International Journal of Cardiology 279 (2019) 64–71

Contents lists available at ScienceDirect

International Journal of Cardiology

journal homepage: www.elsevier.com/locate/ijcard

The effects of interleukin 17A on left stellate ganglion remodeling are mediated by neuroimmune communication in normal structural hearts

Jielin Deng [1], Xiaoya Zhou [1], Menglong Wang, Meng Wang, Liping Zhou, Guannan Meng, Yuhong Wang, Zhenya Wang, Yifeng Zhang, Zhen Zhou, Lilei Yu, Hong Jiang *

Department of Cardiology, Renmin Hospital of Wuhan University, Wuhan, Hubei, China
Cardiovascular Research Institute, Wuhan University, Wuhan, Hubei, China
Hubei Key Laboratory of Cardiology, Wuhan, Hubei, China

ARTICLE INFO

Article history:
Received 25 September 2018
Received in revised form 6 December 2018
Accepted 2 January 2019
Available online 4 January 2019

Keywords:
Interleukin-17A
Cardiac autonomic nerves system
Left stellate ganglion
Neuroimmune

ABSTRACT

Background: It is reported interleukin (IL)-17A, a classical proinflammatory cytokine, is implicated in neuroimmune-associated remodeling in neural plasticity and pathological conditions. However, the effect of IL-17A on left stellate ganglion (LSG) remodeling remains unclear.

Objective: This study was performed to determine whether exogenous IL-17A promotes LSG remodeling and destabilize ventricular electrophysiological properties (EPs) in normal canines.

Methods: 24 beagles were randomly allocated into three groups. In the first group, animals were subjected to 0.1 ml phosphate buffer saline (PBS) microinjection of into LSG ($n = 8$), an equivalent IL-17A was administrated in the second group ($n = 8$), and an equivalent anti-IL-17A mAb plus IL-17A was administrated in the third group ($n = 8$). The ventricular EPs, neural function and activity of the LSG were determined at baseline and 30 min after administration. In the end, LSG tissues were collected.

Results: Compared with the control group, the experimental group had a significantly shorter effective refractory period (ERP) and action potential duration $(APD)_{90}$, an increased ERP, APD_{90}, S_{max} dispersion, and APD alternans cycle length; and steepened APD restitution curves. In addition, IL-17A enhanced the neural function and activity of the LSG, upregulated the expressions of neuropeptides and proinflammatory cytokines and cells. And all these effects were attenuated by anti-IL-17A mAb. Importantly, IL-17 receptor A (IL-17R-A) was detected in sympathetic neurons in the LSG.

Conclusion: IL-17A promoted LSG remodeling by regulating the neural inflammation response. It did so by binding to IL-17R-A, resulting in unstable ventricular electrophysiology in normal structural hearts.

1. Introduction

The Th17 subset of CD4+ T cells has been associated with several adaptive immunological disorders within the nervous system [1]. As a characteristic proinflammatory cytokine of Th17 cells, interleukin (IL)-17A has long been thought to affect several neurological diseases, such as epilepsy [2], multiple sclerosis [3] and ischemic stroke [4], by participating in neuroimmune communications via binding with IL-17 receptor A (IL-17R-A) on neuron surfaces. However, whether IL-17A-induced neuroinflammation regulates the functions of the left stellate ganglion (LSG) remains unclear.

LSG, the sympathetic ganglion of the cardiac autonomic nervous system, plays an important role in the initiation and maintenance of ventricular arrhythmia (VA) [5–7]. Previous studies have shown that the etiology of VA can be attributed to LSG neuroinflammation-induced neuronal overactivity [8–10]. In addition, our previous study demonstrated that IL-1β can induce LSG remodeling by regulating neuroinflammation to promote VA under acute ischemic stress conditions [11].

Prior studies on sudden cardiac death have mainly been performed using a combination of stressors, such as ischemia with activation of the LSG [12]. However, in certain normal structural hearts, increased sympathetic activity may also lead to lethal VA [13]. In the present study, we estimated the effect of IL-17A repletion on LSG activation and susceptibility to VA via neuroimmune communication. Our understanding of the effect of IL-17A on ventricular electrophysiological

* Corresponding author at: Department of Cardiology, Renmin Hospital of Wuhan University, No. 238 Jiefang Road, Wuchang District, Wuhan City, Hubei Province 430060, China.
E-mail address: whujianghong@163.com (H. Jiang).
[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.ijcard.2019.01.010
0167-5273/

properties (EPs) may provide possible mechanistic insights into the incidence of clinical malignant VA events in normal structural hearts.

2. Materials and methods

2.1. Animal preparation and surgical procedures

All animal studies were reviewed and approved by the animal experimental administration of Wuhan University and performed in accordance with the recommendations in the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health. 24 adult beagle dogs weighing between 10 and 12 kg were anesthetized with 3% Na-pentobarbital at an initial dose of 1 ml/kg followed by a maintenance dose of 2 ml/h. All dogs were intubated and ventilated with a positive pressure ventilator. Normal saline was delivered to the right femoral veins to resupply any spontaneous fluid loss. The left femoral artery was dissected, and an electrode catheter was inserted into the artery to record the systemic arterial pressure. The core body temperature was maintained at approximately 36.5 °C via a heating pad. The heart rate (HR), blood pressure (BP), and standard electrocardiogram leads were continuously monitored throughout the procedure via a computer-based apparatus (Lead 7000, Jinjiang Inc., Chengdu, China). All efforts were made to minimize animal suffering. A left thoracotomy was performed at the third intercostal space to carefully expose the LSG. Then, a unilateral thoracotomy was performed at the fourth and fifth intercostal space. Multielectrode catheters were sutured to three different sites, including the left ventricular apex (LVA), left ventricular base (LVB), and median area of the left ventricle (LVM), to measure ventricular EPs. All methods and procedures were performed as described in our previous studies [11,14,15].

2.2. Experimental design

Adult beagle dogs were randomly divided into three groups as follows: the control group ($n = 8$) was microinjected with 0.1 ml phosphate buffer saline (PBS) containing 0.1% albumin into the LSG, the IL-17A group ($n = 8$) was microinjected with recombinant canine IL-17A (Sino Biological Inc., Beijing, China) diluted in PBS containing 0.1% albumin (0.1 ml, 25 μg/ml) into the LSG, and the IL-17A antagonist plus IL-17A group ($n = 8$) was microinjected with anti-IL-17A monoclonal antibody (anti-IL-17A mAb, Biochempartner, Shanghai, China, 2.5 mg/ml) in volume of 0.1 ml 30 min before IL-17A administration. The neural activity and neural function of the LSG and ventricular EPs were measured at different sites at baseline and 30 min postinjection. In addition, LSG tissues were subsequently harvested to conduct biological and pathological examinations. The flow chart protocol for these experiments is outlined in Fig. 1.

