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Original Article

Propofol preconditioning alleviates myocardial ischemia-reperfusion injury in rats through the Wnt/β-catenin signaling pathway

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Abstract: Objective: Myocardial ischemia/reperfusion (I/R) injury seriously endangers the safety of patients. Therefore, this study aimed to explore the protective effect of propofol preconditioning and the mechanism of Wnt/β-catenin signaling pathway in myocardial I/R injury in rats. Methods: Forty male Sprague-Dawley (SD) rats were randomly allocated into 4 groups of 10 rats each: Sham group, IR group, P+IR group, K+IR group. Myocardial I/R rat models were established in the IR group, P+IR group and K+IR group. In the P+IR group, intravenous injection of propofol (6 mg/kg) was given via the tail vein 15 min before the modeling. In the K+IR group, Wnt inhibitor was injected 1 h before modeling. The coronary artery of rats in the sham operation group was not ligated. At the end of reperfusion, the rats in each group were sacrificed, and the serum was collected. The contents of serum interleukin 6 (IL-6), tumor necrosis factor (TNF-α), lactate dehydrogenase (LDH), creatine kinase (CK) were determined by enzyme-linked immunosorbent assay (ELISA). Additionally, the contents of myocardial malondialdehyde (MDA) and superoxide dismutase (SOD), which is representative of oxidative stress, in ground heart tissues were measured by ELISA. Moreover, the expression levels of Bcl-2, Bax, Wnt and β-catenin in the myocardium were detected by Western blot. Results: The contents of IL-6, TNF-α, LDH, CK, MDA and SOD as well as the expression levels of Wnt and β-catenin in the IR group increased significantly compared with those in the Sham group. While the above indexes in the P+IR group and K+IR group decreased significantly compared with those in the IR group (all $P<0.05$). Conclusion: Propofol alleviates myocardial I/R injury in rats by inhibiting the Wnt/β-catenin signaling pathway.

Keywords: Propofol, myocardial ischemia/reperfusion, Wnt/β-catenin, apoptosis

Introduction

With the increase of daily stress and the fast paced changes in lifestyle, ischemic cardiomyopathy is more and more common in young people. In surgery delayed recanalization of blood vessels triggers myocardial ischemia and hypoxia, and may even lead to permanent loss of local myocardial function. Therefore, the rapid recovery of myocardial blood flow is crucial to prognosis of the patients [1-3]. Exacerbated or even irreversible tissue damage caused by restoration of blood flow after ischemia is called ischemia-reperfusion (I/R) injury [4-6]. It has recently been reported that up to 20% of patients with myocardial ischemia suffer from I/R injury [7]. Therefore, reliving myocardial I/R injury with timely intervention can reduce the mortality of patients.

At present, thrombolysis, minimally invasive surgery, as well as stent implantation are commonly used to restore coronary blood flow after myocardial ischemia [8, 9]. However, the treatment of myocardial I/R injury after blood flow recovery still puzzles clinicians [10, 11]. Calcium overload and oxidative stress induced by myocardial I/R may lead to cardiomyocyte apoptosis or autophagic cell death, which is related to inflammation and cell death and cannot be reversed by general anti-inflammatory and anti-apoptotic methods, thus bringing difficulties to clinical treatment [12-14]. Therefore, exploring the underlying pathogenesis may provide better clinical guidance.

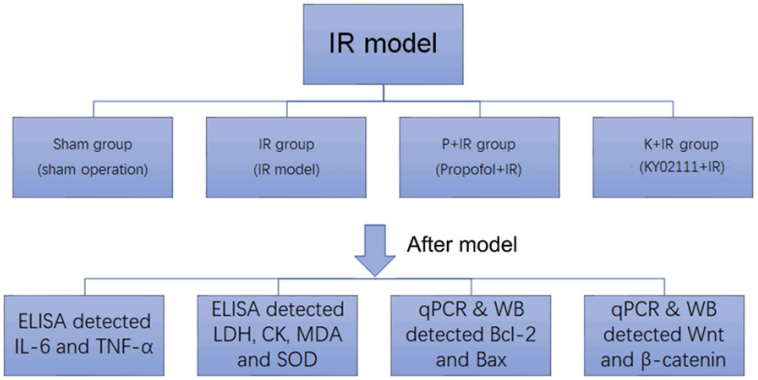

Figure 1. Technical diagram. IL-6: interleukin 6, TNF-α: tumor necrosis factor, LDH: lactate dehydrogenase, CK: creatine kinase, MDA: myocardial malondialdehyde, SOD: superoxide dismutase, Bax: pro-apoptotic protein, Bcl-2: anti-apoptotic protein; Sham: control group, IR: myocardial ischemia-reperfusion injury, P: propofol, K: Wnt inhibitor KY02111; EILSA: enzyme linked immunosorbent assay, qPCR: fluorescence quantitative PCR.

