

Figure S1. Short time treatment of dapagliflozin increased the activity of AMPK and but had no effect on autophagic flux. **(A)** HK-2 cells were treated with dapagliflozin (20 μ M, for 24 h) under HG conditions, and p-AMPK, p-S6RP were assessed by western blotting. **(B)** Densitometric evaluation of **(A)** (n=4). **(C)** HK-2 cells were treated with Cont, HG (30 mM), HG (30 mM) with dapagliflozin (20 μ M) for 24 h and chloroquine (50 μ M) or vehicle for 1 h. Autophagic flux was determined with the LC3-II ratio in the presence and absence of chloroquine. **(D)** Densitometric evaluation of **(C)** (n=4). All data represent the means \pm standard deviation (SD). *p<0.05 vs. the indicated group, **p<0.01 vs. the indicated group. Cont, control; HG, high glucose; DAPA, dapagliflozin; and CQ, chloroquine.

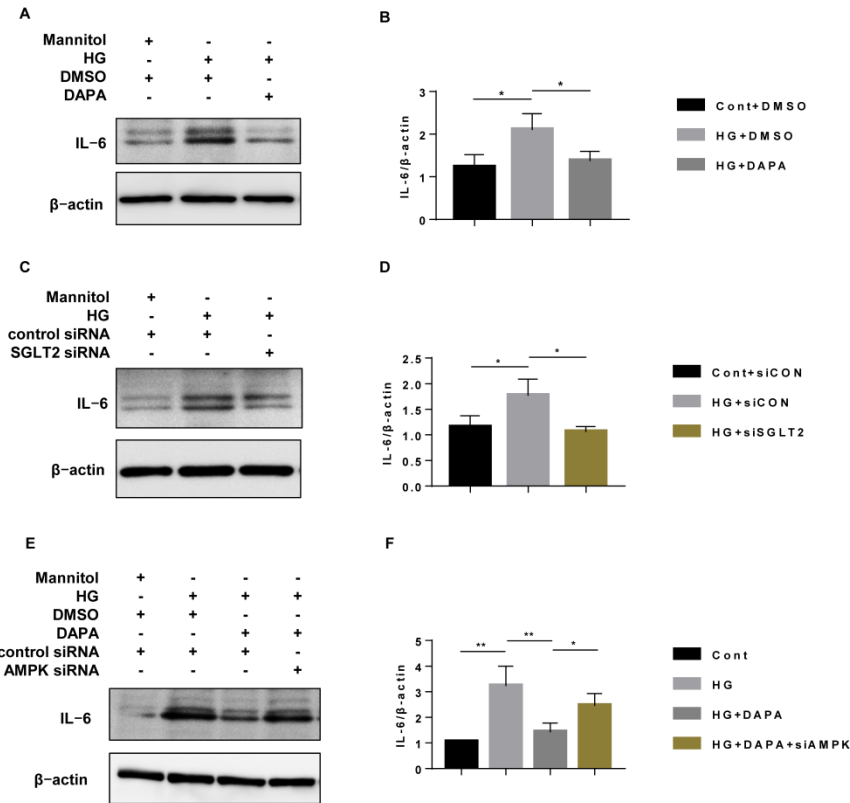


Figure S2. SGLT2 inhibition suppressed HG-induced IL-6 via activating AMPK. (A) HK-2 cells were incubated with Cont, HG (30 mM), or HG (30 mM) dapagliflozin (20 μ M) for 48 h, and IL-6 was assessed by western blotting. (B) Densitometric evaluation of (A) (n=3). (C) HK-2 cells were transfected with SGLT2 siRNA (200 nM) or control siRNA for 6 h and then incubated with Cont or HG (30 mM) medium for 48 h. IL-6 was assessed by western blotting. (D) Densitometric evaluation of (A) (n=4). (E) HK-2 cells were transfected with AMPK siRNA (200 nM) or control siRNA for 6 h and then incubated with Cont, HG (30 mM) or HG (30 mM) with dapagliflozin (20 μ M) for 48 h. The levels of IL-6 in cytoplasmic protein extracts were assessed by western blotting. (F) Densitometric evaluation of (E) (n=4). All data represent the means \pm standard deviation (SD). *p<0.05 vs. the indicated group, **p<0.01 vs. the indicated group (n=3). Cont, control; HG, high glucose; siCON, control siRNA; siAMPK, AMPK siRNA.

