Activating the interleukin-6-Gp130-STAT3 pathway ameliorates ventricular electrical stability in myocardial infarction rats by modulating neurotransmitters in the paraventricular nucleus

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Research article

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Abstract

Background: Malignant ventricular arrhythmias are the most common cause of death in acute myocardial infarction. Recent studies have revealed direct paraventricular nucleus (PVN) involvement in occurrence of ventricular arrhythmias (VAs)，however, the mechanisms are incompletely understood. In this study we investigated the changes of Interleukin-6 (IL-6)-glycorprotein130-STAT3 pathway in the PVN during acute myocardial infarction and its effect on ventricular stability. Methods: The rats were divided into control group, myocardial infarction group, and paraventricular injected anti-IL-6 antibody group and paraventricular injected SC144 group to observe how interleukin-6 and its downstream glycoprotein130-STAT3 pathway in paraventricular nucleus effect the ventricular stability. Left anterior descending coronary artery was ligated to make myocardial infarction. After that, anti-Interleukin-6 antibody and SC144 were injected into the paraventricular. All data are expressed as the mean ± SEM and were analyzed by ANOVA by a post-hoc LSD test. $p<0.05$ was considered statistically significant. Results: After myocardial infarction (MI), the concentration of inflammatory factor interleukin-6 increased and its downstream glycorprotein130-STAT3 pathway was activated in the PVN. After anti-interleukin-6 antibody and glycorprotein130 inhibitor (SC144) injection into the PVN in MI rats, glutamate increased and γ-aminobutyric acid (GABA) decreased in the PVN. We also observed a higher plasma norepinephrine concentration, which increased the vulnerability to ventricular arrhythmias. Conclusions: In summary, interleukin-6 in the PVN plays a protective role in MI rats, and the glycorprotein130-STAT3 pathway plays a key role in this process. We anticipate that our findings will provide new ideas for the prevention and treatment of arrhythmia after MI.

Background

Acute myocardial infarction (AMI) is myocardium necrosis caused by acute, persistent ischaemia and hypoxia in the coronary arteries [1]. There are some complications of MI including heart failure, arrhythmia, heart rupture, pericarditis, papillary muscle rupture and so on. Arrhythmia comes up in most of MI patients and the most common within 24h [2]. Furthermore, lethal ventricular arrhythmias are the most common cause of death among patients with acute MI. It is well known that autonomic imbalance, especially the excessive activation of sympathetic nerves, plays the most important role in promoting the occurrence of arrhythmia, which is called Sympathetic storm. In recent years, there have been many reports on the mechanism how peripheral autonomic nerves regulate the arrhythmia, such as cardiac local nerve, renal sympathetic nerve, star ganglion and so on [3，4]. The mechanism of the central nervous system efffect on VAs is still not clear.

Lampert et al. have proven that ventricular tachycardia and ventricular fibrillation (VF) can be induced by psychological stress, sudden changes in mental state, brain trauma, and elevated intracranial pressure [5]. Davis et.al demonstrate that brain tissue regions and nuclei, from the medulla to the cerebral cortex, play an important role in the development of arrhythmia, and there are complex and variable interconnections [6]. Stimulating different brain regions and nerve nuclei can lead to different types of arrhythmias, among these regions, PVN is the main accumulation area of sympathetic preganglionic

neurons, and innervates other autonomic nuclei including midbrain periaqueductal gray, parabrachial region , rostral ventro lateral medulla, nucleus of solitary tract dorsal vagal nucleus and nucleus ambiguous. Moreover, PVN is also an important integrative site within the brain composed of magnocellular and parvocellular neurons. Parvocellular neurons project to other sites within the central nervous system (CNS), including regions that are important in autonomic control [7, 8]. However, the exact mechanism of the PVN's effect on arrhythmia remains unclear and needs further investigation. Changes in neurochemical factors, such as reactive oxygen species and inflammatory cytokines, in the hypothalamic PVN during MI may be important factors in the increase in sympathetic nerve sensitivity during MI. Kang et. al have shown that microinjection of pro-inflammatory cytokine inhibitors into the central nervous system can alleviate the symptoms of MI, and the effect is significantly better than that of the peripheral route of administration [9, 10]. The neurotransmitter play an important role in this process. For example, glutamate is enhanced and GABA declines in the PVN during MI, thereby affecting sympathetic over-activation and further affecting heart function [11]. Glutamate is one of the most important excitatory amino acids in the central nervous system, and it regulates sympathetic nerve activity and cardiovascular function through the NMDA receptor. Stimulation of NMDA receptors in the PVN can increase sympathetic discharge. GABA is the main inhibitory neurotransmitter in the PVN of the hypothalamus. Injecting GABA into the PVN of the hypothalamus can reduce heart rate and reduce arrhythmia. GAD67 is a rate-limiting enzyme of GABA and a marker for the recognition of GABAergic neurons. Its distribution is parallel to GABA.