Propofol, a commonly used sedative drug, has the functions of inhibiting the release of inflammatory factors and anti-oxygenation [15]. It also inhibits Na^+/K^+-ATPase in cardiomyocyte membranes and regulates the release of chemokines in myocardial I/R injury; so as to reduce inflammatory cell infiltration and inflammatory factor release [16]. A study has found that propofol reduces lung injury induced by intestinal I/R injury in pigs, possibly by inhibiting neutrophil respiratory bursts and reducing cell apoptosis [17].

The Wnt/β-catenin signaling pathway is involved in regulating cell growth and inhibiting inflammation and cell death [18]. Activation of Wnt receptor complex triggers dissociation of multifunctional kinase GSK-3β from a regulatory APC/Axin/GSK-3β complex. When Wnt signaling is lost (OFF state), β-catenin, an integral E-cadherin act as an intercellular adhesion adapter protein and a transcriptional co-regulator; is targeted by coordinated phosphorylation by CK1 and APC/Axin/GSK-3β-complex, leading to its ubiquitination and proteasomal degradation by β-TrCP/Skp pathway. In the presence of the Wnt ligand (ON state), co-receptor LRP5/6 is brought into the complex with Wnt-bound Frizzled. Stable β-catenin is translocated to the nucleus through Rac1 and other factors, where it binds to LEF/TCF transcription factors, displacing co-repressor and recruiting additional co-activator to Wnt target genes. In addition, β-catenin cooperates with other transcription factors to regulate specific targets. Researchers have found that point mutation of β-catenin in human tumors prevents phosphorylation of GSK-3β and leads to its abnormal aggregation. There is also evidence that Wnt signaling promotes nuclear accumulation of other transcriptional regulators associated with cancer. Moreover, GSK-3β participates in glucose metabolism and other signaling pathways, which indicates that its inhibition is related to diabetes and neurodegenerative diseases. However, whether Wnt/β-catenin signaling pathway is involved in myocardial I/R injury still remains unknown.

In the early stages of this experiment, it was found that Wnt was significantly increased in cytoplasmic proteins in myocardial I/R injury. Therefore, this study explored the mechanism of propofol regulating the Wnt/β-catenin pathway to prevent myocardial I/R injury in rats.

Materials and methods

Animals and groups

Forty male Sprague-Dawley (SD) rats were randomly allocated into 4 groups of 10 rats each: Sham group, IR group, P+IR group and K+IR group. Myocardial I/R rat models were established in the IR group, P+IR group and K+IR group. In the P+IR group, intravenous injection of propofol (6 mg/kg) (Shanghai Jichun Industrial Co., Ltd., item number: SJ-JC14680) was administered via the tail vein 15 min before the modeling. In the K+IR group, Wnt inhibitor (Wuhan Chundu Biology Co., Ltd., KY02111, item number: YZJ-23207) was injected 1 h before modeling. The coronary artery of rats in the sham operation group was not ligated (**Figure 1**).

Rat model of myocardial I/R Injury

Rats were intraperitoneally anesthetized with 1% pentobarbital sodium (30 mL/kg). After tracheal intubation, the catheter was fixed with

Table 1. Primers of specific genes

Genes	Primers
GAPDH	R: 5'-GGTATGACAACGAATTTGGC-3'
	F: 5'-GAGCACAGGGTACTTTATTG-3'
Bax	R: 5'-TCCACCAAGAAGCTGAGCGAG-3'
	F: 5'-GTCCAGCCCATGATGGTTCT-3'
Bcl-2	R: 5'-TTCTTTGAGTTCGGTGGGGTC-3'
	F: 5'-TGCATATTTGTTTGGGGCAGG-3'
Wnt3a	R: 5'-CATCGCCAGTCACATGCACCT-3'
	F: 5'-CGTCTATGCCATGCGAGCTCA-3'
β-catenin	R: 5'-CAAACTGCTAAATGACGAGG-3'
	F: 5'-GGGAAAGGTTGTGTAGGGTC-3'

surgical suture and connected to a rodent ventilator (animal ventilator, WRD Life Science, Shenzhen, China), with a cardiac and respiratory rate of 90 beats/min and a tidal volume of 1 mL. In the sham group the coronary artery was not ligated after thoracotomy. After successful anesthesia, the rats in the IR group were fixed on the plate in a natural supine position. Tracheotomy and tracheal intubation were performed, then the ventilator was connected. Afterwards, an animal electrocardiogram (ECG) instrument was connected to the limb of the rat to monitor ECG changes during the surgery. Next, an incision was made in the left chest from the third to fourth rib (intercostal space). The wound was fixed and the heart was exposed. The left coronary artery was ligated approximately 2.5 mm below the left atrial appendage. Afterwards, the myocardium in left ventricular anterior wall could be seen turning white, the heart beats became weak, moreover, the electrocardiogram showed that the ST-T segment was elevated and the extensive malformation lasted for more than 30 min, then the ligation was loosened and the blood was reperfusion for 2 h. Rats were placed in a clean and warm place to collect sers and heart tissues. The collected tissues were stained by Nagar-Olsen assay. Normal myocardium showed blue, ischemic myocardium showed red, and infarcted myocardium showed white, which was used to judge the model establishment.

