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Subcutaneous Nerve Activity and Spontaneous Ventricular Arrhythmias in Ambulatory Dogs

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Abstract

Background—Stellate ganglion nerve activity (SGNA) is important in ventricular arrhythmogenesis. However, because thoracotomy is needed to access the stellate ganglion, it is difficult to use SGNA for risk stratification.

Objective—To test the hypothesis that subcutaneous nerve activity (SCNA) in canines can be used to estimate SGNA and predict ventricular arrhythmia.

Methods—We implanted radio transmitters to continuously monitor left stellate ganglion and subcutaneous electrical activities in 7 ambulatory dogs with myocardial infarction, complete heart block and nerve growth factor infusion to the left stellate ganglion.

Results—Spontaneous ventricular tachycardia (VT) or ventricular fibrillation (VF) was documented in each dog. SCNA preceded a combined 61 episodes of VT and VF, 61 frequent bigeminy or couplets and 61 premature ventricular contractions within 15 s in 70%, 59% and 61% of arrhythmias, respectively. Similar incidence of 75%, 69% and 62% was noted for SGNA. Progressive increase in SCNA (48.9 (95% CI 39.3–58.5) vs. 61.8 (95% CI 45.9–77.6) vs. 75.1 (95% CI 57.5–92.7) mV-s) and SGNA (48.6 (95% CI 40.9–56.3) vs. 58.5 (95% CI 47.5–69.4) vs.

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69.0 (95% CI 53.8–84.2) mV-s) integrated over 20 s intervals was demonstrated 60 s, 40 s and 20 s prior to VT/VF ($p < 0.05$). The Pearson's correlation coefficient for integrated SCNA and SGNA was 0.73 ± 0.18 ($p < 0.0001$ for all dogs, $n = 5$). Both SCNA and SGNA exhibited circadian variation.

Conclusions—SCNA can be used as an estimate of SGNA to predict susceptibility to VT and VF in a canine model of ventricular arrhythmia and sudden cardiac death.

Keywords

atrioventricular block; autonomic nervous system; myocardial infarction; sudden cardiac death; ventricular arrhythmia

Introduction

Sympathetic activation is associated with increased risk of ventricular arrhythmias and sudden cardiac death (SCD).¹ We have shown that stellate ganglion nerve activity (SGNA) precedes spontaneous ventricular tachycardia (VT) and ventricular fibrillation (VF) in an ambulatory canine model of SCD.² Because sympathetic nerve activity is important in arrhythmia initiation, it is highly desirable to develop a reliable and less invasive method to measure sympathetic outflow for arrhythmia prediction and risk stratification. Heart rate variability and microneurography have been used to assess sympathetic tone in patients. However, due to technical problems, those methods are not widely used for arrhythmia prediction.

The skin is well innervated by sympathetic nerve fibers.^{3,4} Studies in dogs and rats demonstrated that the somata of the cutaneous sympathetic nerve fibers of the upper body are located in the middle cervical and stellate ganglia.^{5,6} Innervation of the pectoralis muscle, which is located underneath the hypodermis in the upper chest wall stems from the brachial plexus,⁷ which communicates with the stellate ganglion.⁵ In addition, light microscopy has revealed that large nerve trunks are present in the subcutaneous tissues.⁵ These observations suggest that recording skin and/or muscle directed postganglionic sympathetic nerve activity, which we will collectively term as “subcutaneous nerve activity” (SCNA), from the hypodermis of the upper trunk may be used as a less invasive surrogate for SGNA to measure sympathetic outflow. We have recently shown that SCNA correlates well with SGNA and heart rate in normal ambulatory dogs.⁸ It is not clear, however, if SCNA can be used to predict the onset of ventricular arrhythmias and SCD in diseased animals. The aim of the present study is to employ an established canine model of SCD and investigate the relationship between SCNA, SGNA, ventricular arrhythmias and SCD.

Methods

Surgical preparation

We re-analyzed data from seven ambulatory dogs with complete heart block, myocardial infarction, and nerve growth factor infusion to the left stellate ganglion from a previous study.² The protocol was approved by the Institutional Animal Care and Use Committee of Cedars-Sinai Medical Center, Los Angeles, California. One pair of electrodes was used to record nerve activity from the left stellate ganglion; another pair of bipolar electrodes was

implanted in the subcutaneous tissues of the right upper and left lower quadrant of the chest. In that study,² the subcutaneous electrodes were placed for the purposes of electrocardiogram (ECG) recording. In the current investigation, signals from those electrodes were high pass filtered and inspected for nerve signals. The recording was made in a bipolar mode, with two widely spaced bipoles. The electrodes were the stainless steel wires that came with the Data Sciences International (DSI, St Paul, MN) D70-EEE radiotransmitter. The terminal 5 mm of the wires were stripped of their insulation and used for electrical recording. Subcutaneous inter-electrode distance was not measured at the time of the study, but in similar size dogs it is estimated at 28 cm.