Different from other inflammatory factors, Interleukin-6 is a pleiotropic regulator, which has multiple functions, not only pro-inflammatory but also tissue regeneration, metabolism and more. IL-6 increases significantly during the acute injury and plays a key role in mediation acute phase response. IL-6 has two kinds of IL-6 receptors, membrane bound receptor and soluble receptor, which both can bind to Glycoprotein 130 (Gp130). After the formation of dimerization, intracellular signalling carried on. They were called IL-6 classic signalling and IL-6 trans-signalling. Interesting, these two pathways strongly differ in their biologic influences. While classic signalling is primarily in protection, promoting tissue regeneration and maintaining physiological homeostasis, trans-signalling has deleterious effects [12]. For the central nervous system, Suzuki et al. have demonstrated that IL-6 plays a protective role in the early stage of brain injury. Intracerebroventricular injection of rhIL-6 dramatically reduced ischaemic brain damage when measured 24 hours after middle cerebral artery occlusion [13, 14, 15].

Gp130 is the receptor of IL-6 and is the main signalling molecule for intracellular signal transduction. Currently it is known that there are three signal pathways associated with Gp130, which are the JAK-STAT pathway, the EKA pathway, and the PI3K/Akt pathway. The most prominent proteins recruited to gp130 are the transcription factors of the family of signal transducers and activators of transcription (STAT) STAT3 and (to a certain extent) STAT1. Furthermore, these days it is well accepted that mainly STAT3 and to a much lesser extent STAT1 is activated by IL-6. Binding to IL-6 causes phosphorylation of Gp130 and then activates the cytoplasmic region [16]. Phosphorylated Gp130 exposes a binding site to STAT3 to induce STAT3 phosphorylation and then enters the nucleus to initiate transcription. Habecker et al. have confirmed that Gp130 mediates the conversion of peripheral sympathetic neurons to cholinergic neurons

after MI [17]. The sympathetic co-release of Ach and NE impairs adaptation to high heart rates and increases arrhythmia susceptibility. In the central nervous system, the Gp130 pathway promotes the differentiation and growth of nerves [18, 19]. However, the effect of Gp130 on neurontransmitter conversion in the PVN has not been studied.

The aim of this study was to investigate whether IL-6 in the hypothalamus PVN plays a protective role in the incidence of ventricular arrhythmia after MI and whether the Gp130-STAT3 pathway plays a key role in this process.

Methods

Animals

Adult male Sprague Dawley rats (200-250 g) were purchased from the Animal Experimental Center of Second Affiliated Hospital of Harbin Medical University. Housing conditions included 8 rats per cage, 12 hours light, freely available food and water, temperature 23±2°C, and relative humidity 40%-50%.

Coronary ligation and paraventricular nucleus injection (PNI) [20, 21]

Rats underwent sterile surgery under anaesthesia (Ulatan, 150mg/kg i.p.) for induction of MI by ligation of the left anterior descending coronary artery (MI group) or the same surgery without ligation of the vessel (Sham group). The PVN in each rat was injected with artificial cerebrospinal fluid (ACSF for Sham rats and MI rats), anti-IL-6 antibody or Gp130 antagonist (SC144) according to the rat stereotaxic atlas coordinates, each group included 12 rats (n=12) [22, 23, 24].