ELISA

The blood was collected by right thoracic puncture. The collected venous blood was placed at room temperature for 2 h and centrifuged at 3,000 g and 4°C for 15 min to obtain the sera. The sera were detected by interleukin 6 (IL-6), tumor necrosis factor (TNF-α), lactate dehydrogenase (LDH), creatine kinase (CK), myocardial malondialdehyde (MDA) and superoxide dismutase (SOD) Quantikin ELISA kits from R&D Company.

Quantitative real time polymerase chain reaction (qRT-PCR)

Total RNAs was extracted from rat tissues by Trizol kit (Invitrogen, USA), and cDNAs were as synthesized using a reverse transcription kit (11939823001, Merck, USA). qRT-PCR system (7500 software v2.0.6) (10 μL): 0.5 μL of forward primer, 0.5 μL of reverse primer, 2 μL of cDNA, 3 μL of ddH_2O, 5 μL of (2×) SYBR® Premix Ex TaqTM II. Reaction conditions: predenaturation at 95°C for 2 min, denaturation at 95°C for 15 s, annealing at 55°C for 35 s, for a total of 40 cycles, then extension at 72°C for 5 min. Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) was used as an internal reference. Primers shown in **Table 1** were synthesized by Sangon Bioengineering Co., Ltd. The expressions of the products were calculated by $2^{-\Delta\Delta Ct}$.

Western blot

The rats were sacrificed by right thoracic puncture. The thoracic cavity was opened, the inferior vena cava and thoracic aorta were separated and severed respectively to obtain the cardiac tissue. The tissue was cut and placed in eppendorf (EP) tubes (2 mL) frozen at -80°C in advance, homogenized (65 HZ, 60 s, 4°C) twice, and centrifuged at 12,000 rpm and 4°C for 5 min. Afterwards, the supernatant was collected and placed in 1.5 mL centrifuge tubes. The concentration of proteins was determined using the bicinchoninic acid method. The proteins were separated by sodium dodecylsulphate polyacrylamide gel electrophoresis (SDS-PAGE) and then transferred to a membrane. Then the membrane was blocked, then incubated at 4°C for 12 h with rabbit anti-rat β-actin (1:2,000), rabbit anti-rat Bax (1:1,000, Cell Singling Technology, USA), Bcl-2 (1:1,000, Cell Singling Technology, USA), Wnt3a (1:1,000, Cell Singling Technology, USA), β-catenin (1:1,000, Cell Singling Technology, USA) primary antibodies. Next, horseradish peroxidase (HRP)-labeled goat anti-rabbit secondary antibody (1:1,000, Cell Singling

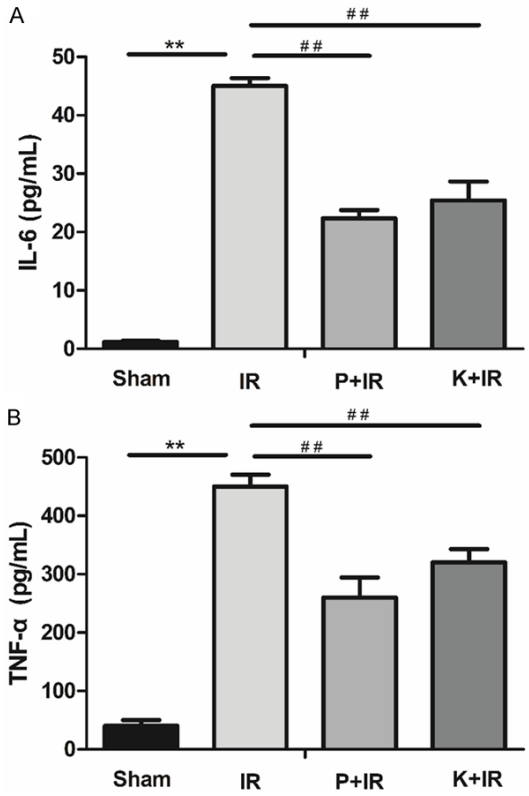

Figure 2. The expression changes of *inflammatory factors in rats.* A. The expression of IL-6; B. The expression of TNF-α; Sham: control group, IR: myocardial ischemia-reperfusion injury, P+IR: propofol and myocardial ischemia-reperfusion injury group, K+IR: Wnt inhibitor and myocardial ischemia-reperfusion injury group; **P<0.01 compare to sham; ##P<0.01 compare to IR.

