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ORIGINAL ARTICLE

Selective Activation of Cannabinoid Receptor 2 Attenuates Myocardial Infarction *via* Suppressing NLRP3 Inflammasome

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Abstract—The administration of cannabinoid receptor 2 (CB2R) agonist has been reported to produce a cardioprotective effect against the pathogenesis and progression of myocardial infarction (MI). Here in this study, we investigated the specific mechanism related to inflammatory suppression. JWH-133 was used for the activation of CB2R. MI mice models and cardiomyocytes under oxygen-glucose deprivation (OGD) challenge were used for the *in vivo* and *in vitro* studies, respectively. Detection of cardiac infarct size and levels of myocardial enzymes as well as echocardiographic examination were applied to assess MI severity and cardiac function. Cell viability and lactate dehydrogenase (LDH) release were detected *in vitro*. Real-time-polymerase chain reaction (RT-PCR) and enzyme-linked immunosorbent assay (ELISA) were used to detect the levels of proinflammatory cytokines. Western blot was used for the analysis of the NOD-like receptor family, pyrin domain containing 3 (NLRP3) inflammasome activation. We found that the administration of CB2R agonist attenuated the severity of MI through reducing infarct size ratio and levels of myocardial enzymes and improved cardiac function in ejection fraction (EF), fractional shortening (FS), left ventricular end-systolic diameter (LVESD), and left ventricular end-diastolic diameter (LVEDD) in MI mice. JWH-133 also produced a cardioprotective effect in murine primary cardiomyocytes by improving cell viability and LDH release. JWH-133 largely reduced the production and secretion of proinflammatory cytokines, which was significantly attenuated by AM630. HU308 showed the same effects as JWH-133. Taken together, we demonstrated for the first time the cardioprotective effect of CB2R agonist and its NLRP3 inflammasome-related mechanism in MI.

KEY WORDS: cannabinoid receptor 2; myocardial infarction; NLRP3 inflammasome; agonist.

INTRODUCTION

Myocardial infarction (MI), well known as heart attack, is one of the most serious diseases which leads to large mortality as well as other public problems. The occurrence of MI causes a deterioration of left ventricle cardiomyocytes and the subsequent damage of cardiac function, which eventually leads to heart failure [1]. It is widely acknowledged that the post-MI inflammatory and immune responses function as important pathways for cardiac repair [2]. However, the overreaction of those self-defensive responses leads to further damage of cardiomyocytes, thus aggravating the

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severity and outcome of MI in infarct size, cardiac function, and ventricular remodeling [3–5]. Consequently, the way to diminish overreactive inflammatory and immune reaction may probably serve as an effective and potential strategy in the treatment of MI.

Cannabinoid receptors belong to a class of G protein-coupled receptor superfamily that located on the surface of cellular membrane [6]. So far, two members of cannabinoid receptors have been known, namely cannabinoid receptor 1 (CB1R) and CB2R [7]. Although CB1R and CB2R are about 44% similar in sequence [8], the distribution and functions of those two members are largely different [9]. It has been reported that CB1R is mostly located in central nervous system, while CB2R is widely expressed in peripheral cells and tissues including the immune cells and tissues as well as other peripheral tissues including heart and brain, functioning in the suppression of inflammatory and immune reaction in various kinds of diseases [10–13]. In MI, although the protective role of activating CB2R has been demonstrated, the specific mechanisms remain to be clarified [14, 15].

The NOD-like receptor family, pyrin domain containing 3 (NLRP3) inflammasome, is one of the most well-studied forms of inflammasomes, which has been reported to be involved in the pathological process of various kinds of diseases, including central nervous system diseases, cardiovascular disorders, metabolic diseases, and auto-immune diseases *etc.* [16–21]. So far, numerous studies have highlighted the vital role of the NLRP3 inflammasome in the pathogenesis and progression of MI, pointing out that targeting on the suppression of the NLRP3 inflammasome provided a potential pathway in the treatment of MI [22–24]. However, few antagonists have been proven to be effective in clinical practice. In addition, although activating CB2R has been demonstrated to inhibit the NLRP3 inflammasome [25], whether this process is involved in the alleviative effect of activating CB2R on MI remains unclear.

Here in this study, we used JWH-133, a selective CB2R agonist, to explore the role and mechanism of CB2R in MI related to the NLRP3 inflammasome suppression. We believe that the current study may provide a novel insight in the treatment of MI in the development of new drugs.

MATERIALS AND METHODS

Animal Care and Use

C57BL/6J mice were purchased from Shanghai Super-B&K Laboratory Animal Corp. Ltd., Shanghai, China. Mice were kept at controlled environment (21 ± 4 °C, 12 h light/dark) with an ambient humidity of 50–80%. Experimental mice had free access to water and standard chow diet. All experiments were conducted according to the guidelines of Zhejiang Chinese Medical University.