Cardiac electrophysiological studies

After the surgery, we recorded the arrhythmia occurrence in rats within 24 hours using a single-lead dynamic electrocardiogram (Good friend, Shenzhen, China). Rats were anesthetized by intraperitoneal injection Ulatan (150mg/kg i.p.), a second thoracotomy was carried out to perform the open-chest electrophysiological study, including ventricular fibrillation threshold (VFT)and the VF inducibility. A 1.9F electrophysiological catheter (Scisense, Canada) was placed on the left ventricular with eight poles recording electrocardiograms by the Electrophysiology Lab Amplifier (GY-6000, Huanan Medical Science and Technology, Henan, China). VFT, the minimum voltage to induce sustained VF, 60ms S1-S1 stimuli were repeatedly applied to the left ventricular apex with an increase of stimuli intensity by 0.5 V each time until VF was induced. VF inducibility, ten bursts of ventricle pacing (25 Hz), lasting for 10 s each time, were used to assess the inducibility of VF. VF was defined as >1000 ms of irregular ventricle arrhythmia.

Method of animal euthanasia and tissue collection

Animals were sacrificed by rapid excision of heart to confirmation of permanent cessation of the circulation under anaesthesia (Ulatan, 150mg/kg i.p.). For the immunohistochemistry, 4% paraformaldehyde was inserted into the left ventricle and the ascending aorta to fix the brain tissue, then

the rats were decapitated to get the brain. For the western blot, brain tissue was quickly extracted in a low temperature environment, and Palkovits's microdissection procedure was used to isolate the PVN.

Immunohistochemistry

After the brain tissue embedded with paraffin, the part between the optic chiasm and mammillary body was resected in the rostral to caudal direction. The tissue underwent serial sectioning on a paraffin slicer, and sections that were approximately 1.50 mm from the bregma were obtained [25, 26]. After dewaxing the slice, 3% H_2O_2 was used to extinguish endogenous peroxidase and 0.01M citric acid was used to repair antigen. Then the slices were incubated with primary antibodies overnight in 4℃. Immunohistochemical labelling was performed on sections to identify IL-6 (Bioss, China, 1:100), Gp130 (Santa Cruz, America, 1:20), pSTAT3 (Bioss, China, 1:50), and NMDA (Bioss, China, 1:50), GAD67(Abcam, England, 1:2000), then incubated with secondary antibodies□anti-mouse for Gp130 and GAD67, anti-rabbit for NMDA, pSTAT3 and IL-6□for 20 minutes in room temperature. For each rat, the positive neurons within the bilateral borders of the PVN were manually counted in three consecutive sections, and an average value was reported.

Western blot analysis

The tissue was homogenized in RIPA buffer containing protease inhibitor cocktail (Beyotime Biotechnology). A BCA protein assay (Beyotime Biotechnology) was used to determine protein concentrations. Equal amounts of protein were separated by SDS-PAGE electrophoresis and then transferred electrophoretically to a polyvinylidene fluoride membrane (Bio-Rad) [27, 28]. The membranes were incubated with the following primary antibodies for 2 hours at room temperature: IL-6 (1:1000, Abcam, England), Gp130 (1:1000, Santa, America), pSTAT3 (1:1000, Bioss, China), and NMDA (1:1000, Bioss, China). GAPDH (1:1000, Solarbio, China), Goat anti-mouse IgG (Bioss, China, Gp130, 1:1000 and GAD67,1:1000) or goat anti-rabbit IgG (Bioss, China, pSTAT3,1:1000 , NMDA, 1□3000 and IL-6□1:1000) was used as a secondary antibody for a 2 hours incubation at room temperature. Finally, the membrane was placed in a gel imaging analysis system for exposure and analysis (AlphaView-fluorChemFC3).

Measurement of glutamate and GABA in PVN tissues

Brain tissue was separated as previously described. Perchloric acid (0.1 mol/L, Sigma) was added to the brain tissue. Then, the tissue was dissolved on an ice pack or crushed ice, fully mashed and homogenized, and sonicated for 5 minutes. Finally, the samples were centrifuged at 12,000 rpm for 10 minutes at 4°C. The supernatant was aspirated, diluted and filtered through a filtration membrane. The concentrations of glutamate and GABA were measured using a liquid chromatography mass spectrometer (Singapore, Xevo).

