative phosphorylation levels of p-AMPK through Western blot analysis in HK-2 cells. NG: cells treated with normal glucose 5.56 mmol/L; NG+MA: cells treated with normal glucose and mannitol 54.44 mmol/L; HG: cells treated with high glucose 60 mmol/L. Each bar represents the mean \pm SEM for groups of three. ** $P < 0.01$, means HG vs NG.

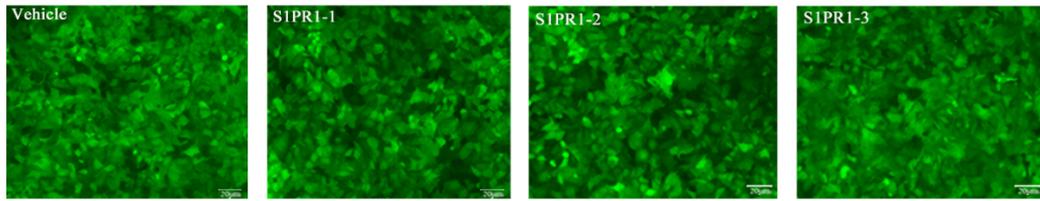


Fig S4. The expression of green fluorescent protein in HK-2 cells cultured with NG.

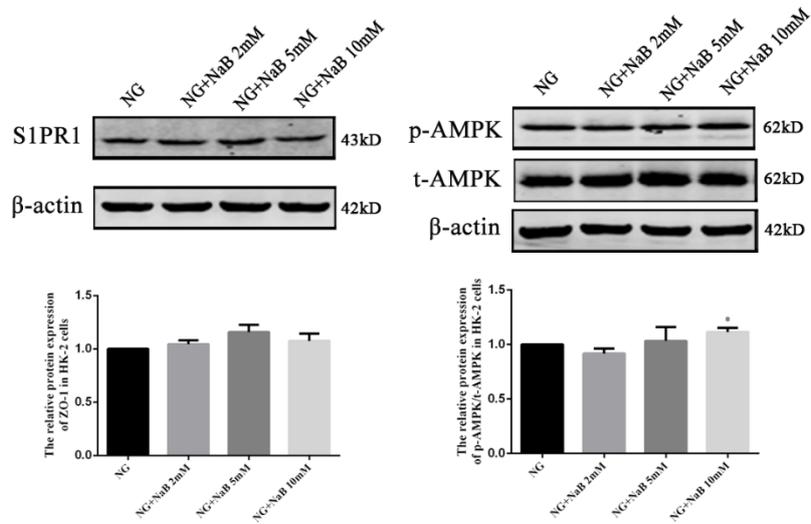


Fig S5. Effects of NaB on S1PR1/AMPK signaling pathway of NG cultured HK-2 cells. NG: cells treated with normal glucose 5.56 mmol/L; HG: cells treated with high glucose 60 mmol/L. HG+NaB 2 mM: cells treated with 60 mmol/L glucose and 2 mM NaB; HG+NaB 5 mM: cells treated with 60 mmol/L glucose and 5 mM NaB; HG+NaB 10 mM: cells treated with 60 mmol/L glucose and 10 mM NaB. Each bar represents the mean \pm SEM for groups of three. * $P < 0.05$, compared to NG as indicated.

