Long-term intermittent high-amplitude subcutaneous nerve stimulation reduces sympathetic tone in ambulatory dogs

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BACKGROUND Reducing sympathetic efferent outflow from the stellate ganglia (SG) may be antiarrhythmic.

OBJECTIVE The purpose of this study was to test the hypothesis that chronic thoracic subcutaneous nerve stimulation (ScNS) could reduce SG nerve activity (SGNA) and control paroxysmal atrial tachycardia (PAT).

METHODS Thoracic ScNS was performed in 8 dogs while SGNA, vagal nerve activity (VNA), and subcutaneous nerve activity (ScNA) were monitored. An additional 3 dogs were used for sham stimulation as controls.

RESULTS Xinshu ScNS and left lateral thoracic nerve ScNS reduced heart rate (HR). Xinshu ScNS at 3.5 mA for 2 weeks reduced mean average SGNA from 5.32 μV (95% confidence interval [CI] 3.89–6.75) at baseline to 3.24 μV (95% CI 2.16–4.31; $P = .015$) and mean HR from 89 bpm (95% CI 80–98) at baseline to 83 bpm (95% CI 76–90; $P = .007$). Bilateral SG showed regions of decreased tyrosine hydroxylase staining with increased terminal deoxynucleotidyl transferase dUTP nick-end labeling–positive nuclei in 18.47% (95% CI 9.68–46.62) of all ganglion cells, indicating cell death. Spontaneous PAT episodes were reduced from 9.83 per day (95% CI 5.77–13.89) in controls to 3.00 per day (95% CI 0.11–5.89) after ScNS ($P = .027$). Left lateral thoracic nerve ScNS also led to significant bilateral SG neuronal death and significantly reduced average SGNA and HR in dogs.

CONCLUSION ScNS at 2 different sites in the thorax led to SG cell death, reduced SGNA, and suppressed PAT in ambulatory dogs.

KEYWORDS Arrhythmia; Autonomic nervous system; Nerve Recording; Neuromodulation; Stellate ganglion

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Neuromodulation methods that reduce sympathetic efferent outflow may be helpful in controlling cardiac arrhythmia.[1] However, other than surgical resection of stellate ganglia (SG) to achieve permanent left sympathetic denervation, no less invasive method is available to effectively inhibit sympathetic nerve output to the heart. We previously reported that vagal nerve stimulation (VNS) could effectively induce left stellate ganglion (LSG) remodeling and suppress stellate ganglion nerve activity (SGNA).[2] Nonetheless, clinical applications of VNS for arrhythmia control are often limited by the technical difficulties and potential

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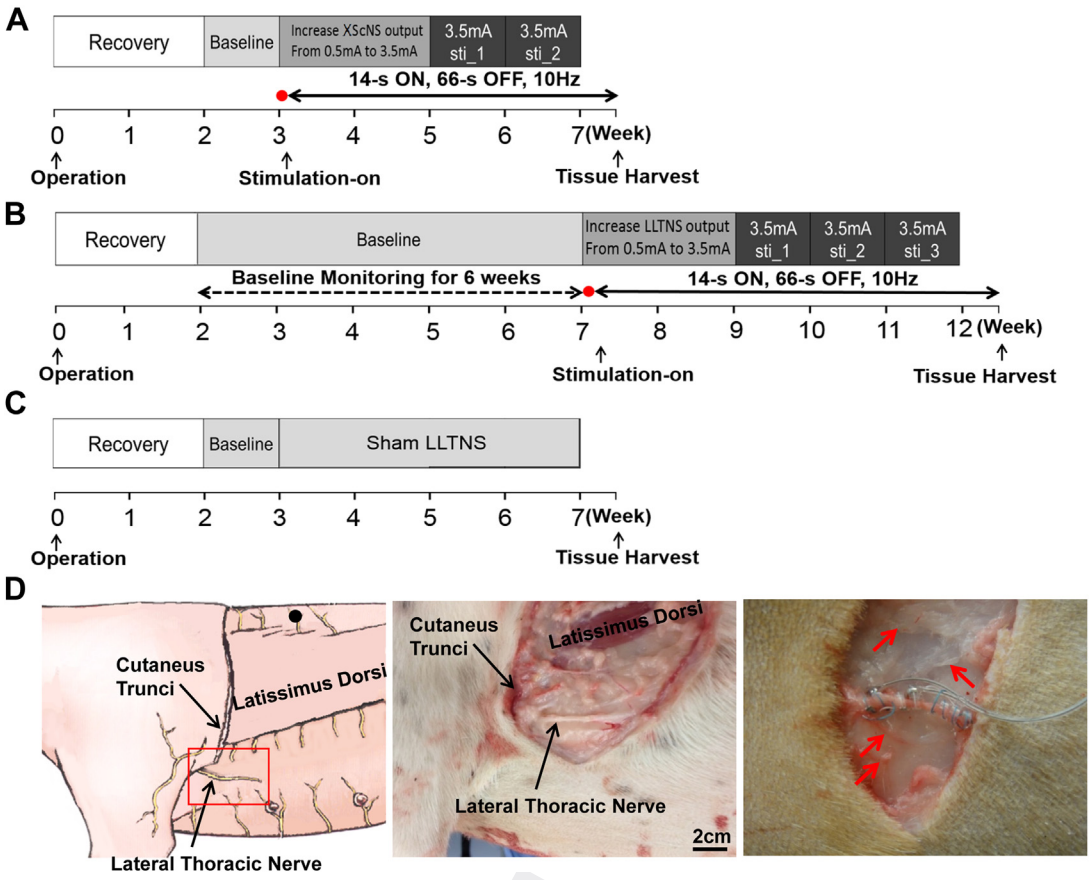

