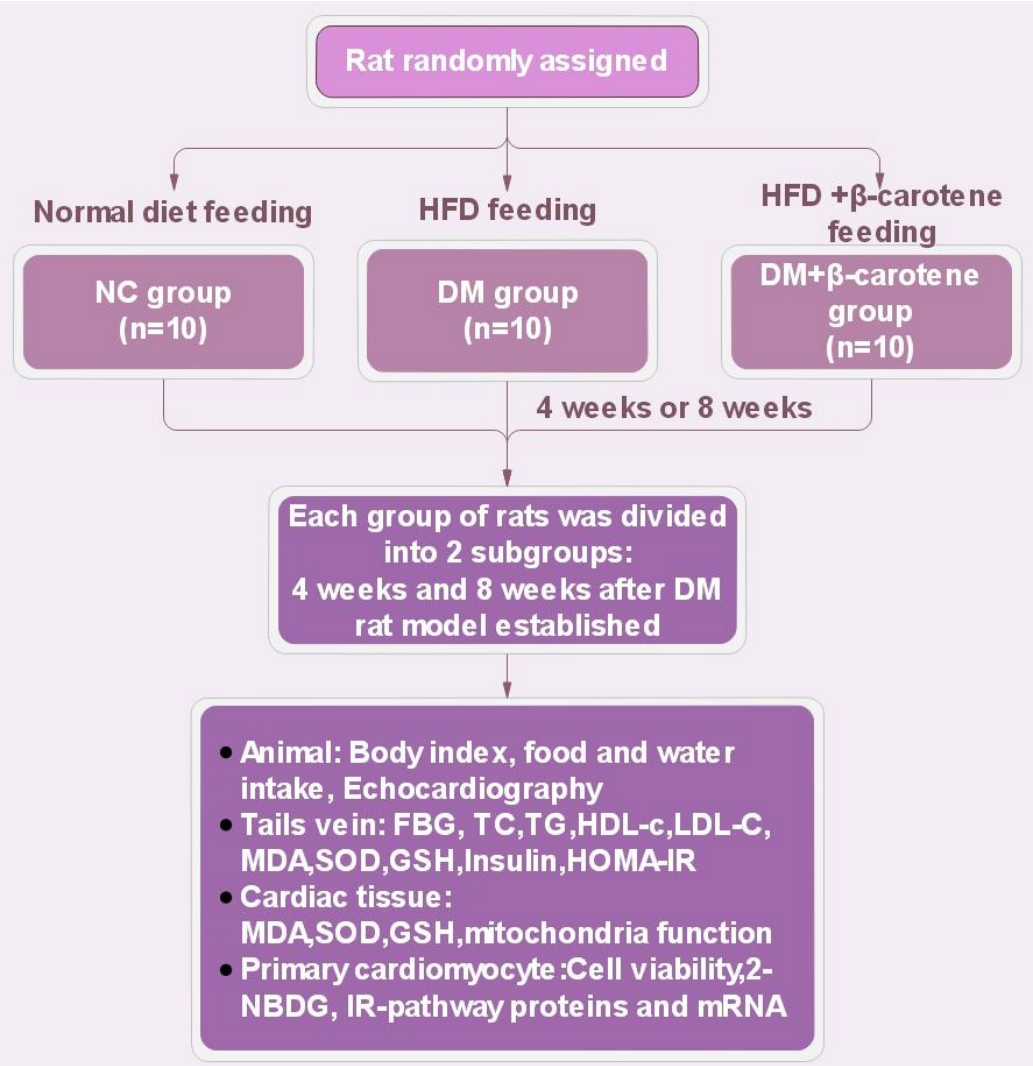

Figure1. Flow chart of the entire procedure of animal experiment

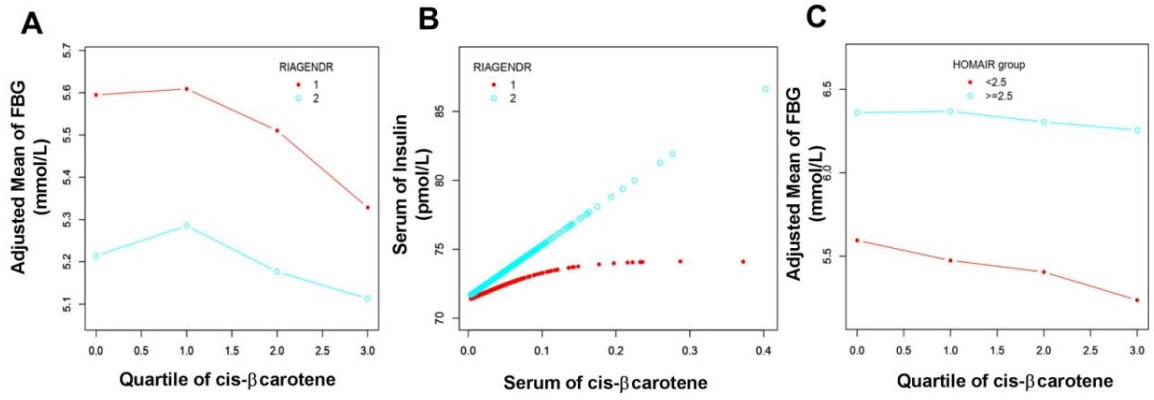

Figure 2. Line chart showing the positive effect of cis-β-carotene on glycemic control in Table 3

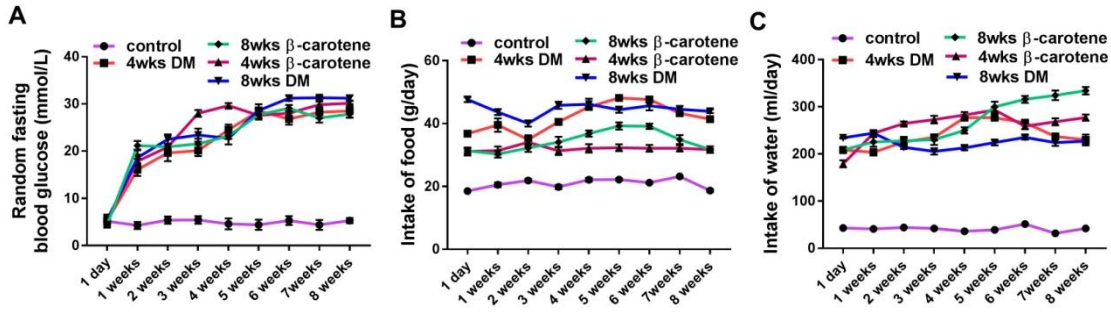

Figure 3. Blood glucose levels, food and water intake in rats.

(A). Blood glucose levels (mg/dl) obtained via tail vein puncture during the study period at 4- to 8-week intervals. (B) Food and (C)water intake in Diabetes(DM) and Diabetes+β-carotene groups.Values are given as the means ± S.E.M. n=10.