Myocardial Infarction Mice Model Creation and Treatment

Permanent occlusion of the left anterior descending coronary artery was conducted on 8-week-old mice for the creation of MI mice model. Specifically speaking, mice were anesthetized with isoflurane (5%) in a ventilating equipment. Lateral thoracotomy was conducted for the exposure of the heart tissue. Left main coronary artery was ligated with a 9–0 prolene suture at 1 mm below the ostium. Experimental mice were placed in a warming pad until recovery after the chest cavity was closed. In *in vivo* studies, experimental mice were intraperitoneally injected with selective CB2R agonist JWH-133 at the doses of 1, 3, or 10 mg/kg as well as HU308 (2 mg/kg), another selective CB2R agonist, and AM630 (2 mg/kg), a selective CB2R antagonist as previously reported [26].

Triphenyltetrazolium Chloride Staining

Triphenyltetrazolium chloride (TTC) staining was conducted for the assessment of cardiac infarct size ratio. Specifically speaking, after anesthesia, heart tissues were isolated from experimental mice at 6-h post-MI. After washed with saline, heart tissues were frozen and cut into 5 transverse slices from apex to base of equal thickness (1–2 mm). The slices were then stained in 1% TTC followed by incubated in a 37 °C incubator for 8–10 min and fixed in 4% (*w*/*v*) paraformaldehyde for 30 min. The infarct size ratio was assessed using ImageJ software in a blinded fashion.

Echocardiographic Detection

After anesthesia, mice were subjected to echocardiographic detection with an M-mode echocardiography in a Vevo 2100 system (Vevo 2100, Visual Sonics) for the assessment of cardiac function. Left ventricular end-systolic diameter (LVESD) and left ventricular end-diastolic diameter (LVEDD) were obtained from at least three separate cardiac cycles. Ejection fraction (EF) and fractional shortening (FS) were analyzed by the equations described as follows: EF = (LVEDV-LVESV)/LVESV ×

100%; FS = (LVEDD-LVESD)/LVESD × 100%; left ventricular end-systolic diameter (LVESD) = 7.0 × $LVESD^3$/(2.4 + LVESD); left ventricular end-diastolic diameter (LVESD) = 7.0 × $LVEDD^3$/(2.4 + LVEDD).

Culture and Treatment of Murine Primary Cardiomyocytes

Murine primary cardiomyocytes were isolated from neonatal C57BL/6J mice. Specifically speaking, after harvested, heart tissues were dissociated with 0.05% trypsin (Gibco, Grand Island, NY, USA) and 0.08% type II collagenase (Gibco, USA). Cells were plated onto a 25-cm^2 cell culture flask for 90 min at 37 °C in a 5% CO_2 incubator cultured by Dulbecco's modification of Eagle's medium (DMEM; Gibco) supplemented with 10% fetal bovine serum (FBS) and 0.1% penicillin-streptomycin. Unadherent cells were plated onto 6-well (1×10^6 cells/well) or 96-well (1×10^5 cells/well) plates. For *in vitro* experiments, murine primary cardiomyocytes were cultured in glucose-free Earle's balanced salt solution (DMEM; Gibco) with 1% O_2 at 37 °C (oxygen-glucose deprivation, OGD) and used for the creation of *in vitro* model of MI. In some *in vitro* studies, JWH-133 was administrated for the selective activation of CB2R at the doses of 1, 10, or 100 nM in primary cardiomyocytes.

Cell Counting Kit-8

Cell counting kit-8 (CCK-8) (Dojindo, Kamimashiki-gun Kumamoto, Japan) was applied for the detection of cell viability according to the manufacturer's protocol. In brief, murine primary cardiomyocytes were seeded in 96-well plates in the density of 1×10^5 cells/well. After stimulation, CCK-8 reagent was added and a microplate reader (Tecan Group Ltd., Männedorf, Switzerland) was used for the assessment of absorbance at the wavelength of 450 nm.

Creatine Kinase-Muscle/Brain Analysis and Lactate Dehydrogenase Release

For the detection of serum creatine kinase (CK)-muscle/brain (MB) and lactate dehydrogenase (LDH), blood samples were collected from mice and centrifuged at 3000 rpm (soft) for 30 min at 4 °C. The serum in the upper layer was obtained and restored in − 80 °C refrigerator for further analysis. For the detection of LDH release in cardiomyocytes, cells were seeded in 96-well plates in the density of 1×10^5 cells/well followed by stimulation. Levels of CK-MB (Nanjing Jiancheng Bioengineering Research Institute, Nanjing, Jiangsu, China) and LDH release (Nanjing Jiancheng Bioengineering Research Institute, China) were detected by the related commercially available assays according to the manufacturer's instructions.