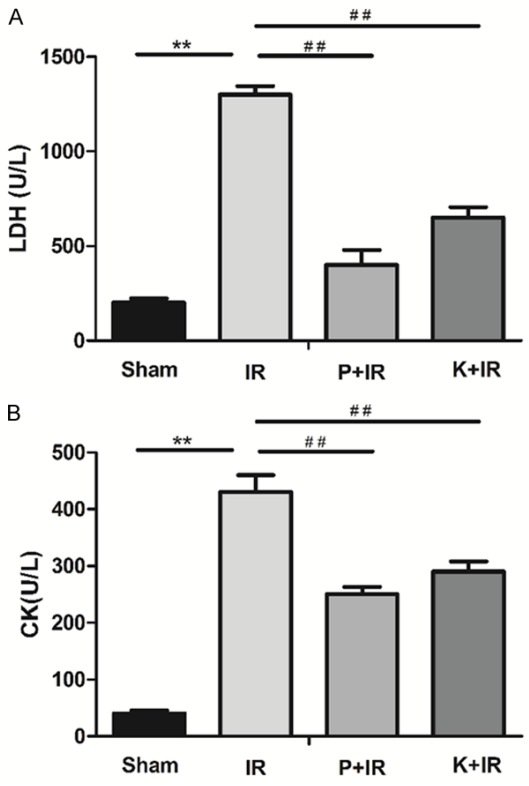

Figure 3. The expression changes of myocardial enzyme release in rats. A. The expression changes of lactate dehydrogenase (LDH); B. The expression changes of creatine kinase (CK); Sham: control group, IR: myocardial ischemia-reperfusion injury, P+IR: propofol and myocardial ischemia-reperfusion injury group, K+IR: Wnt inhibitor and myocardial ischemia-reperfusion injury group; **P<0.01 compare to sham; ##P<0.01 compare to IR.

Technology, USA) was added to the membrane. Exposure and development were performed with reference to the enhanced chemiluminescence (ECL) kit (hypersensitive ECL kit, Beyotime, China). The gray value of each band was analyzed using ImageJ software.

Statistical analysis

The data in this study were analyzed by SPSS 22.0. All statistical data were measurement data. First, the Kolmogorov-Smirnov test was conducted for normality test. Data conforming to normal distribution were expressed by the mean ± standard deviation ($\bar{x}$ ± sd). Multi-group comparison was conducted with one-way analysis of variance, and following pair-wise comparison was conducted with Student-Newman-Keuls test. The difference was statistically significant with P<0.05.

Results

Propofol alleviates inflammation

The levels of serum IL-6 and TNF-α in the IR group increased significantly compared with those in the sham group (P<0.01). While the levels in the P+IR group and K+IR group decreased significantly compared with those in the IR group (P<0.01). See **Figure 2**.

Propofol reduces myocardial enzyme release

The contents of serum LDH and CK in the IR group increased significantly compared with those in the sham group (P<0.01). While the contents in the P+IR group and K+IR group decreased significantly compared with those in the IR group (P<0.01). See **Figure 3**.

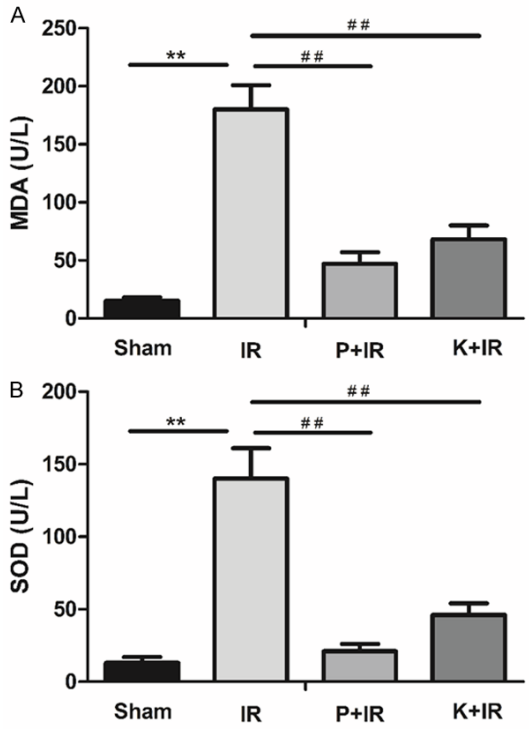

Figure 4. The expression changes of myocardial oxidative stress in rats. A. The expression changes of myocardial malondialdehyde (MDA); B. The expression changes of superoxide dismutase (SOD); Sham: control group, IR: myocardial ischemia-reperfusion injury, P+IR: propofol and myocardial ischemia-reperfusion injury group, K+IR: Wnt inhibitor and myocardial ischemia-reperfusion injury group; **P<0.01 compare to sham; $^{\#\#}$P<0.01 compare to IR.