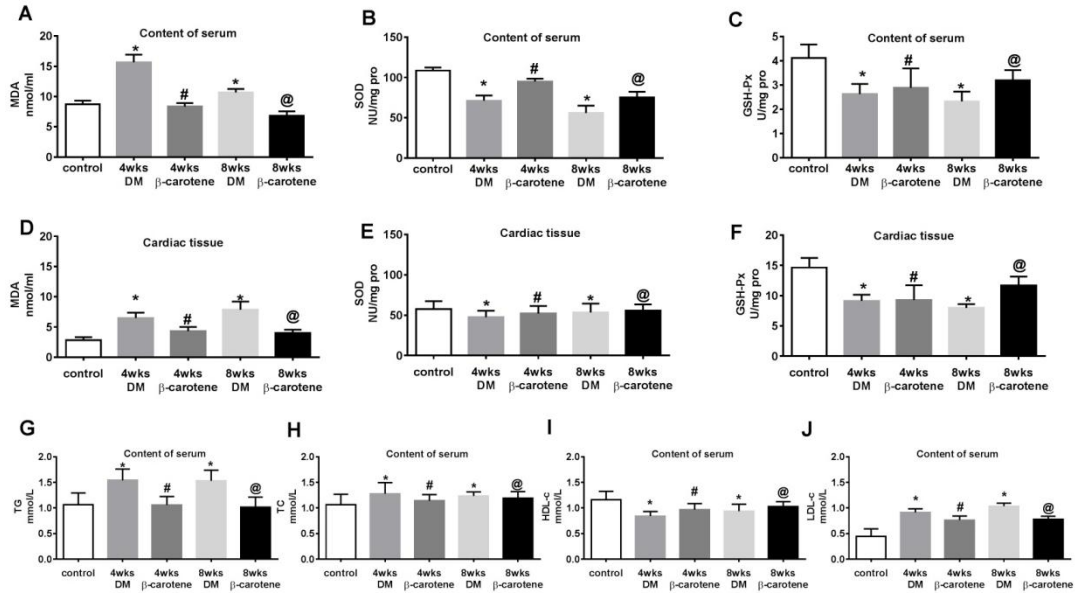

Figure 4.Effect of β-carotene on abnormal glucose and lipid metabolism, including: total triglycerides (TG) ,cholesterol (TC), HDL-cholesterol (HDL-c) and LDL-cholesterol levels (LDL-c).MDA content in serum(A) and cardiac tissue(D). SOD activity in serum(B) and cardiac tissue(E).GSH-px activity in serum(A) and cardiac tissue(D).Serum TG (G), TC (H) , HDL-c (I) and LDL-c (J) were measured throughout the study and average levels were calculated. **P*<0.05 vs. control group; #*P*<0.05 vs. 4 weeks DM group; @ *P*<0.05 vs. 8 weeks DM group, respectively (n=10).