Real-time-Polymerase Chain Reaction

Total RNA from heart tissues and spleens isolated from experimental mice was isolated by Trizol (Invitrogen, Carlsbad, CA, USA). The first-strand cDNA was synthesized using PrimeScript RT Master Mix (Takara, Otsu, Shiga, Japan), and a 2ΔΔCT method was used to analyze the results of PCR with β-actin as the internal reference. 7500 Real-Time PCR System and the Fast Start Universal SYBR Green Master (Roche, Basel, Switzerland) were used for RT-PCR. Primers used were listed as follows: IL-1β: forward, 5′-GAAATGCCACCTTTTGACAGTG-3′ and reverse, 5′-TGGATGCTCTCATCAGGACAG-3′; IL-18: forward, 5′-GTGAACCCCAGACCAGACTG-3′ and reverse, 5′-CCTGGAACACGTTTCTGAAAGA-3′; IFN-γ: forward, 5′-AGTGGCATAGATGTGGAA-3′ and reverse, 5′-CTCAAACTTGGCAATACTC-3′; tumor necrosis factor-α (TNF-α): forward, 5′-CAGGCGGT GCCTATGTCTC-3′ and reverse, 5′-CGATCACC CCGAAGTTCAGTAG-3′; β-actin: forward, 5′-GTAA AGACCTCTATGCCAACA-3′ and reverse, 5′-GGAC TCATCGTACTCCTGCT-3′.

Enzyme-Linked Immunosorbent Assay

Blood samples were collected from mice and centrifuged at 3000 rpm (soft) for 30 min at 4 °C. The serum in the upper layer was obtained, and the levels of proinflammatory cytokines including IL-1β, IL-18, IFN-γ, and TNF-α in serum were analysis using commercial available ELISA kits (R&D system, New York, NY, USA) according to the manufacturer's instructions.

Western Blot

Heart tissues and primary cardiomyocytes were harvested and lysed in lysis buffer. The protein concentration was determined and separated using SDS-PAGE and electro-transferred to nitrocellulose membranes (Millipore, Billerica, MA, USA). Immunoblotting was conducted using the primary antibodies targeting NLRP3 (1:1000, Santa Cruz Biotechnology, Dallas, TX, USA), caspase-1 (Casp1, 1:1000, Abcam, Cambridge, MA, USA), IL-1β (1:1000, R&D, Minneapolis, MN, USA), and β-actin (1:5000, Beyotime

Biotechnology, Shanghai, China). Membranes were then incubated with an IRDye800CW-conjugated secondary antibody (Rockland; Gilbertsville, PA, USA). Images were obtained by the Odyssey infrared imaging system (LI-COR Bioscience, Lincolin, NE, USA).

Statistical Analysis

Data are presented as mean ± SEM. Statistical analysis was conducted using a one-way analysis of variance (ANOVA) followed by Bonferroni *post hoc* test or a Kruskal-Wallis test followed by a Dunn's post-test for the comparison of parametric or nonparametric data, respectively. $P < 0.05$ was considered as statistical difference.

RESULTS

Selective Activation of CB2R by JWH-133 Alleviates Myocardial Infarction Severity and Cardiac Function in Myocardial Infarction Mice

JWH-133, a selective CB2R agonist, was used for the activation of CB2R and study of CB2R at the doses of 1, 3, or 10 mg/kg body weight or vehicle at 5 min before ischemia. The administration of JWH-133 significantly attenuated the severity of MI in reducing the ratio of infarct size (Fig. 1a, b) and levels of CK-MB and LDH (Fig. 1c, d) in mice at 6-h post-MI. The dose of 10 mg/kg body weight presented the best effects in the attenuation of MI severity. In addition, at 1 week post-MI, JWH-133 treatment largely improved the cardiac function through the analysis of EF, FS, LVESD, and LVEDD (Fig. 2a–e). The dose of 10 mg/kg body weight presented the best effects in the improvement of cardiac function.

The Administration of JWH-133 Plays a Protective Role in Cardiomyocytes under Oxygen-Glucose Deprivation Challenge

Murine primary cardiomyocytes were treated with JWH-133 at the doses of 1, 10, or 100 nM or vehicle at 10 min before being exposed to OGD challenge. The administration of JWH-133 significantly improved cell viability (CCK-8) and reduced LDH release (Fig. 3a, b). The dose of 100 nM showed the best effects in the protection of cardiomyocytes under OGD challenge.

JWH-133 Treatment Suppresses the Production and Secretion of Proinflammatory Cytokines in Myocardial Infarction Mice

JWH-133 was administrated at the doses of 1, 3, or 10 mg/kg body weight or vehicle at 5 min before ischemia. The administration of JWH-133 significantly suppressed the proinflammatory cytokines from secretion in serum (Fig. 4a) and production in mRNA level (Fig. 4b). The dose of 10 mg/kg body weight presented the best effects in the suppression of proinflammatory cytokines. The administration of HU308 showed the same effects as JWH-133 on the proinflammatory cytokines from secretion in serum, while AM630 was demonstrated to attenuate the anti-inflammatory effect of JWH-133 in MI mice (Supplemental Fig. 1).

The Administration of JWH-133 Inhibits the Initiation and Activation of the NLRP3 Inflammasome in Myocardial Infarction Mice

JWH-133 was administrated at the dose of 10 mg/kg body weight or vehicle at 5 min before ischemia. The administration of JWH-133 significantly inhibited the initiation and activation of the NLRP3 inflammasome in heart tissues (Fig. 5a, b) and spleens (Fig. 5c, d).