2.3. Microinjection of IL-17A and anti-IL-17A mAb into the LSG

A left thoracotomy was performed to expose the LSG, and 0.1 ml of a 25 μg/ml solution of IL-17A was microinjected into the LSG under direct vision. The entire administration procedure was completed within 1 min. The dose of IL-17A chosen for the present study was based on findings reported by Frank Richter [16], who found that performing an intraarticular injection of 50 ng of recombinant mouse IL-17A sensitizes the joint afferents. We calculated the equivalent dose for our study species based on body surface area [17] and determined that the equivalent dosage of IL-17A in beagles weighing 10–12 kg should be approximately 4 μg. However, we ultimately applied a relatively lower dosage of 2.5 μg in a volume of 0.1 ml to the LSG according to pre-experimental data because we were performing a very local injection. As for the concentration of anti-IL-17A mAb, 2.5 mg/ml was used according to our previous study [10].

2.4. Examination of ventricular EPs

The ventricular effective refractory period (ERP) was recorded at LVA, LVB, and LVM at baseline and 30 min after the microinjection of the IL-17A or PBS solution. The ERP at each site was determined by a programmed stimulation that consisted of 8 consecutive stimuli (S_1-S_1 = 330 ms cycle length) followed by a premature stimulus (S_2). The coupling S_1-S_2 interval was progressively decreased from an initial 250 ms in increments of 10 ms and then 2 ms until ERP was reached. ERP was defined as the longest S_1-S_2 interval that failed to capture the ventricles. The ERP dispersion was defined as the coefficient of variation (CV) of the ERP [18].

Monophasic action potentials were recorded in the left ventricle at the same sites as the ERP. A dynamic steady-state pacing protocol (S_1-S_1) was performed to determine the action potential duration (APD) restitution properties with a LEAD 7000 workstation system (Lead 7000, Jinjiang). The pulse train was delivered at an initial S_1-S_1 that was slightly shorter than the sinus cycle length and was continued for 30 s to achieve a steady state. The subsequent pulse train was delivered in decreasing increments of 20 ms until 300 ms was reached and then in 10 ms increments until an interval that induced the occurrence of APD alternans was reached. The APD at 90% repolarization was set as APD_{90}. To construct the APD restitution, each APD_{90} against the diastolic interval (DI) was plotted. APD_{90} and DI were measured at baseline and after each 10 ms decrease in pacing cycle length. The maximal slope (S_{max}) of the restitution curve was determined at the shortest DI. The spatial dispersion of the APD_{90} was calculated offline as the CV of the APD_{90} among the three recording sites [18].

2.5. Direct LSG function and neural activity recordings

LSG function and neural activity were measured to indicate LSG activation according to our previous studies [14,15]. Briefly, high-frequency stimulation (HFS) (20 Hz, 0.1 ms pulse duration) was delivered by a Grass-S88 stimulator (Astro-Med, West Warwick, Rhode Island, USA) to the LSG. Maximal systolic blood pressure (SBP) changes were used to reflect LSG function in response to HFS at voltages of 5, 10, 15, 20, and 25 V. LSG neural activity was recorded at baseline and 30 min after microinjection using a pair of coated tungsten electrodes inserted into the LSG and a ground lead connected to the chest wall. Data were analyzed with a PowerLab data acquisition system (AD Instruments, New South Wales, Australia). Signal channel filters were set to range from 50 Hz-1 kHz.

2.6. Quantitative real-time PCR (RT-PCR) and western blot analysis

At the end of the experiment, LSG tissues were immediately harvested, dissected into small portions and stored at −80 °C until assayed. Total RNA was extracted using TRIzol reagent (Invitrogen, Carlsbad, US) and then reverse-transcribed to cDNA using a PrimeScrip RT reagent Kit with gDNA Eraser (Takara Bio, Otsu, Japan) according to the manufacturer's instructions. Quantitative polymerase chain reaction (qPCR) was performed according to the manufacturers' instructions using SYBR Premix Ex Taq (Takara Bio, Otsu, Japan) with specific primers (Genecreate, Wuhan, China) on a StepOne Real-Time PCR analyzer (Life Technologies, Carlsbad, US) under the following conditions: initial denaturation for 1 min at 95 °C followed by 40 cycles of denaturation for 15 s at 95 °C, annealing for 20 s at 58 °C, and extension for 45 s at 72 °C.

Tissues used for western blot analysis were washed with PBS, homogenized to extract total protein, and then loaded onto gels for SDS-PAGE (ASPEN, Wuhan, China). After the gels were run, the proteins were transferred to polyvinylidene fluoride membranes (Millipore). The membranes were blocked and then incubated overnight at 4 °C with primary antibodies against c-fos (Santa Cruz Biotechnology, Dallas, Texas, USA), p-TrkA (Absin, Shanghai, China), nuclear factor NFκB activator 1 (ACT1, Absin, Shanghai, China), p65 (Abcam, Cambridge, England), and phosphorylated p65 (p-p65, Abcam, Cambridge, England). Protein signals were standardized to β-actin (Abcam, Cambridge, England).

2.7. Histopathological staining

At the end of the experiment, fresh LSG tissues were harvested and immediately fixed in 4% paraformaldehyde at room temperature. Double immunofluorescence staining was used to examine the expression profiles of neuronal IL-17R-A (Abcam, Cambridge, England) and tyrosine hydroxylase (TH, Servicebio, Wuhan, China), an indicator of sympathetic innervation. Tissues were also co-stained for c-fos and TH (Proteintech, Wuhan, China) to determine the severity of neuronal remodeling. All immunoreactivity results were quantified using commercial software (ImagePro; Media Cybernetics, Inc., Rockville, Maryland).

2.8. Statistical analysis

All data were expressed as the means ± standard error of measurement, and Prism 7.0 (GraphPad Software, Inc., La Jolla, CA, USA) was used for all analyses. Two-way ANOVA was used for comparisons of variables measured at different time points between three groups. One-way ANOVA were used to compare the means between three groups. $P < 0.05$ was considered statistically significant.