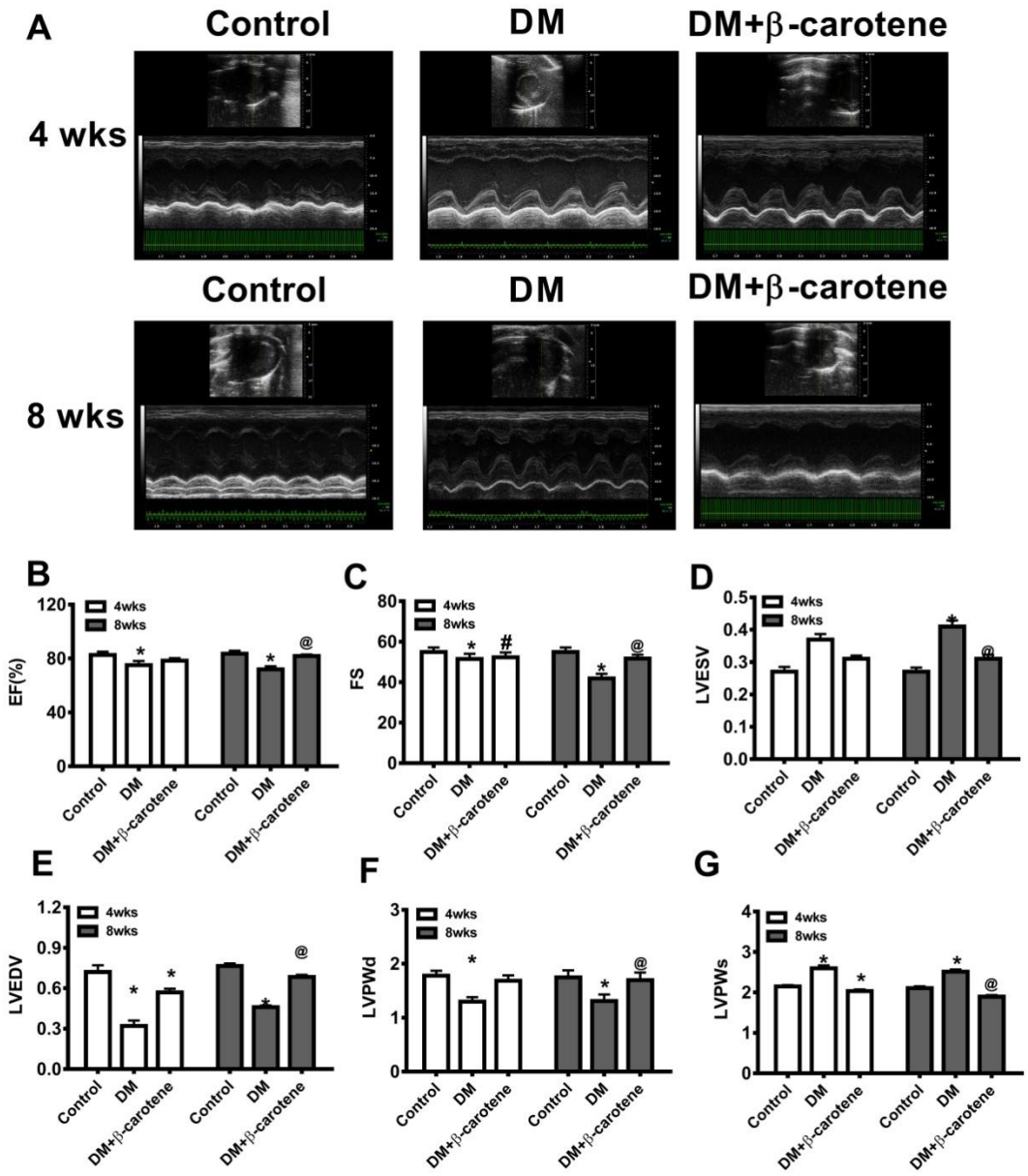

Figure 5. Cardiac dysfunction in diabetic rats. (A) Representative M-mode images of diabetic rats and controls. Serial changes in (B) ejection fraction (EF), (C) fractional shortening (FS), (D) LVESV, LV end-systolic volume; (E) LV end-diastolic volume (LVEDV);(F) LV posterior wall end diastole (LVPWd) and (G) LV posterior wall end systole (LVPWs) were assessed by echocardiography. *P<0.05 vs. control group; #P<0.05 vs. 4 weeks DM group; @ P<0.05vs.8 weeks DM group, respectively (n=10).

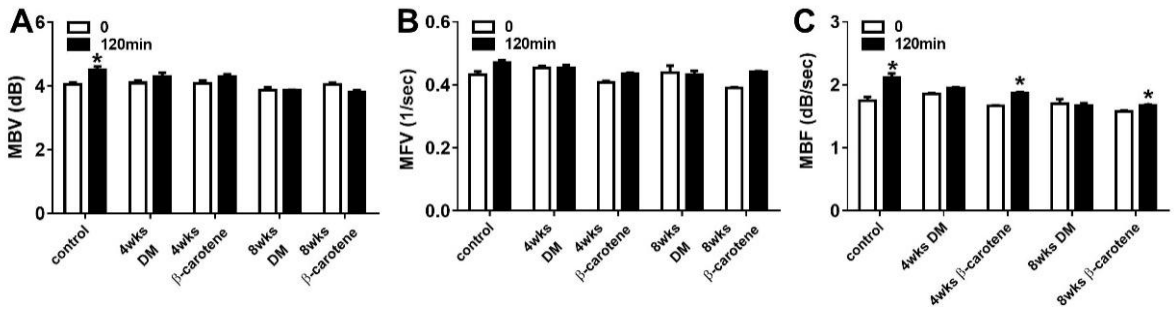

Figure 6. Changes of cardiac muscle microvascular parameters . Myocardial MBV (A), MFV (B), and MBF (C) at baseline (blank square) and at end of 120-min insulin infusion (black square). **P* <0.05 vs 0 min (n=10).