Measurement of circulating catecholamine levels

The arterial blood was drawn from the left heart chamber and placed in a centrifuge, centrifuged at 3,000 rpm for 15 min at 4 °C and got the supernatant, Stored in a refrigerator at -80 °C. The ELSIA kit to test NE was purchased from Bioss. Standard dilution and loading: After dilution, the volume of each well was 50 μl, the concentration was 120 ng / L, 80 ng / L, 40 ng / L, 20 ng / L, 10 ng / L. The sample to be tested on the enzyme-labeled plate is first diluted with a sample of 40 μl, and then 10 μl, of the sample to be tested is added. 50 μl of enzyme labeling reagent per well. After sealing with a sealing film, incubate at 37 °C for 30 minutes. Zeroing with blank holes, measuring the absorbance of each hole in sequence at 450nm and 630 nm wavelength. The concentration of sample was calculated based on absorbance

Statistical analysis

All analyses were carried out with SPSS v17.0 software.All data were expressed as the mean ± SEM and were analysed by ANOVA followed by a post-hoc LSD test. Statistical significance was accepted at $p < 0.05$ for all analyses.

Results

Cardiac Electrophysiological study

In fig.1A, showed a typical VF graph induced by electrophysiological catheter, fig.1B showed ventricular premature beat recorded by dynamic electrocardiogram. In fig.1C,D,E, MI rats showed an increased incidence of spontaneous ventricular arrhythmias(5.5±0.8 vs 1.3±0.6)(p<0.01) than Sham rats. The incidence of spontaneous ventricular arrhythmias (54.67±5.59 and 200.8 ±19.12) of paraventricular nucleus injected (PNI) anti-IL-6 antibody-treated rats or PNI SC144 treated rats was significantly higher than that in sham rats and MI rats (p<0.0001). To further study the changes in cardiac electrophysiology, we measured cardiac-induced VF and VF threshold in each group of rats. The induced VF of MI rats was over 50% higher than that of sham rats（31.83±3.43% vs 13.33±1.41%）(p<0.01), meanwhile PNI anti-IL-6 rats and PNI SC144 rats had much higher induced VF (58.17±5.74% and 81.33±6.01%) than MI rats (p<0.0001). For the VF threshold, compared to sham rats (10.75±1.20V), MI rats (7.25±0.63V) showed a decreased VF (10.75±1.20V vs 7.25±0.63V) threshold (p<0.0001), and the VF (6.16±0.60V and 3.66±0.45V) threshold was lower in PNI anti IL-6 rats and PNI SC144 rats than MI rats (p<0.05).

IL-6, Gp130 and pSTAT3 expression in the PVN

The immune system was activated when the left anterior descending coronary artery was ligated. As shown in Fig 2-A, the immunohistochemical image shown the expression IL-6 in four groups. For the fig 2-C, demonstrated the immunohistochemical expression of Gp130 in four groups and fig 2-E account for the immunohistochemical expression of pSTAT3 in four groups. Figure fig 2-B, D, F were the densitometric analysis of IL-6, Gp130 and pSTAT3 respectively. The figue2-G showed the representative immunoblot image of IL-6, Gp130 and pSTAT3. From this picture we could observe that compared to sham rats, the IL-6 concentration in MI rats' PVNs increased significantly (p<0.0001), which then activated the Gp130 receptor (p<0.0001) and its downstream mediator pSTAT3 (p<0.01). With the

reduction in IL-6 content by injection of anti-IL-6 antibody, Gp130-pSTAT3 activation was blunted (p<0.05). Gp130-STAT3 activation was blunted conspicuously by SC144, which could bind to Gp130 and eventually abrogate STAT3 phosphorylation and nuclear translocation, Figure 2-H-J showed densitometric analysis of protein expression of IL-6,Gp130 and pSTAT3.