3. Results

3.1. Effect of IL-17A on ventricular EPs

Ventricular EP indexes, including ventricular ERP, APD, dispersion of ERP, APD and APD restitution properties, were evaluated in three groups. As shown in Fig. 1B–F, 30 min after microinjecting IL-17A into the LSG significantly decreased ERP and APD_{90} but increased the dispersion of ERP, APD_{90}, and APD alternans cycle length compared to baseline, indicating that APD alternans occurred more easily during the dynamic pacing protocol. In addition, IL-17A also increased the S_{max} of the restitution curve at all three sites and the dispersion of S_{max} ($P < 0.05$). While pre-injection of anti-IL-17A mAb significantly attenuated the changes induced by IL-17A ($P < 0.05$). No obvious changes were observed in the control group ($P > 0.05$).

3.2. Effect of IL-17A on the neural function and activity of the LSG

LSG function was estimated as an indicator of sympathetic activity that is consistent with neural activity. Microinjecting IL-17A into the LSG significantly enhanced LSG functions, as reflected by the fact that the maximal SBP induced when the LSG was electrically stimulated at different voltage levels (10, 15, 20, and 25 V) was significantly higher than at baseline ($P < 0.05$) (Fig. 2B). Conversely, no obvious changes

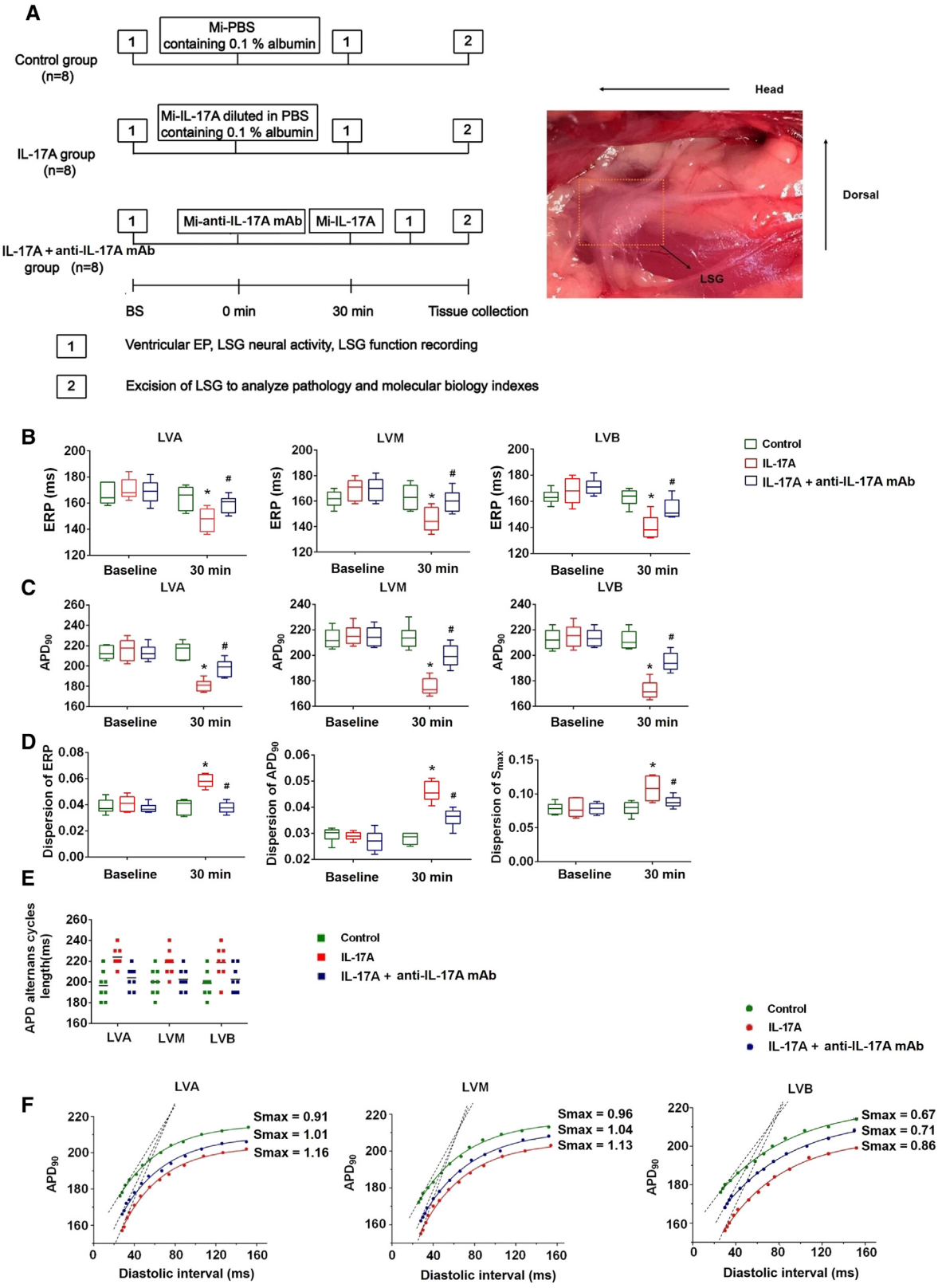

Fig. 1. The effect of IL-17A administration on ventricular EPs. The flow chart of the present study and the location of the LSG (A). The ERP (B), APD_{90} (C) were shortened, while the dispersion of ERP, APD_{90} and S_{max} (D), the pacing cycle length of APD alternans (E), and the S_{max} of the restitution curve (F) were increased at each site in normal structural hearts after IL-17A administration. These effects were attenuated by pre-injection of anti-IL-17A mAb. * $P < 0.05$ vs baseline in IL-17A-treated group, # $P < 0.05$ vs 30 min in IL-17A group. APD_{90} = action potential duration at 90% repolarization, EPs = electrophysiological properties, ERP = effective refractory period, Mi-IL-17A = microinjection of interleukin-17A, LSG = left stellate ganglion. LVA = left ventricular apex, LVB = left ventricular base, LVM = median area of the left ventricle, Mi-PBS = microinjection of phosphate buffer saline, S_{max} = the maximal slope of the restitution curve.