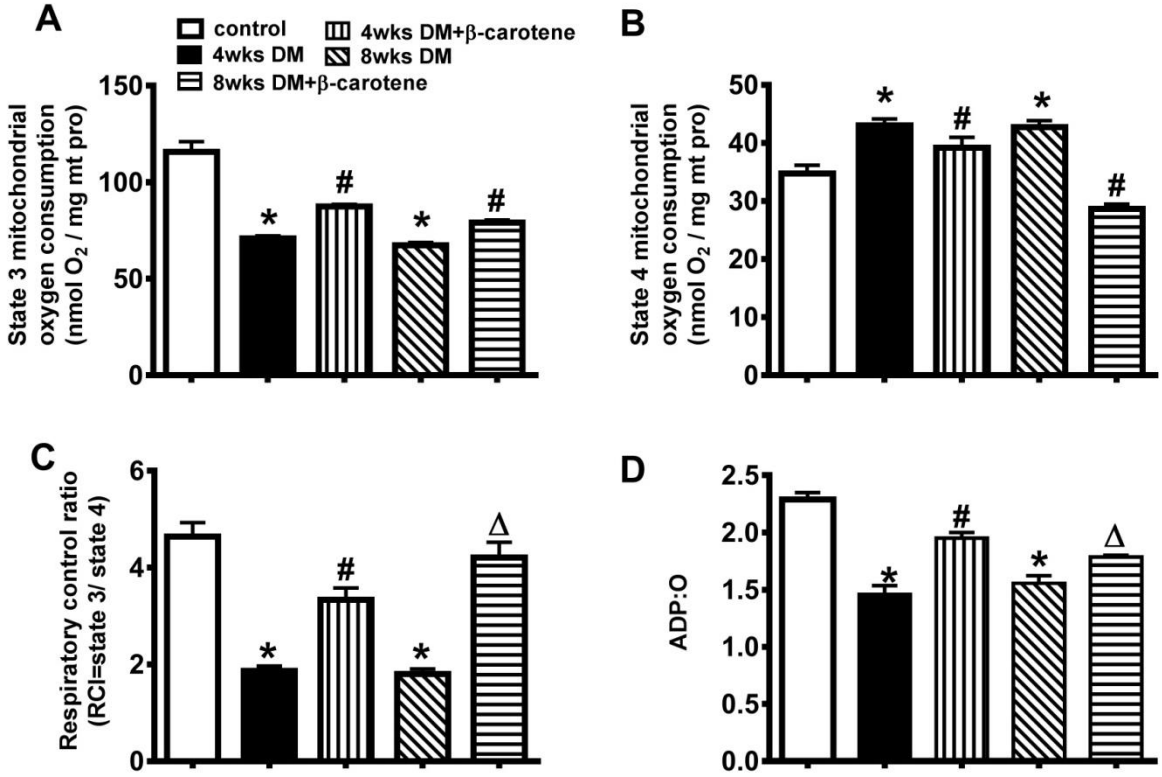

Figure 7. Effect of high glucose and β-carotene treatment in cardiac mitochondria respiratory chain parameters: states 3 (A) and 4 (B) of respiration, RCI(C) and ADP/O index (D). Data are the mean ± SEM of 4 animals from each condition studied. **P*<0.05 vs. control group; #*P*<0.05 vs. 4 weeks DM group; Δ *P*<0.05vs.8 weeks DM group, respectively (n=10).