Figure 1 The study protocols. **A:** Protocol 1, Xinshu subcutaneous nerve stimulation (n = 6). After baseline recording, the neurostimulator was turned on (*red dot*) and programmed 14-s ON (10 Hz, 500-μs pulse duration) and 66-s OFF. Output current was increased gradually from 0.5 to 3.5 mA in 2 weeks. After an additional 2 weeks of stimulation, the dogs were euthanized. **B:** Protocol 2, left lateral thoracic nerve stimulation (n = 2). Baseline recording extended to 6 weeks. The neurostimulator was turned on at week 8, and output was gradually increased to 3.5 mA over a 2-week period. After an additional 3 weeks of stimulation at 3.5 mA, the dogs were euthanized. **C:** Protocol 3, left lateral thoracic nerve sham stimulation (n = 3). **D:** Anatomy of Xinshu acupoint and left lateral thoracic nerve (LLTN). **Left:** *Black dot* indicates site of Xinshu acupoint. **Middle:** Incision at the upper portion of the red box reveals LLTN beneath the cutaneus trunci. **Right:** Electrodes wrapped around the LLTN. *Red arrows* point to subcutaneous nerves, which are also found at the Xinshu acupoint.

complications associated with surgical implantation of the vagal nerve stimulators. Just like vagal nerves, subcutaneous nerves in dogs contain sympathetic components.[3] The postganglionic sympathetic nerve fibers of the neck and thorax came primarily from the SG.[4] Thoracic subcutaneous nerve activity (ScNA) and superficial skin sympathetic nerve activity closely correlate with the SGNA,[3,5,6] further supporting a connection between thoracic subcutaneous nerves and the SG. We hypothesized that thoracic subcutaneous nerve stimulation (ScNS) can rapidly excite the SG and in turn cause neurotoxicity in the SG, which would reduce its sympathetic outflow. We first performed stimulation at the Xinshu acupoint (BL15, approximately 5 cm lateral to the spine at T5 level) because Xinshu acupoint stimulation has been reported to change mRNA expression in sympathetic ganglia in rats,[7] and acupuncture at the Xinshu acupoint has prevented atrial fibrillation recurrence after cardioversion in humans.[8] We also stimulated the left lateral thoracic nerve (LLTN) in additional dogs to determine whether stimulation outside an acupoint can achieve the same antiarrhythmic effects. A sham control group was studied for comparison. We designed this study to test the hypothesis that long-term rapid ScNS may cause neuronal death in the SG, leading to reduced sympathetic outflow and decreased spontaneous paroxysmal atrial tachyarrhythmias (PAT) in ambulatory dogs.

Methods

The animal protocol was approved by the Institutional Animal Care and Use Committee and conformed to the Guide for Care and Use of Laboratory Animals. Figure 1 summarizes the study protocols and the location of electrode implantation. A detailed experimental method is included in the Supplementary Methods.

Protocol 1: The effect of Xinshu acupoint stimulation

A total of 6 mongrel dogs were studied. A radiotransmitter (D70EEE, Data Sciences International, St. Paul, MN) was implanted to record SGNA, vagal nerve activity (VNA), and ScNA. A Cyberonics model 304 bipolar vagal stimulating lead (Cyberonics Inc, Houston, TX) was implanted around subcutaneous nerves at Xinshu acupoint and connected to a Cyberonics Demipulse neurostimulator. After