Propofol relieves oxidative stress

The contents of MDA and SOD in the IR group increased significantly compared with those in the sham group (P<0.01). While the contents in the P+IR group and K+IR group decreased significantly compared with those in the IR group (P<0.01). See **Figure 4**.

Propofol decreases cardiomyocyte apoptosis

The mRNA and protein expression of pro-apoptotic protein Bax in the IR group increased significantly compared with that in the sham group, while the expression of anti-apoptotic protein Bcl-2 decreased significantly (P<0.01). The mRNA and protein expression of Bax in P+IR group and K+IR group decreased significantly compared with that in the IR group, while the expression of Bcl-2 increased significantly (P<0.01). Therefore, propofol could inhibit cardiomyocyte apoptosis in rats with myocardial I/R injury (**Figure 5**).

Propofol reduces myocardial I/R injury in rats by inhibiting the Wnt/β-catenin signaling pathway

The mRNA and protein expression of Wnt/β-catenin in the IR group rats increased significantly compared with that in the sham group (P<0.01). While the expression in the P+IR group and K+IR group decreased significantly compared with that in the IR group (P<0.01). See **Figure 6**.

Discussion

Myocardial infarction is a cardiovascular disease with high mortality and disability, seriously affecting the quality of life of patients [19]. The severity and prognosis are closely related to infarct size and cardiomyocyte apoptosis. Therefore, the current clinical treatment of myocardial infarction mainly focuses on narrowing infarct size and anti-cardiomyocyte apoptosis [20, 21].

In this study, the myocardial I/R models were prepared in a traditional way, which better simulated the myocardial infarction and reperfusion process and was more simple, economical, and practical, and easier to be replicated. Myocardial I/R in tissues leads to hypoperfusion, increased anaerobic glycolysis, as well as decreased adenosine triphosphate (ATP). The main function of Na^+-K^+-ATPase is to pump intracellular sodium ions (Na^+) out and extracellular potassium ions (K^+) within the cell. When myocardial I/R occurs, Na^+-K^+-ATPase activity significantly decreases, leading to insufficient cardiomyocyte energy, significantly increased inflammation and inflammatory factor release [22, 23]. Our study found that the expression of inflammatory factors in serum of I/R rats was significantly increased, indicating that the I/R process was related to inflammation. The possible mechanism is that myocardial ischemia causes declined blood flow to the myocardium, inducing local hypoxia, cardiomyocyte necrosis and release of inflammatory factors into the blood; eventually resulting in significant increase of IL-6 and TNF-α in serum. Propofol preconditioning has effects of protecting myocardial membranes, reducing localized cell death, decreasing inflammatory factor release