NMDA and GAD67 expression in the PVN

The fig3-A demonstrated the immunohistochemical expression of NMDA in four groups and Fig 3-C account for the immunohistochemical expression of GAD67 in four groups. Fig 3-B, D were the densitometric analysis of NMDA and GAD67 respectively. The figue3-E showed the representative immunoblot image of NMDA and GAD67, which demonstrated that MI rats had higher NMDA levels and lower GAD67 levels in the PVN than did sham rats (p<0.01). PVN injections of anti-IL-6 antibody and SC144 promoted increases in NMDA and decreases in GAD67 within the PVN of MI rats (p<0.05).

Neurotransmitters in the PVN

In fig.4 A, B,We observed significant changes of excitatory neurotransmitters and inhibitory neurotransmitters in the PVN of MI and PNI rats compared to those in sham rats. Compared to sham rats, MI rats had higher levels of glutamate (0.64 ±0.08 *vs* 0.90±0.09μg/mg, p<0.05) and lower levels of GABA in the PVN (304.0±12.0 *vs* 232.3±9.1 ng/mg, p<0.0001). Furthermore, PNI anti-IL-6 rat and anti SC144 rats had more glutamate (1.28±0.09μg/mg and 2.57±1.13μg/mg) (p<0.0001) and less GABA(GABA:165.9±8.2ng/mg and135.2±8.1ng/mg) (p<0.0001) in the PVN than MI rats did .

Plasma humoral factors

To determine the sympathoexcitation effect of MI and PVN infusion, we measured plasma NE levels in blood using ELISA. In fig.4 C, as expected, compared with sham rats, MI rats had much higher plasma NE (412.7±16.5pg/ml *vs* 630.5±21.2pg/ml) (p<0.0001). Additionally, PNI anti IL-6 rats and PNI SC144 rats had higher levels of plasma NE (850.5±23.2pg/ml and 1002.0±29.9pg/ml) than MI rats did (p<0.0001) (Figure 4).

Discussion

The novel finding of the present study is that IL-6 and its downstream Gp130-STAT3 induce an imbalance between excitatory and inhibitory neurotransmitters and their rate-limiting enzymes in the PVN MI rats, which contributes to sympathoexcitation and incidence of ventricular arrhythmias.

Central nervous system disease can induce multiple arrhythmias including ventricular tachycardia and ventricular fibrillation. Exploring central nervous system-related ventricular premature contractions is of great significance for clinical work. Thus, PVN may be a potential target for prevention and treatment of VA in patients with AMI. Neuroanatomical studies have shown that the PVN sends direct projections to spinal preganglionic neurons of sympathetic ganglia. Stimulation of cell bodies in the PVN causes an increase in blood pressure, heart rate and circulating NE concentrations [29]. Injecting glutamate or GABA

antagonist into the PVN resulted in an increase in renal nerve activity and circulating NE concentration, suggesting that a sympathoadrenal component to cardiovascular responses was associated with PVN stimulation [30]. Recent research demonstrated that pathophysiological changes in the PVN are undoubtedly critical to the elevated sympathetic nerve activity in MI. In response to MI, microglia in PVN become activated and secrete cytokines. In this study, we observed an increase in the expression of IL-6 and the activation of Gp130-STAT3 in PVN neurons of MI rats. Interestingly, while previous studies demonstrated that other cytokines, such as TNF-α and IL-1 in PVN, play a devastating role, IL-6 in PVN plays a protective role in our study. When MI rats were treated with IL-6 antagonist, the sympathetic outflow increased. The role of IL-6 is complicated during the inflammation process, which contributes to both injury and repair processes. However, the peak in IL-6 (24 hours) expression is associated with neuroprotection [12]. And many studies have shown that IL-6 dose-dependently protects neurons against NMDA toxicity. Activation of NMDA receptors can increase sympathetic discharge. In our study, we observed that blocking IL-6 induced the increased concentration of Glu and elevated expression of NMDA in the PVN whereas the decreased concentration of GABA and reduced expression of GAD67 in the PVN.

Moreover, the Gp130-STAT3 pathway plays a key role in this process. When we treated MI rats with a Gp130 antagonist (SC144), also gave rise to the same changes in neurotransmitters in PVN as which injected anti-IL-6, This process lead to the sympathetic outflow increased with incidence of ventricular arrhythmias elevated.