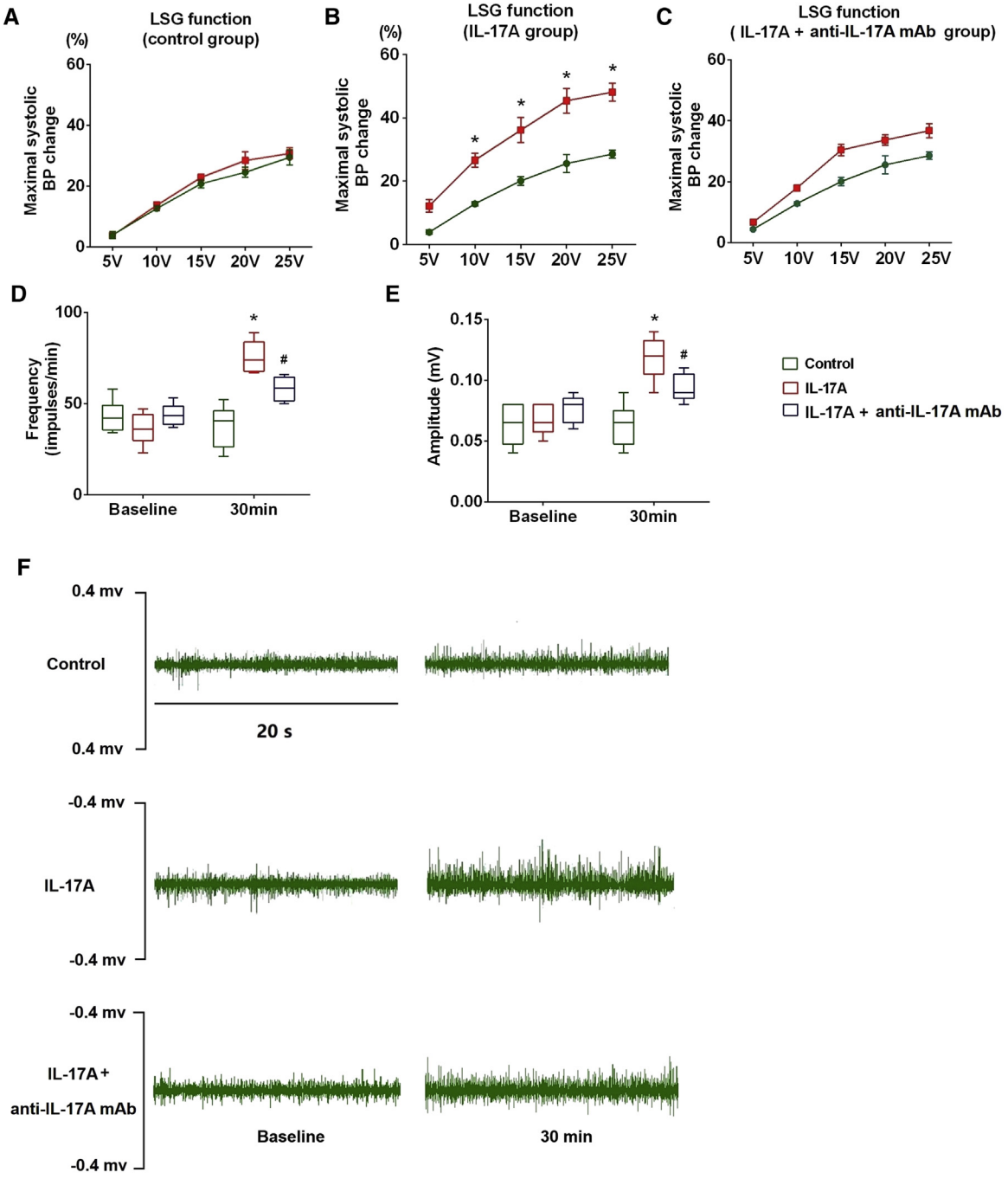

Fig. 2. The effect of IL-17A administration on LSG function and neural activity. The maximal SBP showed no significant changes in the control groups (A), but significant increase in IL-17A-treated groups (B), and less increase in the pre-injection of anti-IL-17A mAb group (C). The neural recordings from LSG including frequency (D) and amplitude (E) both were increased in the IL-17A-treated groups which were reversed by pre-injection of anti-IL-17A mAb. Typical graphic examples of LSG neural activity recorded in the control group, IL-17A-treated groups and pre-injection of anti-IL-17A mAb group (F). * $P < 0.05$ vs baseline in IL-17A-treated group, # $P < 0.05$ vs 30 min in IL-17A-treated group. SBP = systolic blood pressure, LSG = left stellate ganglion.

were observed in the voltage-BP response curve in the control group or pre-injection of anti-IL-17A mAb group ($P > 0.05$) (Fig. 2A and C). LSG neural activity was measured as an indicator of sympathetic activity. A quantitative analysis of LSG neural activity (Fig. 2D and E) clearly showed that microinjecting IL-17A into the LSG markedly increased the frequency and amplitude of LSG neural activity at 30 min after administration ($P < 0.05$). While pre-injection of anti-IL-17A mAb significantly attenuated the increases induced by IL-17A ($P < 0.05$). However, no significant changes were observed in the control group ($P > 0.05$). Typical graphic examples of the LSG neural activity recorded in the three groups at baseline and 30 min after microinjection are shown in Fig. 2F.

3.3. Effect of IL-17A on the expression of proinflammatory cytokines in the LSG

IL-17A-elicited inflammation in the LSG was measured by determining the messenger ribonucleic acid (mRNA) expression levels of proinflammatory cytokines. The results showed that the mRNA level of tumor necrosis factor-α (TNF-α) within the LSG at 30 min after

administration was higher in the IL-17A-treated group than in the control group ($P < 0.05$) (Fig. 3A). While pre-injection of anti-IL-17A mAb attenuated the increased inflammatory response induced by IL-17A ($P < 0.05$). As expected, the mRNA expression levels of IL-6 and IL-1β within the LSG exhibited behavior similar to that observed for TNF-α (Fig. 3B and C). Typical images of inflammatory cell infiltration are shown in Fig. 3D.