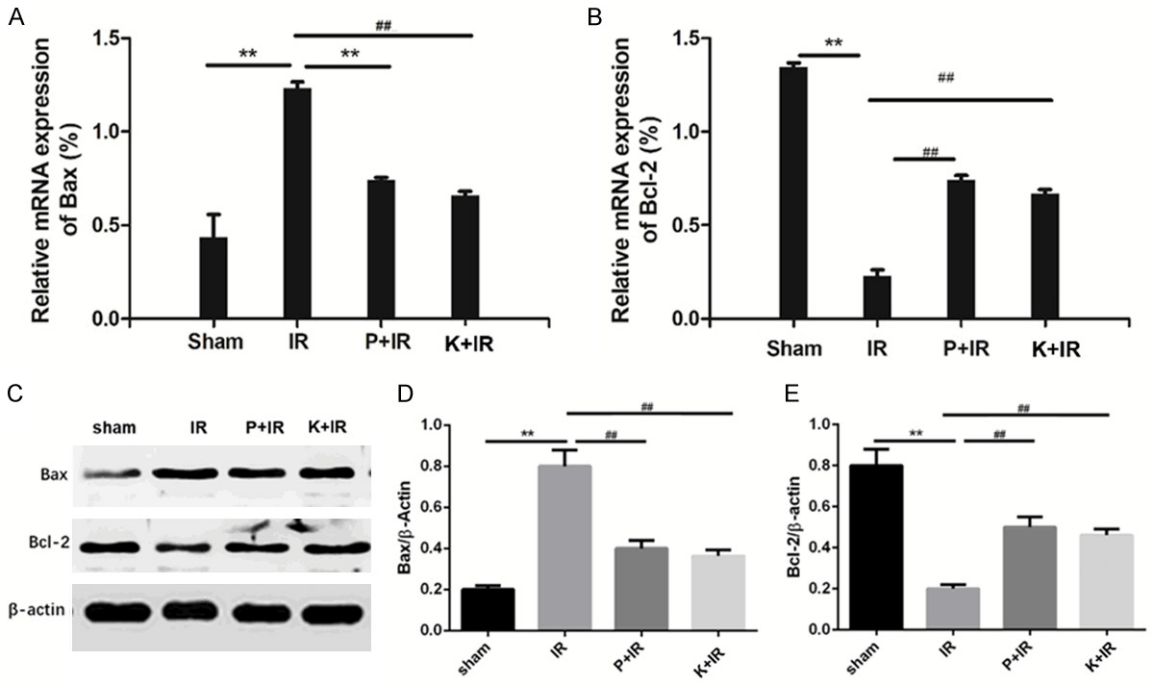

Figure 5. The expression changes of myocardial apoptosis relative protein and mRNA in rats. A. The relative mRNA expression changes of pro-apoptotic protein (Bax); B. The relative mRNA expression changes of anti-apoptotic protein (Bcl-2); C. Western blot results of Bax and Bcl-2; D. Quantitative column chart of relative expression level of Bax; E. Quantitative column chart of relative expression level of Bcl-2; Sham: control group, IR: myocardial ischemia-reperfusion injury, P+IR: propofol and myocardial ischemia-reperfusion injury group, K+IR: Wnt inhibitor and myocardial ischemia-reperfusion injury group; **P<0.01 compare to sham; ##P<0.01 compare to IR.

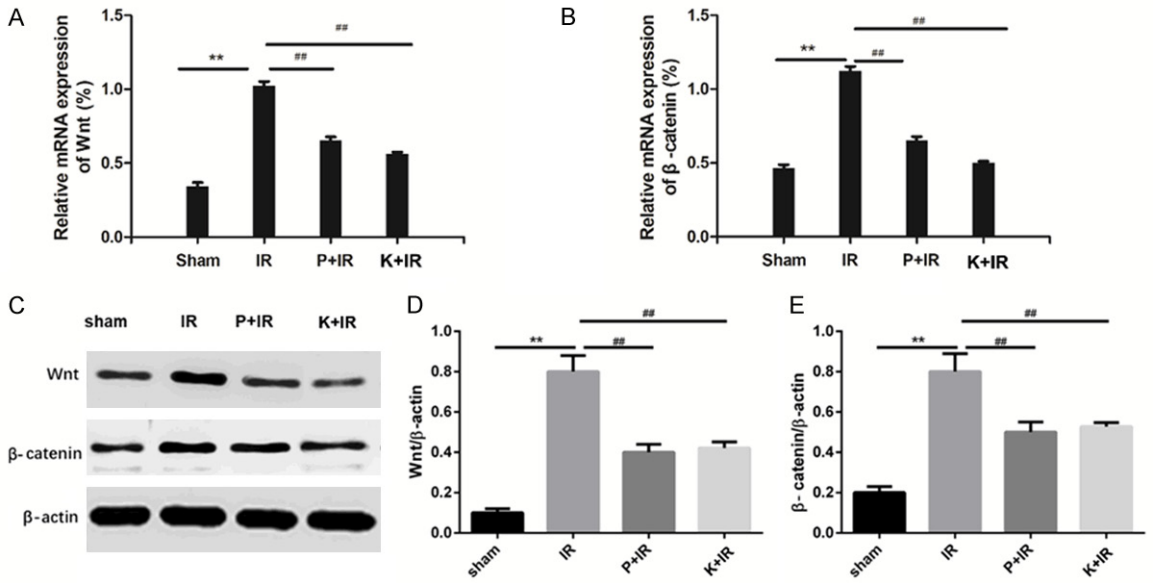

Figure 6. The mRNA and protein expression changes of myocardial Wnt/β-catenin in rats. A. The relative mRNA expression changes of Wnt; B. The relative mRNA expression changes of β-catenin; C. Western blot results of Wnt/β-catenin; D. Quantitative column chart of relative expression level of Wnt; E. Quantitative column chart of relative expression level of β-catenin; Sham: control group, IR: myocardial ischemia-reperfusion injury, P+IR: propofol and myocardial ischemia-reperfusion injury group, K+IR: Wnt inhibitor and myocardial ischemia-reperfusion injury group; **P<0.01 compare to sham; ##P<0.01.

and remarkably relieving inflammation. Yu revealed that propofol could significantly reduce the release of systemic inflammatory factors in liver injury induced by I/R [24, 25].

LDH and CK are markers of myocardial injury reflecting the degree of cardiomyocyte damage [26, 27]. This study found that the contents of serum LDH, CK, MDA and SOD increased significantly after I/R. Propofol preconditioning reduced myocardial injury and decreased the levels of above kinases. The possible reason is that propofol maintains stability and reduces the necrosis of cardiomyocytes by inhibiting the formation of oxides. RH found that propofol could deliver hydrogen to scavenge free radicals and protect cell stability in pulmonary tissues [21].