JWH-133 Inhibits the Initiation and Activation of the NLRP3 Inflammasome in Cardiomyocytes under Oxygen-Glucose Deprivation Challenge

Murine primary cardiomyocytes were treated with JWH-133 at the dose of 100 nM or vehicle at 10 min before exposed to OGD challenge. The administration of JWH-133 significantly inhibited the initiation and activation of the NLRP3 inflammasome in cardiomyocytes under OGD challenge (Fig. 6a, b).

DISCUSSION

Here in this study, JWH-133 was chosen to selectively activate CB2R for the study of the role and mechanism of CB2R in MI. JWH-133, with the chemical formula of $C_{22}H_{32}O$, is regarded as an ideal selective CB2R agonist with a Ki of 3.4 nM. The selectivity for CB2R is about 200-fold compared to that for CB1R. JWH-133 has been administrated for the activation of CB2R in the study of various kinds of diseases, including cerebral hemorrhage, liver disorders, allergy, atherosclerosis, *etc*. [27–32]. In MI, it was previously reported that intraperitoneal injection of

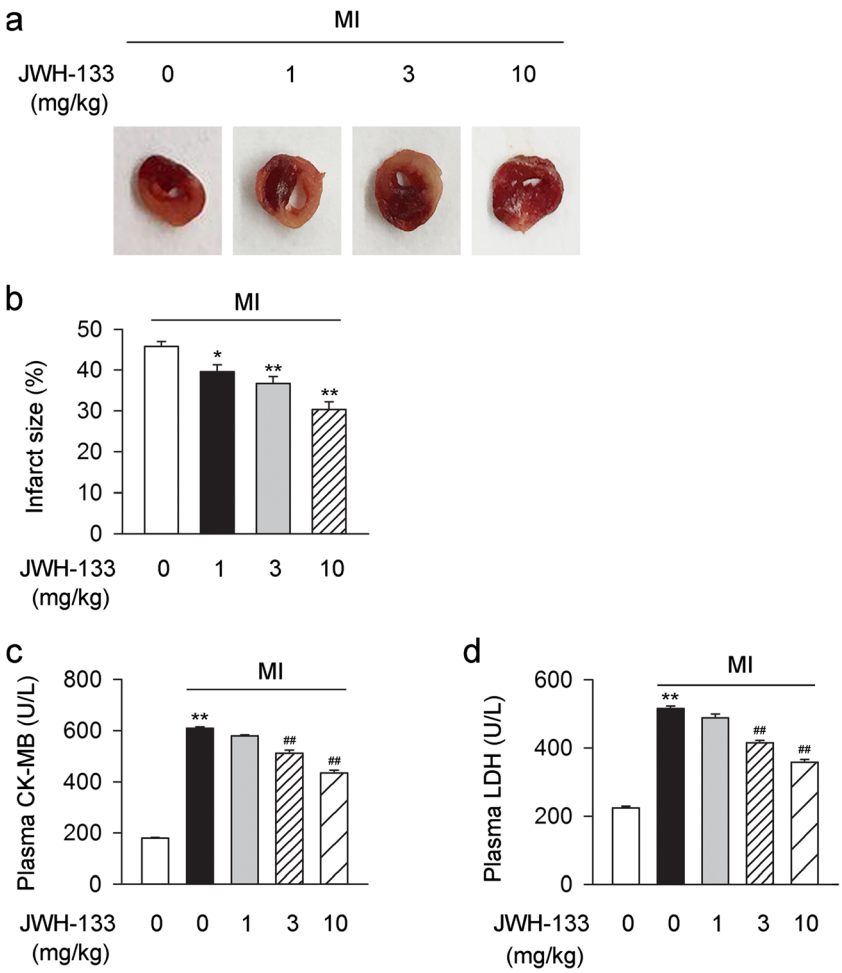

Fig. 1. Selective activation of CB2R by JWH-133 attenuates MI severity in MI mice. MI mice models were created through occlusion of left anterior descending coronary artery. Mice were intraperitoneally injected with selective CB2R agonist JWH-133 at 5 min before ischemia. **a** Representative images of heart tissues isolated from mice at 6 h post-MI. **b** Quantitative analysis of myocardial infarct size ratio ($n = 7$). $^{*}P < 0.05$ *vs.* MI group without JWH-133; $^{**}P < 0.01$ *vs.* MI group without JWH-133; data are presented as mean ± SEM. **c**, **d** Quantitative analysis of plasma CK-MB and LDH at 6-h post-MI. $^{**}P < 0.01$ *vs.* sham group; $^{\#\#}P < 0.01$ *vs.* MI group without JWH-133; data are presented as mean ± SEM.