3.4. IL-17A upregulated p-TrkA and c-fos expression in the LSG

The levels of neuron-associated proteins, including neuropeptides and neurotransmitters, were measured to evaluate the extent of remodeling in the LSG. P-TrkA is the activated receptor for nerve growth factor (NGF), which contributes to remodeling in sympathetic neurons, and c-

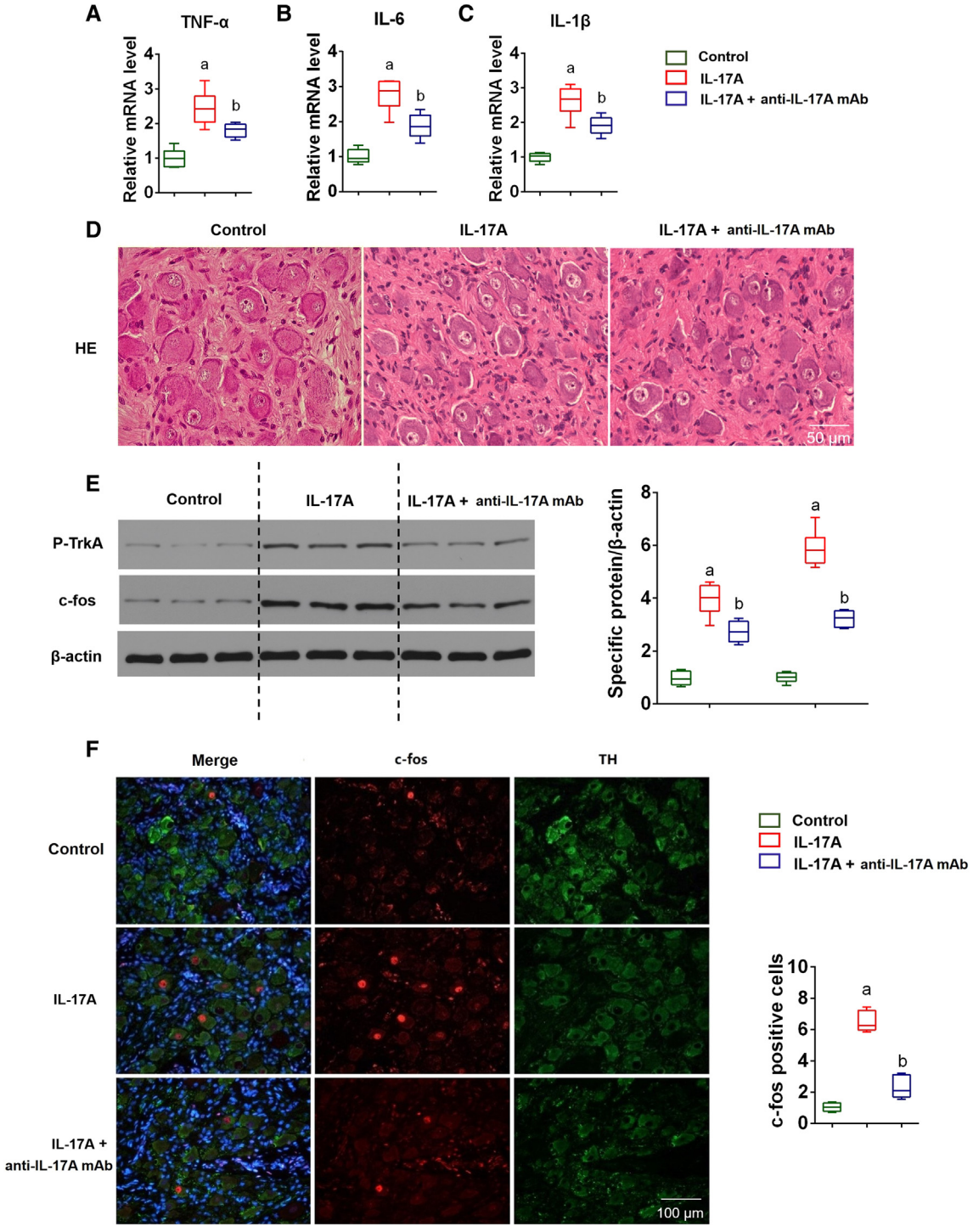

Fig. 3. The effect of IL-17A administration on expressions of inflammatory substances and neuropeptides of LSG. The expression of TNF-α (A), IL-6 (B), and IL-1β (C) were significantly upregulated in the IL-17A-treated groups. An image is shown to demonstrate the typical increased levels of inflammatory cells (D) within the LSG in the IL-17A-treated groups (a portion of the LSG is shown at 400×). Representative bands and quantitative analysis of p-TrkA and c-fos expression (E), representative examples and quantitative analysis of c-fos (red) and TH (green) immunofluorescence staining (F) which showed great increase of neuropeptide expression in the IL-17A-treated groups (a portion of the LSG is shown at 200×). [a] $P < 0.05$ vs control group, [b] $P < 0.05$ vs IL-17A-treated group. HE staining = hematoxylin-eosin staining, mRNA = messenger ribonucleic acid, p-TrkA = phosphorylated Tyr kinase A, TH = tyrosine hydroxylase.

fos is a marker that indicates the level of activation of sympathetic neurons. As shown by the results of western blot analysis, the expression levels of p-TrkA and c-fos were dramatically higher in the IL-17A-administered group than in the control group ($P < 0.05$) (Fig. 3E). While pre-injection of anti-IL-17A mAb significantly attenuated the increases induced by IL-17A ($P < 0.05$). Double-immunofluorescence staining for c-fos (red) and TH (green) within the LSG further indicated that the LSG was activated by IL-17A administration ($P < 0.05$) (Fig. 3F). However, the increased p-TrkA and c-fos expressions induced by IL-17A were attenuated by pre-injection of anti-IL-17A mAb ($P < 0.05$).

3.5. IL-17A binds to IL-17R-A to activate ACT1 and the NFκB-p65 pathway in the LSG

IL-17R-A expression was evaluated within the LSG using immunofluorescence. Fig. 4A shows neurons within the LSG stained for only IL-17R-A. The downstream effects of IL-17R-A are mediated by the cytosolic adaptor molecule ACT1, which subsequently activates the downstream proteins p65 and p-p65 [19]. As shown in Fig. 4B, the expression levels of ACT1 and p-p65 were significantly higher in the IL-17A administration group than in the control group ($P < 0.05$). However, the increased ACT1 and p-p65 expressions induced by IL-17A were attenuated by pre-injection of anti-IL-17A mAb ($P < 0.05$).