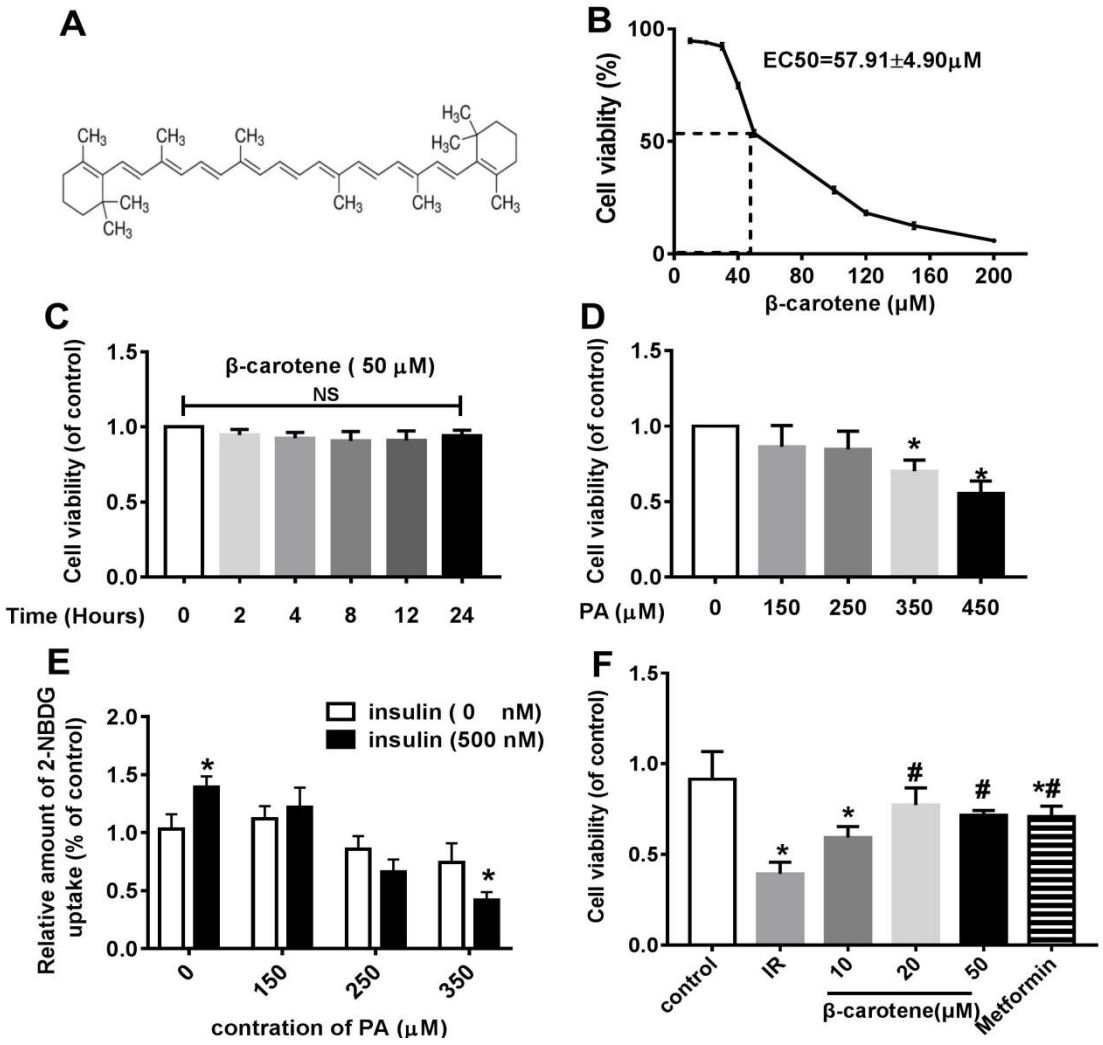

Figure 8. The bioactivity of β-carotene in primary cardiomyocytes. (A) Chemical structure of β-carotene. (B) Cardiomyocytes metabolic activity was analyzed by performing an MTT following treatment with β-carotene dose range (EC50, 57.91 ± 4.9 μM; n=4). (C) Quantification of primary cardiomyocytes viability following treatment with different time with β-carotene treatment. (D) Quantification of primary cardiomyocytes viability following treatment with increasing concentrations of palmitic acid (PA). (E) PA reduces 2-NBDG uptake of cardiomyocytes with or without insulin (500 nM). (F) Quantification of cardiomyocytes viability following different dose of β-carotene treatment following co-incubation with or without PA. Data are presented as the means ± SEM. *P<0.05 vs. control group; #P<0.05 vs.IR group, respectively (n=10).

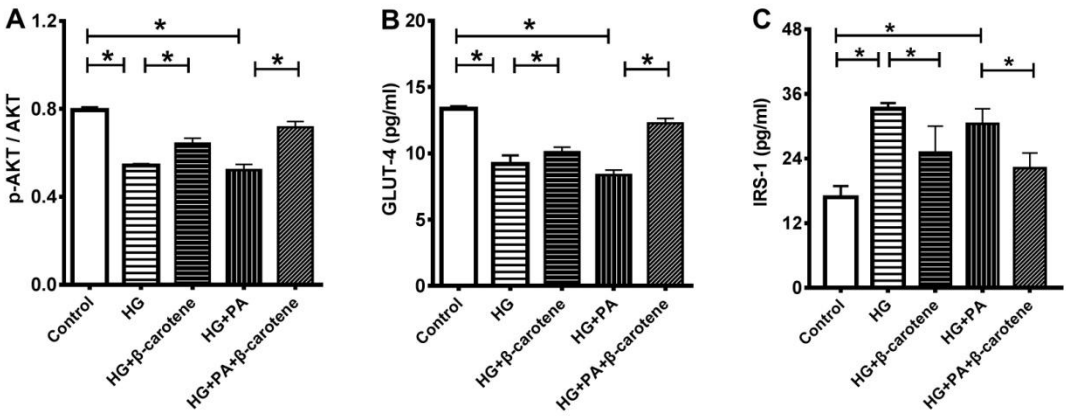

Figure 9. Effects of β-carotene on insulin signaling pathway in insulin-resistant cardiomyocytes induced by PA after 24 hrs treatment. (A) ELISA showing the p-AKT/AKT ratio in cardiomyocytes (B) ELISA showing the GLUT4 level in cardiomyocytes (C) ELISA showing the IRS-1 level in cardiomyocytes .*P<0.05 represents significant differences. Data represent the best of three separate experiments. Each ELISA value is an average of two measurements.