As common signal transducing receptor subunit, Gp130 acts in association with ligand-specific receptors of IL-6. Now in clinic, drugs that antagonize IL-6 mostly targets at IL-6 and IL-6R, whereas they exist detrimental offside effects, such as bacterial infections. In this trail we chose the SC144—a novel specific small molecule inhibitor of Gp130, to block the signal of IL-6 by blocking Gp130, which is a new target for blocking IL-6 signaling [31]. The intracellular signal transduction induced by IL-6 involves the activation of JAK tyrosine kinase family members, leading to the activation of transcription factors of the STAT family. STAT3 is an important element in the JAK-STAT pathway. The phosphorylation of STAT3 at Tyr705 in response to Gp130-stimulating cytokines leads to the formation of STAT3 dimers, followed by the translocation of these dimers to the nucleus where they regulate the transcription of target genes [16].

Increases in cholinergic genes within the stellate ganglion and widespread co-expression of ChAT protein in TH**+** neurons have been detected in MI rats. The acquisition of cholinergic function requires the expression of the Gp130 cytokine receptor in sympathetic neurons. Removal of Gp130 from sympathetic neurons also prevents the local expression of noradrenergic transmission in the left ventricle after acute MI. In this study, Gp130 played the same role in the PVN by transforming glutamate into GABA and inducing an imbalance between excitatory and inhibitory neurotransmitters, thereby further affecting the outflow of sympathetic activity..

In summary, the present study demonstrates that MI rats have higher concentrations of IL-6 and Gp130-STAT3 in the PVN, thereby contributing to sympathetic nerve inhibition and higher ventricular electrical stability. Our findings provide new insights into the treatment of ventricular arrhythmias in MI rats.

Preservation of IL-6-Gp130-STAT3 in the PVN can reduce the occurrence of ventricular arrhythmia in the acute phase of MI.

Limitations

We need to consider the effect of the depth of anaesthesia on autonomic nervous system (ANS). Although we gave anaesthetic according to the weight of rats, there is individual differences in self-efficacy. Another limitation of the study is that we elected to diagnose VAs from a single-lead electrocardiogram during 24 hours recording. Although the diagnostic accuracy is well in human, the rats need more experiments to confirm.

Declarations

Ethics approval and consent to participate

All experimental protocols were approved by local animal care and use committee (Animal Experimental Ethics Association Of the First Affiliated Hospital of Harbin Medical University). The methods were carried out in accordance with the revised Animals (Scientific Procedures) Act 1986.

Consent to publish

Not Applicable

Availability of data and materials

The datasets supporting the conclusions of this article are included within the article.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

All authors have read and approved the manuscript. Conceptualization, MG, DCY and XFQ; Data curation, MG; Formal analysis, MG; Funding acquisition, DCY and XFQ; Investigation, MG; Methodology, MG and JGC; Project administration, XFQ; Resources, XFQ; Software, JGC; Supervision, DCY and XFQ; Writing-original draft, MG; Writing-review and editing, DCY, and XFQ.