JWH-133 significantly led to the reduction of the infarct size in ischemia/reperfusion mice models through the suppression of oxidative stress and neutrophil infiltration in the infarcted myocardium [32]. In addition, it was also demonstrated that the administration of JWH-133 produced a protective effect on cardiomyocytes against ischemia/reperfusion injury through the ERK1/2-related signaling pathway [33]. Consistent with those studies, here in our current study, we demonstrated that the administration of JWH-133 significantly attenuated the severity of MI in the reduction of cardiac infarct size and levels of myocardial enzymes at 6-h post-MI in MI mice models *in vivo*. Those results were further shown in cardiomyocytes undergoing OGD challenge *in vitro* demonstrating that JWH-133 treatment led to a protective effect in cardiomyocytes against OGD challenge in the improvement of cell viability and LDH release. Taken together, our data showed the cardioprotective effect of selective activation of CB2R by JWH-133 against ischemia both *in vitro* and *in vivo*.

The NLRP3 inflammasome is present in immune and inflammatory cells following the activation by inflammation-related stimuli [34]. The activation of the NLRP3 inflammasome mainly includes two steps, namely the initiation (increase in the transcription of inflammasome-related components, including inactive

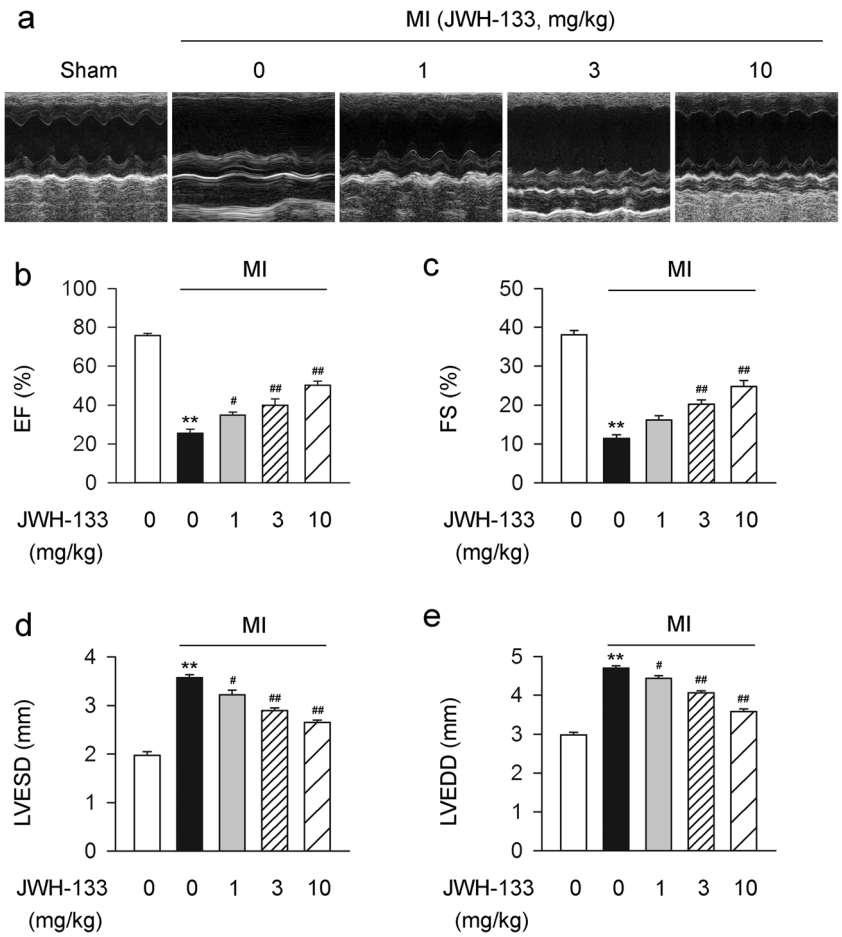

Fig. 2. The administration of JWH-133 improves cardiac function in MI mice. MI mice models were created through occlusion of left anterior descending coronary artery. Mice were intraperitoneally injected with JWH-133 at 5 min before ischemia. **a** Representative M-mode echocardiographic images on mice at 1 week post-MI. **b** Quantitative analysis of cardiac function index including EF, FS, LVESD, and LVEDD ($n = 8$). $^{**}P < 0.01$ *vs.* sham group; $\#P < 0.05$ *vs.* MI group without JWH-133; $\#\#P < 0.01$ *vs.* MI group without JWH-133; data are presented as mean ± SEM.