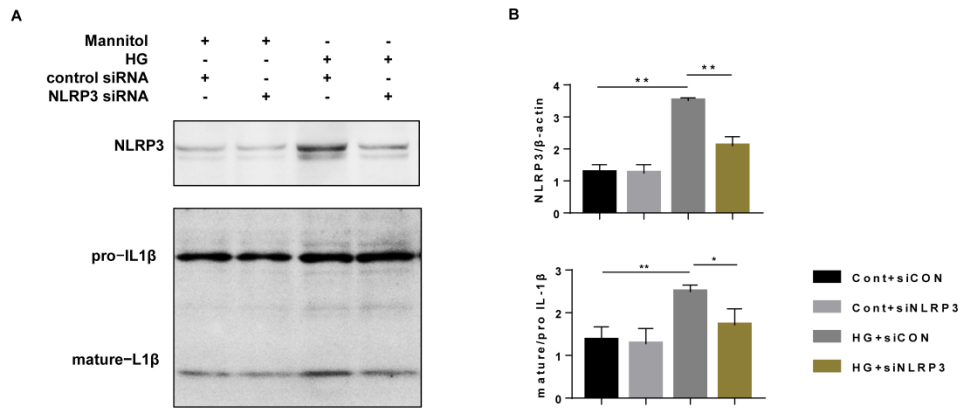


Figure S3. NLRP3 knockdown suppressed HG-induced activation of IL-1 β . **(A)** HK-2 cells were transfected with NLRP3 siRNA (200 nM) or control siRNA for 6 h and then incubated with Cont or HG (30 mM) medium for 48 h. The levels of NLRP3 and IL-1 β were assessed by western blotting. **(B)** Densitometric evaluation of (A) (n=3). All data represent the means \pm standard deviation (SD). **p<0.01 vs. the indicated group. Cont, control; HG, high glucose; siCON, control siRNA; siNLRP3, NLRP3 siRNA.