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Figures

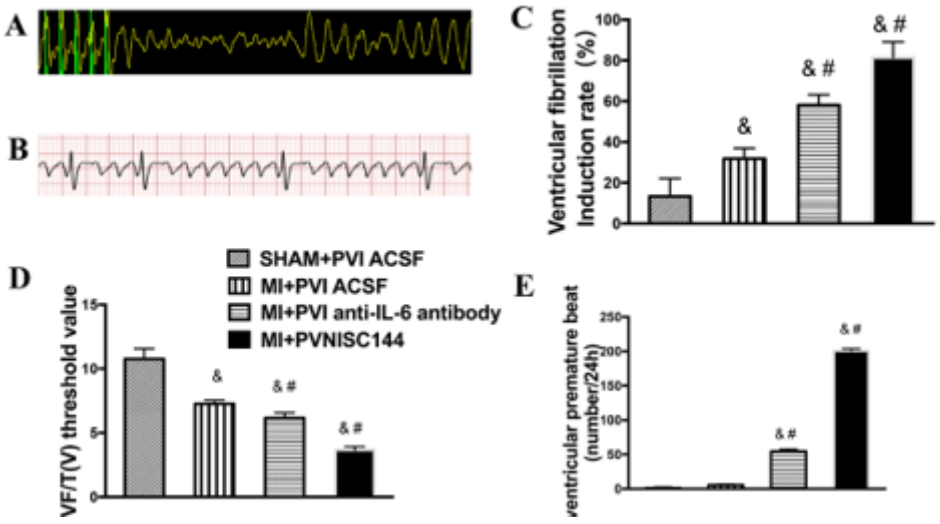

Figure 1

Effects of PVN infusion of anti- IL-6-antibody or SC144 on ventricular electrophysiological activity in MI rats. (A) Recordings of typical inducible ventricular arrhythmias by programmed electrical stimulation; (B) the ventricular premature beat within 24 hours recording by the small animal dynamic electrocardiogram; (C) a quantitate analysis of induced ventricular fibrillation; (D) A quantitate analysis of ventricular fibrillation threshold; (E) A quantitative analysis of premature ventricular beats in 24 hours. Values are mean±SE. &p<0.05 vs sham rats, #p<0.05 vs MI rats. Abbreviations: PVN, paraventricular nucleus; IL-6, Interleukin-6; MI, myocardial infarction.

Figure 2

Effects of PVN infusion of anti-IL-6 antibody or SC144 on IL-6, Gp130 and pSTAT3 levels within the MI in MI rats: (A) A representative immunohistochemical image of IL-6; (B) densitometric analysis of IL-6 staining; (C) a representative immunohistochemical image of Gp130; (D) densitometric analysis of Gp130 staining; (E) a representative immunohistochemical image of pSTAT3; (F) densitometric analysis of pSTAT3 staining. (G) A representative immunoblot image of IL-6, Gp130 and pSTAT3. (H-J) Densitometric analysis of protein expression of IL-6, Gp130 and pSTAT3 (n=4). Values are the mean ± SE. &p<0.05 vs SHAM, #p<0.05 vs MI rats. Abbreviations: PVN, paraventricular nucleus; IL-6, interleukin-6; MI, myocardial infarction; Gp130：glycoprotein130;