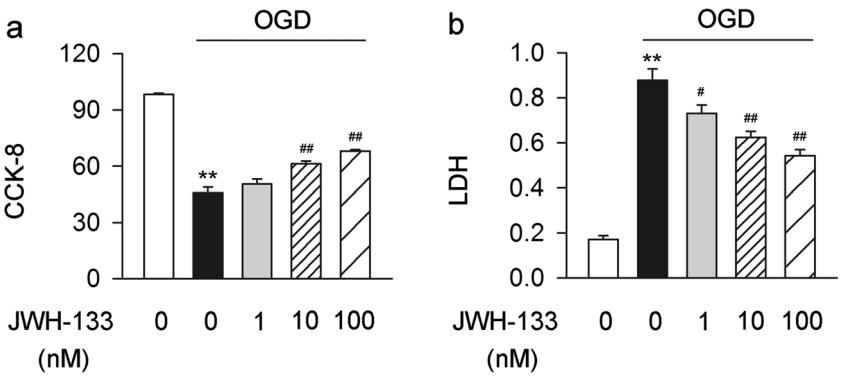

Fig. 3. The administration of JWH-133 protects cardiomyocytes against OGD injury. Murine primary cardiomyocytes were harvested and exposed to OGD for 4 h. JWH-133 was given at 10 min before OGD challenge. **a**, **b** Quantitative analysis of CCK-8 and LDH release in murine primary cardiomyocytes ($n = 6$). $^{**}P < 0.01$ *vs.* normal; $\#P < 0.05$ *vs.* OGD group without JWH-133; $\#\#P < 0.01$ *vs.* OGD group without JWH-133; data are presented as mean ± SEM.

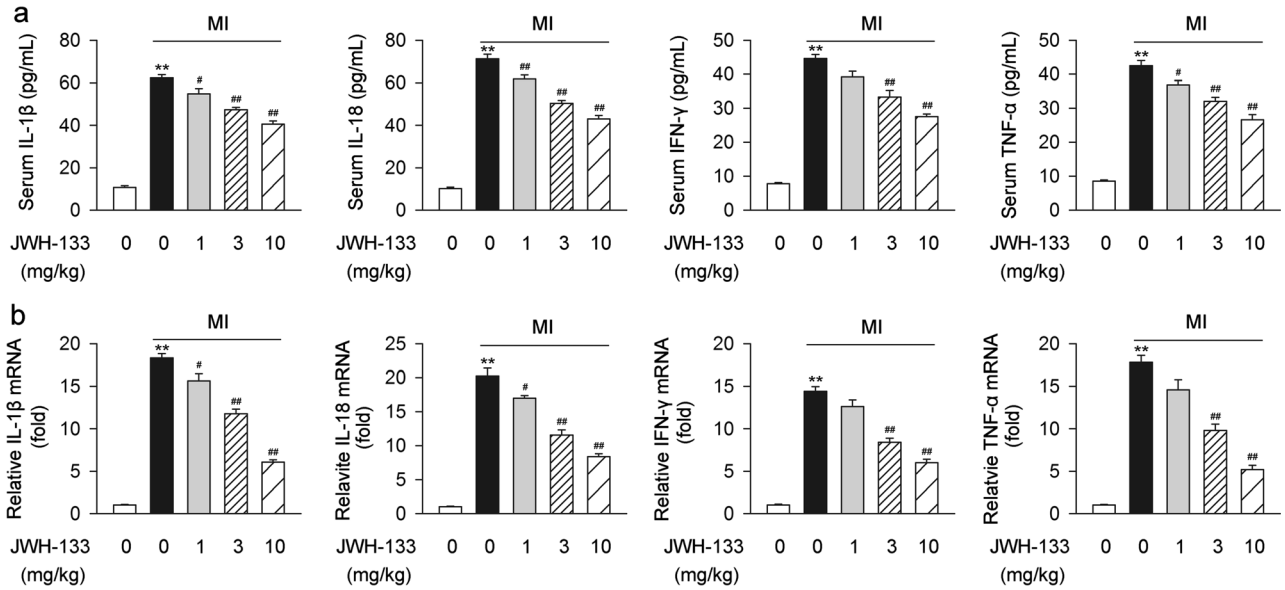

Fig. 4. The administration of JWH-133 suppresses the production and secretion of proinflammatory cytokines. MI mice models were created through occlusion of left anterior descending coronary artery. Mice were intraperitoneally injected with JWH-133 at 5 min before ischemia. **a** Quantitative analysis of proinflammatory cytokines including IL-1β, IL-18, IFN-γ, and TNF-α in serum at 6 h post-MI ($n = 6$). $^{**}P < 0.01$ *vs.* sham group; $^{\#}P < 0.05$ *vs.* MI group without JWH-133; $^{\#\#}P < 0.01$ *vs.* MI group without JWH-133; data are presented as mean ± SEM. **b** mRNAs were isolated from heart tissues at 6 h post-MI. Quantitative analysis of proinflammatory cytokines including IL-1β, IL-18, IFN-γ, and TNF-α in mRNA levels ($n = 6$). $^{**}P < 0.01$ *vs.* sham group; $^{\#}P < 0.05$ *vs.* MI group without JWH-133; $^{\#\#}P < 0.01$ *vs.* MI group without JWH-133; data are presented as mean ± SEM.