Myocardial I/R injury occurs in patients with acute myocardial infarction who have received thrombolysis, coronary angioplasty, cardiopulmonary bypass and cardiopulmonary resuscitation [22]. A study found that propofol relieved myocardial I/R injury, but the mechanism remained unclear [23]. Wnt protein is a highly conserved secreted protein that is rich in cysteine. Activated Wnt/β-catenin binds to the receptor family on the surface of cardiomyocytes, causing β-catenin to accumulate in the cytoplasm and then enter the nucleus, thus activating transcription factor TCF-LET to regulate the expression of target genes and inhibit cardiomyocyte apoptosis [18]. Wnt/β-catenin signaling pathway was reported to be involved in cell proliferation, differentiation and death. Yang et al suggested that the expression of β-catenin protein was significantly reduced in acute pulmonary injury, and significantly elevated when the disease was alleviated by drugs [19]. Our study also showed that the pro-apoptotic protein Bax increased and the anti-apoptotic protein Bcl-2 decreased significantly in the IR group. While Bax decreased and Bcl-2 increased significantly in the P+IR group compared with those in the IR group. The results indicated that propofol could inhibit cardiomyocyte apoptosis in myocardial I/R injury possibly by inhibiting the Wnt/β-catenin signaling pathway.

There are several limitations in this study. First, the mechanism of propofol on myocardial protection in rats with myocardial infarction has been preliminarily discussed, other possible regulatory approaches have not been described in detail. Second, other factors, such as hypoxia inducible factor-1α and NF-κB, may play a role in cardiomyocyte apoptosis during the occurrence and development of myocardial infarction. Third, the possible effects of propofol on cardiomyocyte apoptosis have not been elaborated. Therefore, the mechanism of propofol on myocardial protection will be further discussed.

In summary, propofol alleviates myocardial I/R injury in rats by inhibiting the Wnt/β-catenin signaling pathway.

Disclosure of conflict of interest

None.

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References

[1] Wilson RF. Ischemic cardiomyopathy. Am Heart J 2017; 119-133.

[2] Yokoshiki H, Mitsuyama H, Watanabe M, Mitsuhashi T and Shimizu A. Cardiac resynchronization therapy in ischemic and non-ischemic cardiomyopathy. J Arrhythm 2017; 33: 410-416.

[3] George J, Barsheshet A, Moss AJ, Martin D, Ouellet G, McNitt S and Goldenberg I. Effectiveness of cardiac resynchronization therapy in diabetic patients with ischemic and nonischemic cardiomyopathy. Ann Noninvasive Electrocardiol 2012; 17: 14-21.

[4] Zhao D, Yang J and Yang L. Insights for oxidative stress and mTOR signaling in myocardial ischemia/reperfusion injury under diabetes. Oxid Med Cell Longev 2017; 2017: 6437467.

[5] Fang SJ, Li PY, Wang CM, Xin Y, Lu WW, Zhang XX, Zuo S, Ma CS, Tang CS, Nie SP and Qi YF. Inhibition of endoplasmic reticulum stress by neuregulin-1 protects against myocardial ischemia/reperfusion injury. Peptides 2017; 88: 196-207.

[6] Chen Y, Ba L, Huang W, Liu Y, Pan H, Mingyao E, Shi P, Wang Y, Li S, Qi H, Sun H and Cao Y. Role of carvacrol in cardioprotection against myocardial ischemia/reperfusion injury in rats through activation of MAPK/ERK and Akt/eNOS signaling pathways. Eur J Pharmacol 2017; 796: 90-100.

[7] Yu L, Li S, Tang X, Li Z, Zhang J, Xue X, Han J, Liu Y, Zhang Y, Zhang Y, Xu Y, Yang Y and Wang H. Diallyl trisulfide ameliorates myocardial ischemia-reperfusion injury by reducing oxidative stress and endoplasmic reticulum stress-mediated apoptosis in type 1 diabetic rats: role of SIRT1 activation. Apoptosis 2017; 22: 942-954.

[8] Jayachandra A and Aggarwal V. To evaluate the change in brain natriuretic peptide levels in outpatients with ischemic cardiomyopathy and its association with functional capacity: a pilot study. J Clin Diagn Res 2017; 11: OC26-OC28.

[9] Wang L, Lu MJ, Feng L, Wang J, Fang W, He ZX, Dou KF, Zhao SH and Yang MF. Relationship of myocardial hibernation, scar, and angiographic collateral flow in ischemic cardiomyopathy with coronary chronic total occlusion. J Nucl Cardiol 2019; 26: 1720-1730.

[10] Chen H, Liu S, Liu X, Yang J, Wang F, Cong X and Chen X. Lysophosphatidic acid pretreatment attenuates myocardial ischemia/reperfusion injury in the immature hearts of rats. Front Physiol 2017; 8: 153.