4. Discussion

4.1. IL-17A aggravated ventricular electrophysiological remodeling

Previous studies have suggested that neuroinflammation in the LSG may promote cardiac sympathetic output to exert adverse effects on ventricular electrophysiology, such as shortening the ventricular ERP and APD_{90}. However, when these alterations are induced by certain pro-inflammatory factors, they can be attenuated by preadministration of their receptor antagonist, thus suppressing the incidence of VA [11]. In the present study, we showed that ventricular ERP and APD_{90} were significantly lower after IL-17A administration but were reversed by pre-injection of anti-IL-17A mAb, indicating a potential role of IL-17A in the initiation of VA. It was previously reported that maximizing spatial dispersions effectively induced ventricular fibrillation (VF) [20]. Conversely, decreasing dispersions produced beneficial effects on VF [21]. In the present study, we showed that the dispersions observed in ventricular ERP, APD and APD restitution kinetics were significantly increased by IL-17A administration, and the increase was attenuated by pre-injection of anti-IL-17A mAb, suggesting that IL-17A is capable of promoting VA. The restitution hypothesis proposes that a steep restitution slope indicates a high risk of VF. Conversely, a flattened restitution curve indicated a protective effect [22]. Various interventions aimed at reducing the restitution slope have been implemented as therapeutic approaches to inhibit the risk of VA [23,24]. Our results showed that IL-17A steepened the restitution curve and facilitated APD alternans, while pre-injection of anti-IL-17A attenuated the changes, indicating that IL-17A may play a role in promoting the occurrence of malignant VA.

4.2. IL-17A aggravated LSG remodeling

Studies performed over many years have shown that IL-17A-mediated neuroinflammation may aggravate the remodeling of the sympathetic nervous system. For instance, recent studies have demonstrated that IL-17A exerts a neurotrophic effect to aggravate the remodeling of sympathetic neurites [25]. Similarly, patients with inflammatory bowel disease were found to have increased sympathetic innervation in the intestines, which are regulated by Th17-mediated neuroinflammation [26]. In the present study, IL-17A promoted sympathetic tone, as shown by the increased neural function and activity observed in the LSG following IL-17A microinjection, which were attenuated by pre-injection of anti-IL-17A mAb.

Cardiovascular diseases and sympathetic nerve also communicate in the humoral pathway [27]. Neuropeptides and neurotransmitters contribute to LSG remodeling. NGF plays a crucial role in sympathetic remodeling by binding to TrkA receptors at axon terminals. After NGF activates its receptor, the resulting NGF–phospho-TrkA complex is retrogradely transported back to the neuronal cell bodies [28]. In the present study, we found that IL-17A upregulated the expression of p-TrkA and c-fos to aggravate pathological remodeling in the LSG, and this effect may also contribute to cardiac sympathetic nerve sprouting [7,28]. More importantly, the increased p-TrkA and c-fos in the LSG was attenuated by pre-injection of anti-IL-17A mAb.

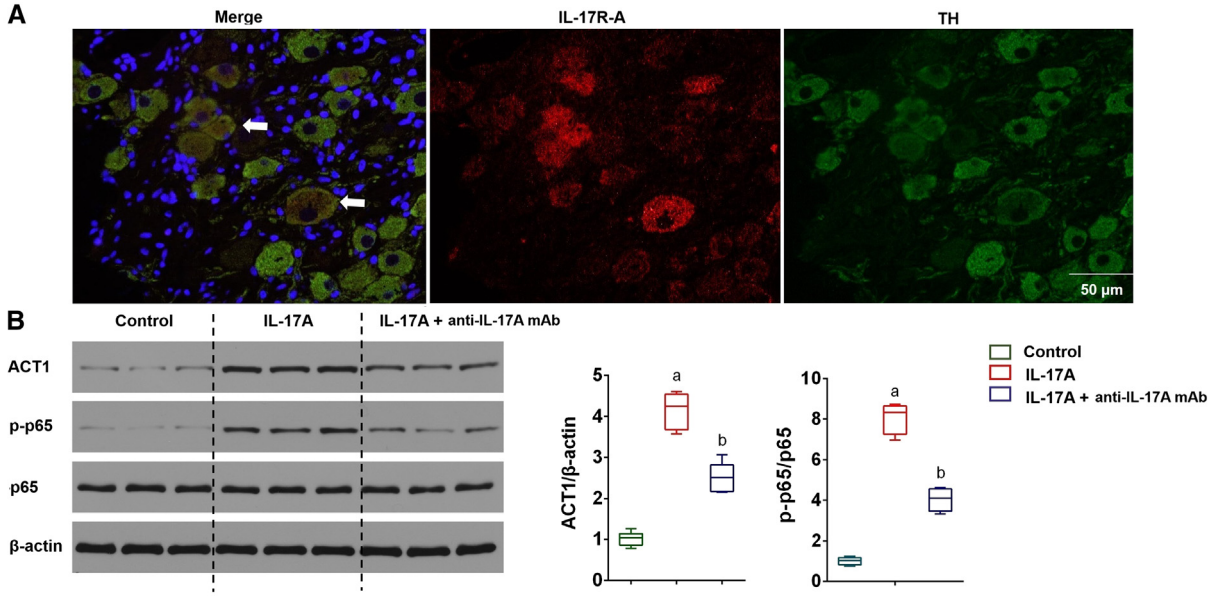

Fig. 4. The effect of IL-17A on IL-17R-A signaling in the LSG. Representative images of double immunofluorescence labeling for IL-17-R-A with TH expression in the LSG (a portion of the LSG is shown at 400×) (arrows in A). Representative images and quantitative analysis of the results of western blot analysis for ACT1, p65, and p-p65 (B). The expression of ACT1 and p-p65 were significantly increased in the IL-17A-treated groups. [a] $P < 0.05$ vs control group, [b] $P < 0.05$ vs IL-17A-treated group. ACT1 = nuclear factor NFκB activator 1, p-p65 = phosphorylated p65.

4.3. Potential mechanisms for IL-17A- and IL-17R-A-mediated LSG remodeling

Several lines of evidence have indicated that IL-17A, as a proinflammatory mediator, participates in neuroimmune interactions via binding with IL-17R-A [2–4]. While IL-17A commonly binds to IL-17R-A and IL-17R-C, it has a higher affinity for IL-17R-A than for IL-17R-C. Moreover, the tissue distributions of IL-17R-A and IL-17R-C are different, and this may result in functional specialization. Unlike other IL-17Rs, IL-17R-A is broadly expressed on neural tissues and is activated during inflammation, indicating a potential role for neuronal IL-17R-A signaling [29,30]. In the central nervous system, IL-17R-A is clearly expressed within brain structures such as the cortical layers [31], hippocampal neurons [32], and spinal cord [33]. Moreover, approximately 95% of the TH-positive postganglionic neurons within the superior mesenteric ganglia express IL-17R-A [25]. Furthermore, IL-17A also targets a diverse array of immune cells, including T cells, and B cells, to actively promote T cell priming and the production of antibodies and proinflammatory cytokines [34]. These effects need to be further investigated within the LSG.