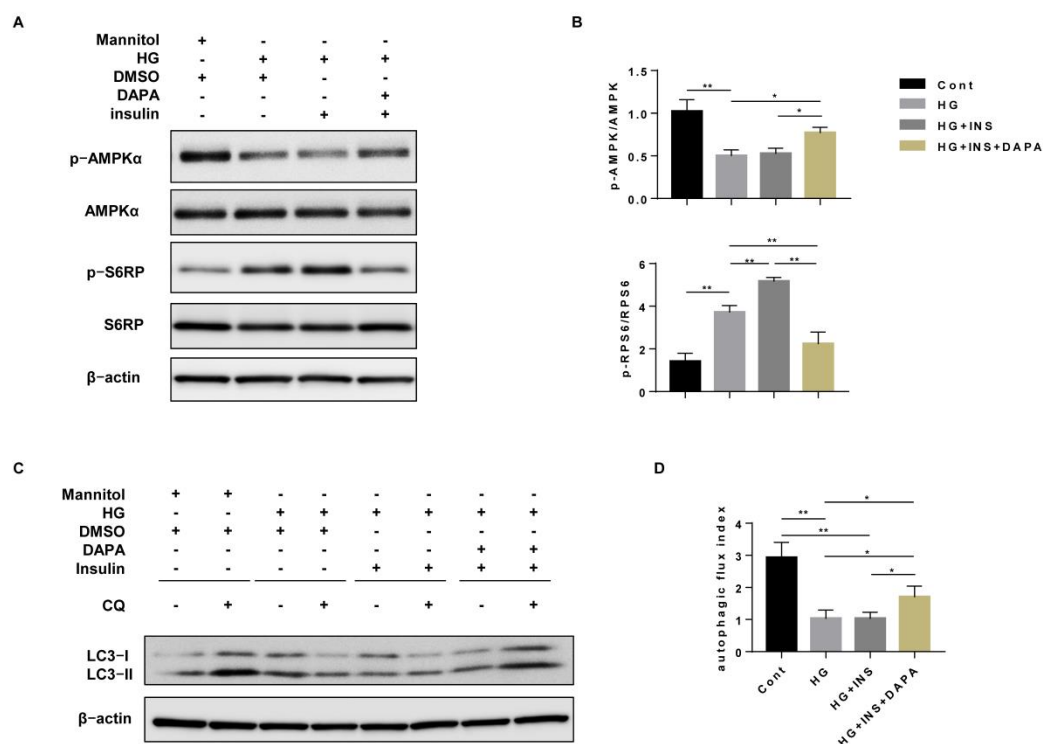


Figure S4. Dapagliflozin increased the activity of AMPK and autophagic flux in insulin-treated HK-2 cells. **(A)** HK-2 cells were treated with Cont, HG (30 mM), HG (30 mM) with insulin (100 nM), HG (30 mM) with insulin (100 nM) plus dapagliflozin (20 μ M) for 48 h, and p-AMPK, p-S6RP were assessed by western blotting. **(B)** Densitometric evaluation of (A) (n=3). **(C)** HK-2 cells were treated with Cont, HG (30 mM), HG (30 mM) with insulin (100 nM), HG (30 mM) with insulin (100 nM) plus dapagliflozin (20 μ M) for 48 h and chloroquine (50 μ M) or vehicle for another 1 h. Autophagic flux was determined with the LC3-II ratio in the presence and absence of chloroquine. **(D)** Densitometric evaluation of (C) (n=5). All data represent the means \pm standard deviation (SD). *p<0.05 vs. the indicated group, **p<0.01 vs. the indicated group. Cont, control; HG, high glucose; DAPA, dapagliflozin; INS, insulin; CQ, chloroquine.

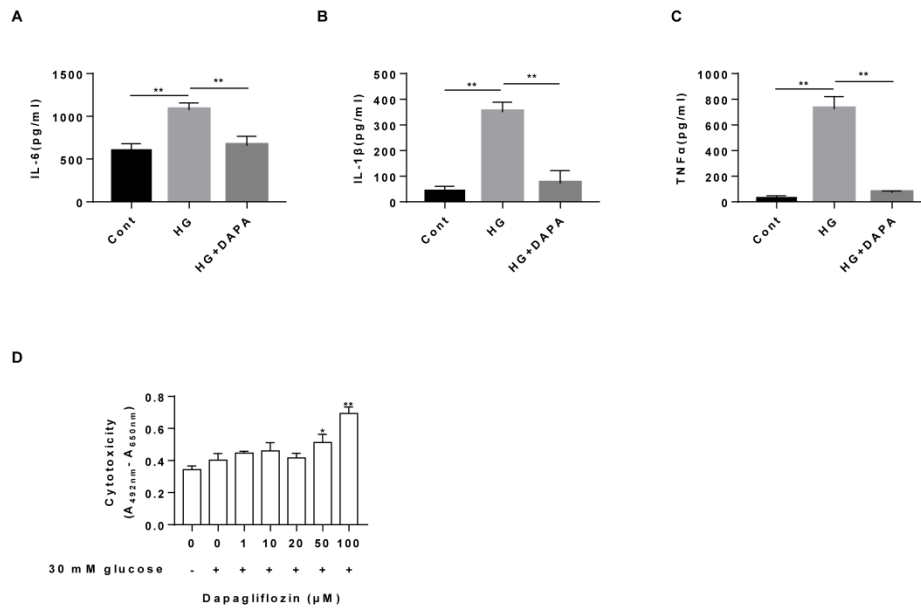


Figure S5. Dapagliflozin suppressed HG-induced pro-inflammatory cytokines in cultured medium. HK-2 cells were incubated with Cont, HG (30 mM), or HG (30 mM) dapagliflozin (20 μM) for 48 h, levels of pro-inflammatory cytokines in cultured medium including (A) IL-6, (B) IL-1β and (C) TNFα were measured by ELISA (n=5). *p<0.05 vs. the indicated group, **p<0.01 vs. the indicated group. (D) Cytotoxicity of dapagliflozin on HG-treated HK-2 cells was assayed by LDH cytotoxicity detection kit after incubation with different concentrations of dapagliflozin (0-100 μM) for 48 h (n=3). *p<0.05 vs. 30 mM glucose group, **p<0.01 vs. 30 mM glucose group. Cont, control; HG, high glucose; DAPA, dapagliflozin.