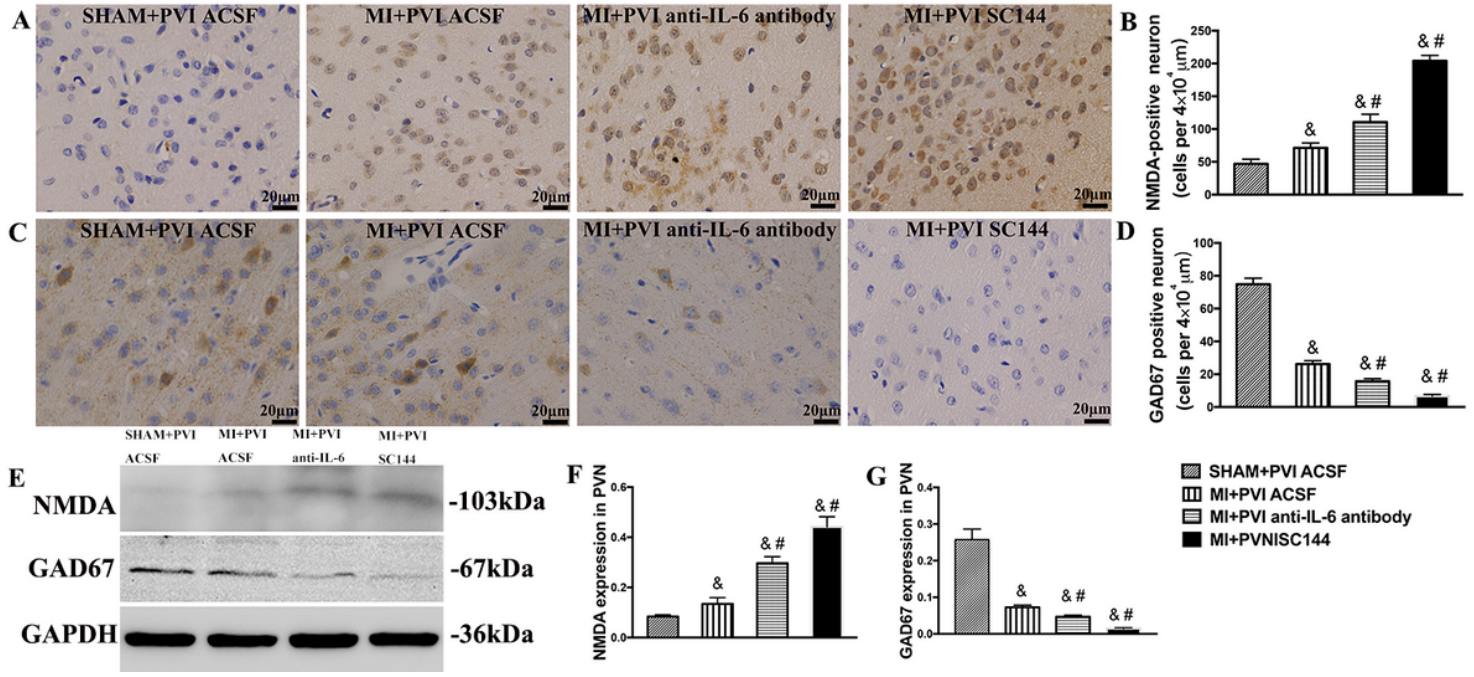

Figure 3

Effects of PVN infusion of anti-IL-6 antibody or SC144 on NMDA and GAD67 within the PVN in MI rats: (A) a representative immunohistochemical image of NMDA; (B) densitometric analysis of NMDA staining; (C) a representative immunohistochemical image of GAD67; (D) densitometric analysis of GAD67

staining; (E) a representative immunoblot image of NMDA and GAD67; (F) densitometric analysis of protein expression of NMDA and GAD67 (n=4). Values are the mean ± SE. &p<0.05 vs SHAM, #p<0.05 vs MI rats. Abbreviations: PVN，paraventricular nucleus；IL-6，interleukin-6；MI，myocardial infarction.

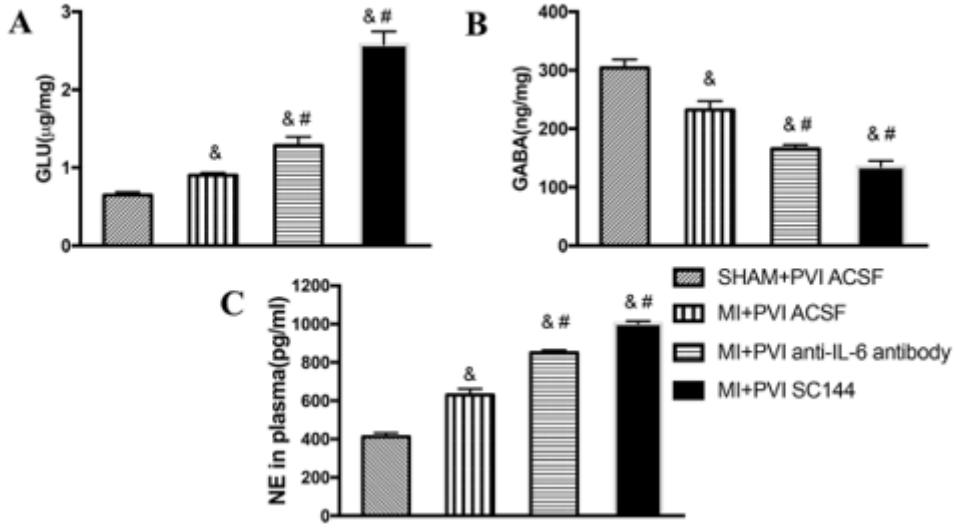

Figure 4

Effects of PVN infusion of anti-IL-6 antibody or SC144 on the PVN glutamate and GABA concentration in MI rats. (A) Glutamate; (B) GABA. Values are the mean ± SE. (C) Quantitative analysis of plasma NE. &p<0.05 vs SHAM rats, #p<0.05 vs MI rats. Abbreviations: PVN, paraventricular nucleus; IL-6, Interleukin-6; MI, myocardial infarction; Gp130:glycoprotein130; GABA,γ-aminobutyric acid.

Supplementary Files

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