In the present study, we demonstrated that IL-17R-A was expressed on neurons and other cells within the LSG. As the Supplementary figure shows, the increased expression of ACT1 and p65 in the IL-17A group further suggested that binding of IL-17A to IL-17R-A recruited ACT1 to the IL-17R (SEFIR) domain, thus activating the NFκB-p65 subunit to produce a variety of proinflammatory cytokines, including TNF-α, IL-1β and IL-6. In addition, IL-17A facilitated the migration of proinflammatory cells to the local region, further promoting LSG remodeling.

4.4. Clinical applications

The microinjection of IL-17A into the LSG aimed to simulate several pathological conditions along with an acute elevation in peripheral IL-17A, which can affect its concentration in the LSG. Several common diseases such as acute coronary syndrome [35] and epileptic seizure [2], can increase the release of IL-17A. Similarly, a growing amount of data indicate that susceptibility to arrhythmia is significantly increased in some of these diseases [36,37]. A trigger and a substrate form the basis for provoking the majority of lethal arrhythmias [38]. The results of the present study suggest several reasons why normal structural hearts without arrhythmogenic substrates that are subjected to high levels of IL-17A are susceptible to VA. And the treatment target excess IL-17A in normal structural hearts with IL-17A antagonist was capable to stabilize the ventricular electrophysiology without changing the structure of the LSG.

4.5. Study limitations

There are several limitations to our study. First, in the present study, we only evaluated the role of IL-17A in normal structural heart conditions, and further studies are therefore needed to determine its effect in tissues under myocardial ischemic stress. Second, the potential mechanisms by which IL-17A-mediated neuroimmune communication occurs were only preliminarily investigated, and further studies should provide us with a better overall and more precise understanding of this crosstalk. In addition, whether IL-17A microinjection into other cardiac ganglia, especially the right stellate ganglion, would exert the same neuronal remodeling-promoting effect remains to be investigated. Finally, all EP recordings were obtained from the epicardium rather than the endocardium of the left ventricle, which did not conform to physiological pacing.

5. Conclusion

In summary, the present study demonstrated that IL-17A administration aggravated LSG remodeling by binding with IL-17R-A to activate downstream inflammatory signaling, thereby contributing to destabilized ventricular EPs. Targeting neuroinflammation-induced LSG remodeling may therefore represent an innovative approach to treating VA in several types of normal structural hearts.

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ijcard.2019.01.010.

Acknowledgement of grant support

This work was supported by grant from National Key R&D Program of China (No. 2017YFC1307800), and grants from National Natural Science Foundation of China (No. 81530011, 81770364, 81570463, 81871486, 81700444, 81600367 and 81600395).

Conflicts of interest

None.

References

[1] M.J. McGeachy, D.J. Cua, Th17 cell differentiation: the long and winding road, Immunity 28 (2008) 445–453.
[2] L.Y. Mao, J. Ding, W.F. Peng, Y. Ma, Y.H. Zhang, W. Fan, et al., Interictal interleukin-17A levels are elevated and correlate with seizure severity of epilepsy patients, Epilepsia 54 (2013) e142–e145.
[3] V. Brucklacher-Waldert, K. Stuerner, M. Kolster, J. Wolthausen, E. Tolosa, Phenotypical and functional characterization of T helper 17 cells in multiple sclerosis, Brain 132 (2009) 3329–3341.
[4] M. Gelderblom, A. Weymar, C. Bernreuther, J. Velden, P. Arunachalam, K. Steinbach, et al., Neutralization of the IL-17 axis diminishes neutrophil invasion and protects from ischemic stroke, Blood 120 (2012) 3793–3802.
[5] P.E. Puddu, R. Jouve, F. Langlet, J.C. Guillen, M. Lanti, A. Reale, Prevention of postischemic ventricular fibrillation late after right or left stellate ganglionectomy in dogs, Circulation 77 (1988) 935–946.
[6] S. Zhou, B.C. Jung, A.Y. Tan, V.Q. Trang, G. Gholmieh, S.W. Han, et al., Spontaneous stellate ganglion nerve activity and ventricular arrhythmia in a canine model of sudden death, Heart Rhythm. 5 (2008) 131–139.
[7] J.M. Cao, L.S. Chen, B.H. KenKnight, T. Ohara, M.H. Lee, J. Tsai, et al., Nerve sprouting and sudden cardiac death, Circ. Res. 86 (2000) 816–821.
[8] D.N. Xanthos, J. Sandkuhler, Neurogenic neuroinflammation: inflammatory CNS reactions in response to neuronal activity, Nat. Rev. Neurosci. 15 (2014) 43–53.
[9] S. Rizzo, C. Basso, D. Troost, E. Aronica, A.C. Frigo, A.H. Driessen, et al., T-cell-mediated inflammatory activity in the stellate ganglia of patients with ion-channel disease and severe ventricular arrhythmias, Circ. Arrhythm. Electrophysiol. 7 (2014) 224–229.
[10] O.A. Ajijola, D.B. Hoover, T.M. Simerly, T.C. Brown, J. Yanagawa, R.M. Biniwale, et al., Inflammation, oxidative stress, and glial cell activation characterize stellate ganglia from humans with electrical storm, JCI Insight 2 (2017).
[11] M. Wang, S. Li, X. Zhou, B. Huang, L. Zhou, X. Li, et al., Increased inflammation promotes ventricular arrhythmia through aggravating left stellate ganglion remodeling in a canine ischemia model, Int. J. Cardiol. 248 (2017) 286–293.
[12] M. Swissa, S. Zhou, I. Gonzalez-Gomez, C.M. Chang, A.C. Lai, A.W. Cates, et al., Long-term subthreshold electrical stimulation of the left stellate ganglion and a canine model of sudden cardiac death, J. Am. Coll. Cardiol. 43 (2004) 858–864.
[13] P.J. Schwartz, A. Zaza, E. Locati, A.J. Moss, Stress and sudden death. The case of the long QT syndrome, Circulation 83 (1991) Ii71–80.
[14] S. Wang, X. Zhou, B. Huang, Z. Wang, L. Zhou, M. Wang, et al., Noninvasive low-frequency electromagnetic stimulation of the left stellate ganglion reduces myocardial infarction-induced ventricular arrhythmia, Sci. Rep. 6 (2016) 30783.
[15] L. Yu, Y. Wang, X. Zhou, B. Huang, M. Wang, X. Li, et al., Leptin injection into the left stellate ganglion augments ischemia-related ventricular arrhythmias via sympathetic nerve activation, Heart Rhythm. 15 (2018) 597–606.
[16] F. Richter, G. Natura, M. Ebbinghaus, G.S. von Banchet, S. Hensellek, C. Konig, et al., Interleukin-17 sensitizes joint nociceptors to mechanical stimuli and contributes to arthritic pain through neuronal interleukin-17 receptors in rodents, Arthritis Rheum. 64 (2012) 4125–4134.
[17] USFDA, Guidance for Industry: Estimating the Maximum Safe Starting Dose in Adult Healthy Volunteer, US Food and Drug Administration, Rockville, MD, 2005.
[18] B. Huang, L. Yu, B. He, S. Wang, Z. Lu, K. Liao, et al., Sympathetic denervation of heart and kidney induces similar effects on ventricular electrophysiological properties, EuroIntervention 11 (2015) 598–604.
[19] W. Lin, N. Wang, K. Zhou, F. Su, Y. Jiang, J. Shou, et al., RKIP mediates autoimmune inflammation by positively regulating IL-17R signaling, EMBO Rep. (2018) e44951.
[20] A.R. Gelzer, M.L. Koller, N.F. Otani, J.J. Fox, M.W. Enyeart, G.J. Hooker, et al., Dynamic mechanism for initiation of ventricular fibrillation in vivo, Circulation 118 (2008) 1123–1129.
[21] B. Huang, L. Yu, B. He, Z. Lu, S. Wang, W. He, et al., Renal sympathetic denervation modulates ventricular electrophysiology and has a protective effect on ischaemia-induced ventricular arrhythmia, Exp. Physiol. 99 (2014) 1467–1477.
[22] M.L. Riccio, M.L. Koller, R.F. Gilmour Jr., Electrical restitution and spatiotemporal organization during ventricular fibrillation, Circ. Res. 84 (1999) 955–963.