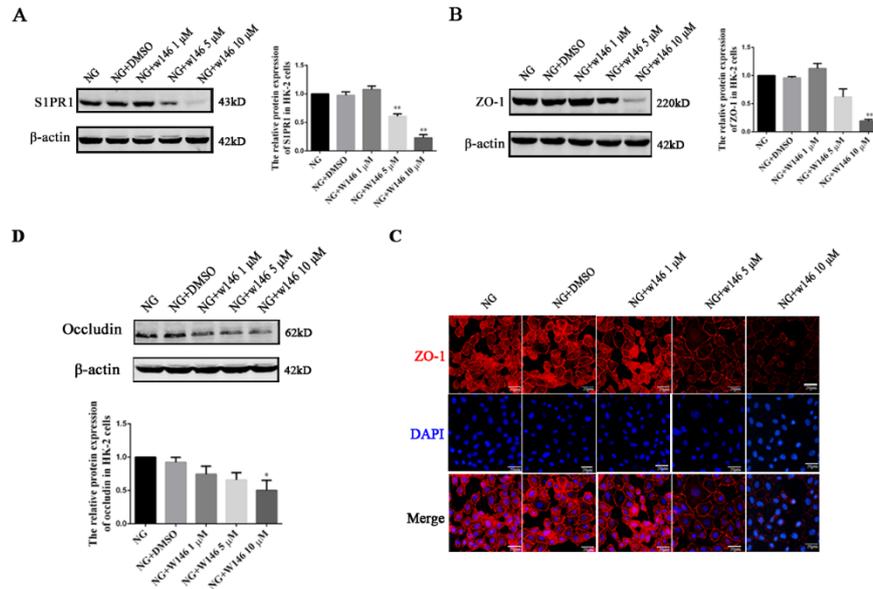


Fig S6. Effects of W146 on S1PR1/AMPK signaling pathway of NG cultured HK-2 cells. (A) The relative protein levels of S1PR1 through Western blot analysis in HK-2 cells exposed to 1 μ M, 5 μ M, and 10 μ M W146. (B) The relative protein levels of ZO-1 through Western blot analysis in HK-2 cells exposed to 1 μ M, 5 μ M, and 10 μ M W146. (C) The relative protein levels and distribution of ZO-1 through IF in NG cultured HK-2 cells exposed to 1 μ M, 5 μ M, and 10 μ M W146. (D) The relative protein levels of Occludin through Western blot analysis in HK-2 cells exposed to 1 μ M, 5 μ M, and 10 μ M W146. NG: cells treated with normal glucose 5.56 mmol/L; NG+DMSO: cells treated with dimethyl sulfoxide; NG+W146 1 μ M: cells treated with 5.56 mmol/L glucose and 1 μ M W146; NG+W146 5 μ M: cells treated with 5.56 mmol/L glucose and 5 μ M W146; NG+W146 10 μ M: cells treated with 5.56 mmol/L glucose and 10 μ M W146; Each bar represents the mean \pm SEM for groups of three. * P < 0.05, ** P < 0.01, compared to NG as indicated.

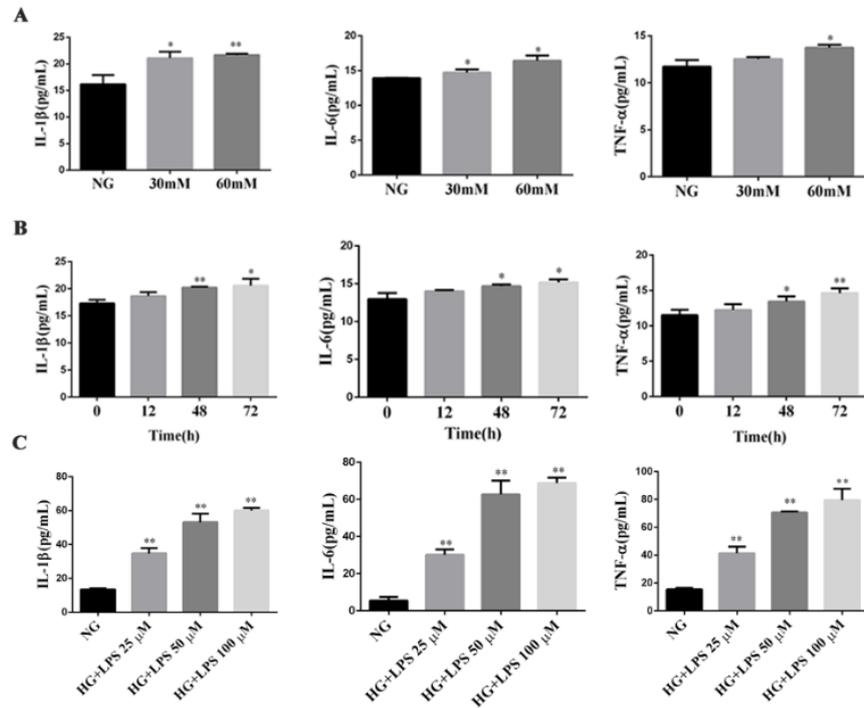


Fig S7. Effects of HG and LPS on the levels of IFs in HK-2 cells. (A) The protein levels of IL-1 β , IL-6 and TNF- α in the culture medium of HK-2 cells exposed to 5.56 mmol/L, 30 mmol/L and 60 mmol/L glucose for 72 h. (B) The protein levels of IL-1 β , IL-6 and TNF- α in the culture medium of HK-2 cells exposed to 60 mmol/L glucose for 0 h, 12 h, 48 h and 72 h. (C) The protein levels of IL-1 β , IL-6 and TNF- α in the culture medium of HK-2 cells exposed to HG and 25 μ M, 50 μ M, and 100 μ M LPS for 72 h. NG: cells treated with normal glucose 5.56 mmol/L; HG+LPS 25 μ M: cells treated with 60 mmol/L glucose and 25 μ M LPS; HG+LPS 50 μ M: cells treated with 60 mmol/L glucose and 50 μ M LPS; HG+LPS 100 μ M: cells treated with 60 mmol/L glucose and 100 μ M LPS. Each bar represents the mean \pm SEM for groups of three. * P < 0.05, ** P < 0.01, compared to NG as indicated.