[11] Yu Z, Wang S, Zhang X, Li Y, Zhao Q and Liu T. Pterostilbene protects against myocardial ischemia/reperfusion injury via suppressing oxidative/nitrative stress and inflammatory response. Int Immunopharmacol 2017; 43: 7-15.

[12] Li JP, Guo LL, Chen Z, Wang R and Wang J. Relationship between calcium overload and myocardial ischemia reperfusion injury and intervention strategy of Chinese herbal medicine. Zhongguo Zhong Yao Za Zhi 2016; 41: 2168-2173.

[13] Qin CY, Zhang HW, Gu J, Xu F, Liang HM, Fan KJ, Shen JY, Xiao ZH, Zhang EY and Hu J. Mitochondrial DNA-induced inflammatory damage contributes to myocardial ischemia reperfusion injury in rats: cardioprotective role of epigallocatechin. Mol Med Rep 2017; 16: 7569-7576.

[14] Zhai M, Li B, Duan W, Jing L, Zhang B, Zhang M, Yu L, Liu Z, Yu B, Ren K, Gao E, Yang Y, Liang H, Jin Z and Yu S. Melatonin ameliorates myocardial ischemia reperfusion injury through SIRT3-dependent regulation of oxidative stress and apoptosis. J Pineal Res 2017; 63.

[15] Jin YC, Kim W, Ha YM, Shin IW, Sohn JT, Kim HJ, Seo HG, Lee JH and Chang KC. Propofol limits rat myocardial ischemia and reperfusion injury with an associated reduction in apoptotic cell death in vivo. Vascul Pharmacol 2009; 50: 71-77.

[16] Li X, Yao L, Liang Q, Qu H and Cai H. Propofol protects hippocampal neurons from hypoxia-reoxygenation injury by decreasing calcineurin-induced calcium overload and activating YAP signaling. Oxid Med Cell Longev 2018; 2018: 1725191.

[17] Yang N, Liang Y, Yang P, Yang T and Jiang L. Propofol inhibits lung cancer cell viability and induces cell apoptosis by upregulating microRNA-486 expression. Braz J Med Biol Res 2017; 50: e5794.

[18] Nusse R and Clevers H. Wnt/beta-catenin signaling, disease, and emerging therapeutic modalities. Cell 2017; 169: 985-999.

[19] Yang X, Lv JN, Li H, Jiao B, Zhang QH, Zhang Y, Zhang J, Liu YQ, Zhang M, Shan H, Zhang JZ, Wu RM and Li YL. Curcumin reduces lung inflammation via Wnt/beta-catenin signaling in mouse model of asthma. J Asthma 2017; 54: 335-340.

[20] Fan D, Yang Z, Yuan Y, Wu QQ, Xu M, Jin YG and Tang QZ. Sesamin prevents apoptosis and inflammation after experimental myocardial infarction by JNK and NF-kappaB pathways. Food Funct 2017; 8: 2875-2885.

[21] Inserte J, Garcia-Dorado D, Hernando V and Soler-Soler J. Calpain-mediated impairment of Na^+/K^+-ATPase activity during early reperfusion contributes to cell death after myocardial ischemia. Circ Res 2005; 97: 465-473.

[22] Chu YF, Ke YS, Yu GH and Yang H. The significance and effect of myocardial ischemia reperfusion injury on gene expression of Na^+-K^+-ATPase isoforms in rat. Chinese Journal of Arteriosclerosis 2007; 15: 173-177.

[23] Ren HM, Ren AJ, Xu XW, Guo ZF, Yang LQ and Yu WF. Effects of propofol on isolated heart function and metabolism of oxygen free radical in rats with obstructive jaundice. Acad J Second Mil Med Univ 2007; 28: 640-643.

[24] Yu X, Sun X, Zhao M, Hou Y, Li J, Yu J and Hou Y. Propofol attenuates myocardial ischemia reperfusion injury partly through inhibition of resident cardiac mast cell activation. Int Immunopharmacol 2018; 54: 267-274.

[25] Gan X, Xing D, Su G, Li S, Luo C, Irwin MG, Xia Z, Li H and Hei Z. Corrigendum to "propofol attenuates small intestinal ischemia reperfusion injury through inhibiting NADPH oxidase mediated mast cell activation". Oxid Med Cell Longev 2017; 2017: 8932871.

[26] Wang Y, Liu MZ, Li HJ, Zhang WP, Gao Q and Li ZH. Effect of endomorphin-1 postconditioning against myocardial ischemia-reperfusion injury in rats and the role of Erk1/2 signaling pathway. Nan Fang Yi Ke Da Xue Xue Bao 2017; 37: 1028-1034.

[27] Zhu JX, Kong LH, Zhang CF, Sun N, Chang JR and Xu Y. Capsaicin alleviate myocardial ischemia reperfusion injury through attenuating mitochondrial oxidative stress. Sichuan Da Xue Xue Bao Yi Xue Ban 2017; 48: 716-720.