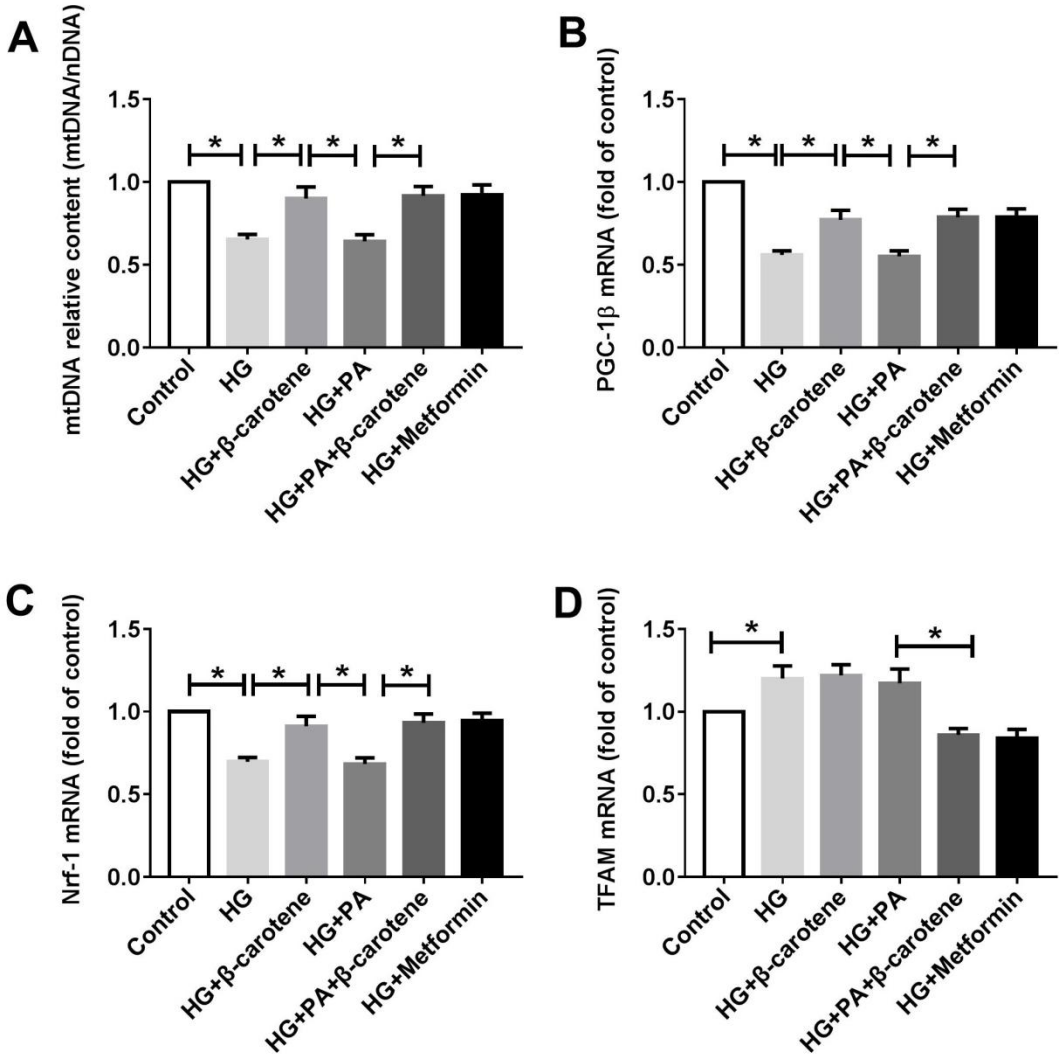

Figure 10. Effect of β-carotene on mitochondria biogenesis of insulin-resistant cardiomyocytes induced by PA after 24 h treatment.(A) Effect of β-carotene on the mtDNA copy number using PCR.(B-D)The relative mRNA levels PGC-1β ,Nrf-1 and TFAM were determined by quantitative real-time PCR assay and calculated by the mean value with the comparative Ct method . Cardiomyocytes were treated with 5.5 mM glucose (lane 1,control), 25 mM glucose (lane 2,HG), HG+50 μM β-carotene (lane 3, HG+β-carotene), HG+350 μM of PA (lane 4， HG+PA) and HG+350 μM of PA+50 μM β-carotene (lane 4, HG+PA+β-carotene)for 24 hrs. β-actin served as the loading control. *P<0.05 represents significant differences. Data represent the best of three separate experiments.