NLRP3, proIL-1β, and proIL-18) and further activation (oligomerization of NLRP3 and subsequent assembly of NLRP3, adapter protein apoptosis-associated speck-like protein (ASC) and procaspase-1 into a complex) [35, 36]. The activation of the NLRP3 inflammasome leads to the secretion of IL-1β and IL-18, involved in the inflammation-related pathological process of diseases [37]. As discussed above, the activation of the NLRP3 inflammasome is involved in the pathogenesis and progression of various kinds of diseases. So far, several agents targeting on the inhibition of the NLRP3 inflammasome have been successfully applied in clinical practice or related fundamental research [36]. In addition, several studies have reported the therapeutic effects on MI taking advantage of inhibiting the NLRP3 inflammasome [38–40]. For example, it was demonstrated that Ghrelin produced a cardioprotective effect against ischemic/reperfusion injury in the improvement of cardiac function through the inhibition of the NLRP3 inflammasome-related pathway [38]. In addition, it was reported that the reperfusion therapy with recombinant human relaxin-2 (serelaxin) contributed to the attenuation of myocardial ischemia/reperfusion injury and the subsequent caspase-1 activation through the eNOS-dependent mechanism [39]. Consist with those findings, in this study, we demonstrated that the administration of JWH-133 for the activation of CB2R significantly suppressed the production and secretion of several proinflammatory cytokines including IL-1β, IL-18, IFN-γ, and TNF-α both in serum and heart tissues in MI mice. In addition, we used HU308 in MI mice model in the same manner of JWH-133 and detected the level of serum IL-1β, IL-18, IFN-γ, and TNF-α at 6 h post-MI. We found that the administration of HU308 significantly suppressed the level of proinflammatory cytokines from secretion in serum. In another set of experiments, we used AM630, a selective CB2R antagonist, at 5 min after the administration of JWH-133. We found that AM630 significantly attenuated the anti-inflammatory role of JWH-133 in MI mice models. We further reported that JWH-133 led to the inhibition of the initiation and activation of the NLRP3 inflammasome in protein levels both *in vivo* in the heart tissues and spleen isolated from MI mice models and *in vitro* in cardiomyocytes under OGD challenge, thus indicating the inhibitory effect of JWH-133 on cardiomyocytes under ischemia.