[23] Q. Jin, X. Chen, W.M. Smith, R.E. Ideker, J. Huang, Effects of procainamide and sotalol on restitution properties, dispersion of refractoriness, and ventricular fibrillation activation patterns in pigs, J. Cardiovasc. Electrophysiol. 19 (2008) 1090–1097.
[24] G.A. Ng, K.E. Brack, V.H. Patel, J.H. Coote, Autonomic modulation of electrical restitution, alternans and ventricular fibrillation initiation in the isolated heart, Cardiovasc. Res. 73 (2007) 750–760.
[25] D.M. Moynes, S.J. Vanner, A.E. Lomax, Participation of interleukin 17A in neuroimmune interactions, Brain Behav. Immun. 41 (2014) 1–9.
[26] I. Monteleone, M. Sarra, F. Pallone, G. Monteleone, Th17-related cytokines in inflammatory bowel diseases: friends or foes? Curr. Mol. Med. 12 (2012) 592–597.
[27] B.A. Habecker, M.E. Anderson, S.J. Birren, K. Fukuda, N. Herring, D.B. Hoover, et al., Molecular and cellular neurocardiology: development, and cellular and molecular adaptations to heart disease, J. Physiol. 594 (2016) 3853–3875.
[28] B.A. Tsui-Pierchala, D.D. Ginty, Characterization of an NGF-P-TrkA retrograde-signaling complex and age-dependent regulation of TrkA phosphorylation in sympathetic neurons, J. Neurosci. 19 (1999) 8207–8218.
[29] S.G. Hymowitz, E.H. Filvaroff, J.P. Yin, J. Lee, L. Cai, P. Risser, et al., IL-17s adopt a cystine knot fold: structure and activity of a novel cytokine, IL-17F, and implications for receptor binding, EMBO J. 20 (2001) 5332–5341.
[30] R.E. Kuestner, D.W. Taft, A. Haran, C.S. Brandt, T. Brender, K. Lum, et al., Identification of the IL-17 receptor related molecule IL-17RC as the receptor for IL-17F, J. Immunol. 179 (2007) 5462–5473.
[31] J.J. He, S. Li, H.F. Shu, S.X. Yu, S.Y. Liu, Q. Yin, et al., The interleukin 17 system in cortical lesions in focal cortical dysplasias, J. Neuropathol. Exp. Neurol. 72 (2013) 152–163.
[32] D.D. Wang, Y.F. Zhao, G.Y. Wang, B. Sun, Q.F. Kong, K. Zhao, et al., IL-17 potentiates neuronal injury induced by oxygen-glucose deprivation and affects neuronal IL-17 receptor expression, J. Neuroimmunol. 212 (2009) 17–25.
[33] X. Meng, Y. Zhang, L. Lao, R. Saito, A. Li, C.M. Backman, et al., Spinal interleukin-17 promotes thermal hyperalgesia and NMDA NR1 phosphorylation in an inflammatory pain rat model, Pain 154 (2013) 294–305.
[34] Y. Liu, R.C. Ho, A. Mak, The role of interleukin (IL)-17 in anxiety and depression of patients with rheumatoid arthritis, Int. J. Rheum. Dis. 15 (2012) 183–187.
[35] X. Cheng, X. Yu, Y.J. Ding, Q.Q. Fu, J.J. Xie, T.T. Tang, et al., The Th17/Treg imbalance in patients with acute coronary syndrome, Clin. Immunol. 127 (2008) 89–97.
[36] P. Sanchez-Borque, B. Gonzalez-Giraldez, J. Benezet-Mazuecos, A. Miracle, J. Crosa, J. M. Rubio, Ictal asystole: a condition between neurology and cardiology, Int. J. Cardiol. (2018 Sep 27), https://doi.org/10.1016/j.ijcard.2018.09.087, pii: S0167-5273(18)34628-X.
[37] J.N. Weiss, K. Nademanee, W.G. Stevenson, B. Singh, Ventricular arrhythmias in ischemic heart disease, Ann. Intern. Med. 114 (1991) 784–797.
[38] G.R. Sutherland, Sudden cardiac death: the pro-arrhythmic interaction of an acute loading with an underlying substrate, Eur. Heart J. 38 (2017) 2986–2994.