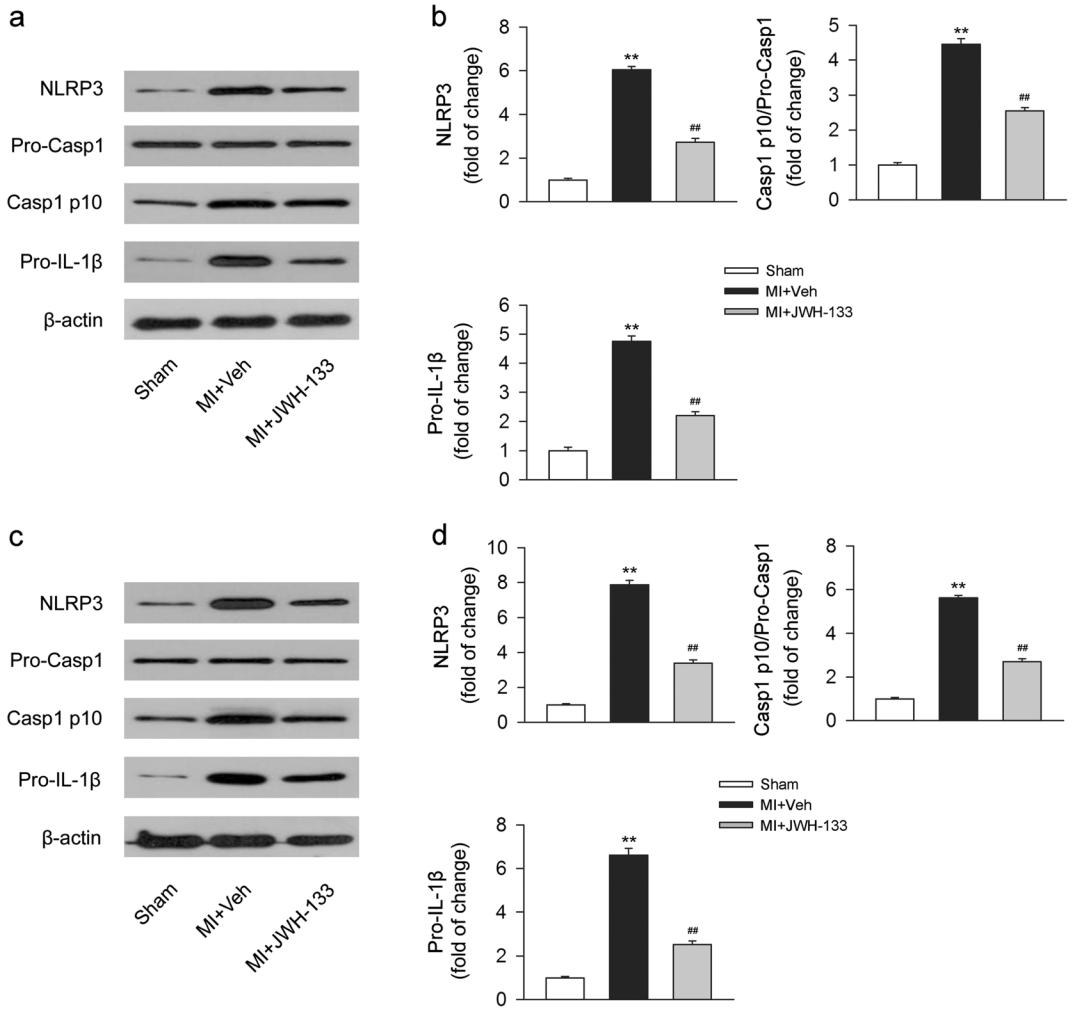

Fig. 5. JWH-133 inhibits the NLRP3 inflammasome in MI mice. MI mice models were created through occlusion of left anterior descending coronary artery. Mice were intraperitoneally injected with JWH-133 at 5 min before ischemia. Heart tissues and spleens were isolated from mice at 6-h post-MI. **a** Representative images of the NLRP3 inflammasome-related proteins in heart tissues detected by Western blot. **b** Quantitative analysis of the NLRP3 inflammasome-related proteins ($n = 5$). $^{**}P < 0.01$ *vs.* sham group; $^{\#\#}P < 0.01$ *vs.* MI + Veh group; data are presented as mean ± SEM. **c** Representative images of the NLRP3 inflammasome-related proteins in spleens detected by Western blot. **d** Quantitative analysis of the NLRP3 inflammasome-related proteins ($n = 5$). $^{**}P < 0.01$ *vs.* sham group; $^{\#\#}P < 0.01$ *vs.* MI + Veh group; data are presented as mean ± SEM.

As we discussed previously, CB2R is widely expressed in peripheral cells and tissues and highly involved in the pathogenesis and progression of various kinds of disorders in cardiovascular system, central nervous system, and metabolic system. For the relations between CB2R and the activation of the NLRP3 inflammasome, it was demonstrated previously that activating CB2R was highly involved in the therapeutic effect of electroacupuncture on inflammatory pain through the inhibition of the NLRP3 inflammasome in inflamed skin tissues as well as the skin macrophages, which also indicated the analgesic effect of activating CB2R [41]. In addition, it was reported that the administration of another selective CB2R agonist HU-308 significantly inhibit the initiation and progression of the NLRP3 inflammasome through the enhancement of the protective autophagy process in microglia, thus leading to the alleviation of multiple sclerosis [25]. This conclusion was proven by Ke et al. in peripheral macrophages as well as the inflammatory bowel disease mice models [42]. Here in our current study, we initially demonstrated the cardioprotective effect of activating CB2R through the inhibition of the NLRP3

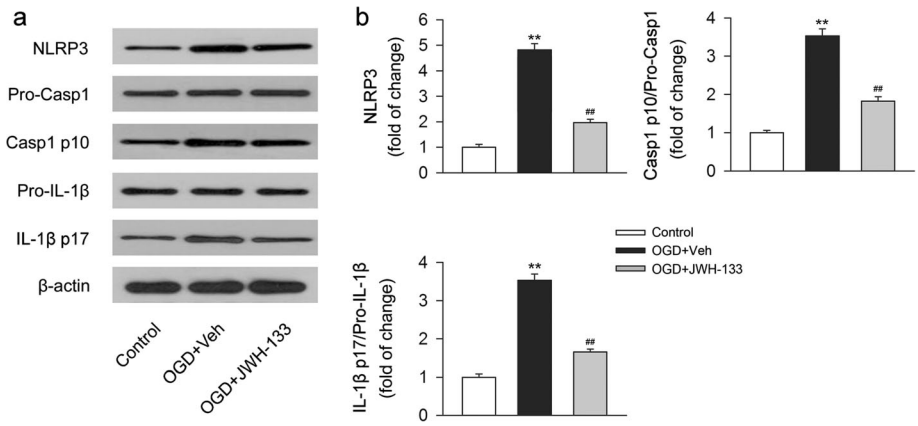

Fig. 6. JWH-133 inhibits the NLRP3 inflammasome in cardiomyocytes under OGD. Murine primary cardiomyocytes were harvested and exposed to OGD for 4 h. JWH-133 was given at 10 min before OGD challenge. **a** Representative images of the NLRP3 inflammasome-related proteins in cardiomyocytes detected by Western blot. **b** Quantitative analysis of the NLRP3 inflammasome-related proteins ($n = 5$). $^{**}P < 0.01$ *vs.* control group; $^{\#\#}P < 0.01$ *vs.* OGD + Veh group; data are presented as mean ± SEM.

inflammasome, thus uncovering the function of selective CB2R agonists through an NLRP3 inflammasome-related mechanism in the protection against MI or cardiac ischemia.

All in all, in our current study, we for the first time reported the role and specific mechanism of the alleviative effect of activating CB2R related to inflammasome suppression in MI. We demonstrated that selective activation of CB2R by JWH-133 significantly attenuated the severity of MI in the reduction of cardiac infarct size and improvement of cardiac function through the inhibition of the NLRP3 inflammasome. However, to develop agents of activating CB2R taking advantage of the NLRP3 inflammasome for the treatment of MI, further efforts are demanded for the detection and exploration.

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COMPLIANCE WITH ETHICAL STANDARDS

Conflict of Interest. The authors declare that they have no conflict of interest.

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