A detailed description of study methods is available in the Online Supplement.

Results

The simultaneous SGNA and SCNA recording lasted 43 ± 26 days.

Presence and characteristics of subcutaneous nerve discharges

Similar to a prior report in normal canines,⁸ all seven dogs demonstrated subcutaneous nerve discharges with similar morphology to the signals recorded from the left stellate ganglion. In addition, SCNA morphology was similar to filtered skin and muscle sympathetic nerve activity obtained in microneurography studies.^{9–11} We previously described two SGNA patterns in this canine model; low-amplitude burst discharge activity (LABDA) with amplitudes between 0.05–0.8 mV and high-amplitude spike discharge activity (HASDA) with amplitudes of 0.9 ± 0.16 mV.² Out of 366 randomly selected 15 s frames, 214 contained SGNA and 186 displayed SCNA. In 88% of frames, the presence or absence of SGNA correlated directly with the presence or absence of SCNA. There were frames containing subcutaneous, but not stellate ganglion discharges, suggesting that the origin of SCNA is not inadvertent recording of SGNA with the subcutaneous electrodes. All subcutaneous discharges demonstrated LABDA pattern with amplitude of 0.07 ± 0.08 mV. In all but one frame, which contained HASDA, SGNA also displayed LABDA pattern with amplitudes of 0.10 ± 0.11 mV. Unlike the SGNA channel, the SCNA channel was more prone to display incompletely filtered ECG signals and pacing artifacts. Figure 1 shows two VT episodes from two different animals. In the first animal (1A) the raw signal is high pass filtered to obtain non-contaminated SCNA. In the second animal (1B) the ECG signals from the raw signals could not be filtered well and contaminate the SCNA channel with ECG artifacts (downward arrows) despite high pass filtering. The onset of the SCNA discharge is still visible (upward arrow); however integration of SCNA (in mV-s) cannot be accurately accomplished due to unfiltered ECG signals.

Subcutaneous nerve activity and sudden cardiac death due to ventricular fibrillation

Two dogs died from SCD due to VF on postoperative days 3 and 52. Figures 2 and 3 show the recordings immediately before and after the onset of VF in these two dogs. In both dogs, VF was preceded by both SGNA and SCNA. The tracings of 2A and 2B as well as 3A and 3B are continuous with the 2A and 3A panels preceding the 3A and 3B panels by 40 s. LABDA discharges before VF are marked by downward arrows (Figure 2 and 3). Massive

SGNA and SCNA (asterisks) occurred after VF, likely responses to acute reduction of blood pressure. Similar to figure 1B, the dog portrayed in Figure 2 was not included in the quantitative nerve integration analyses because the SCNA channel was contaminated with ECG and pacing artifacts.

Subcutaneous nerve activity and ventricular tachycardia

Two episodes of VF and 59 episodes of VT from 6 dogs (7–12 per dog), occurring 23 ± 17 days after surgery with an average heart rate (HR) of 156 ± 44 beats per minute (bpm) and duration of 20 ± 89 s were analyzed. A total of 75% of VT/VF episodes were preceded by SGNA and 70% by SCNA within 15 s of initiation. Out of 61 15 s episodes of AIVR selected within 26 ± 26 minutes, 59% contained SGNA and 43% had SCNA. By using a generalized linear mixed-effects model, the odds ratio of observing SGNA and SCNA 15 s prior to VT/VF compared to observing discharges during episodes of AIVR for a specific dog was 2.32 (95% CI 1.01–5.31; $p=0.0466$) and 3.13 (95% CI 1.45–6.76; $p=0.004$), respectively. Figure 4A shows another representative episode of VT, along with a control AIVR episode in the same animal. The average latency of onset from the beginning of the stellate and the subcutaneous discharge to the onset of VT/VF was 17.1 ± 15.2 s and 17.8 ± 18.4 s, respectively. Because the average VT/VF occurrence was 2.1 ± 1.4 episodes per day, while intermittent LABDA from the subcutaneous tissues occurred throughout the day, <1% of SCNA LABDA episodes were followed by VT/VF. This is similar to data previously reported for SGNA discharges.² Integrated SGNA (iSGNA) and integrated SCNA (iSCNA) were calculated in three 20 s intervals for 60 s prior to 49 episodes of VT, 1 VF episode and 50 20 s frames of AIVR in 5 dogs. By using a linear mixed-effects model, progressive increase in iSGNA and iSCNA was observed before VT/VF (Figure 5).

Subcutaneous nerve activity and frequent bigeminy or couplets

Sixty-one episodes of FBG/C from 6 dogs (9–12 per dog), occurring 21 ± 15 days after surgery with an average HR of 77 ± 14 bpm were analyzed. Sixty-nine percent of FBG/C were associated with SGNA and 59% with SCNA within 15 s before initiation. In contrast, in 61 15 s frames of AIVR, occurring within 28 ± 26 minutes, SGNA was recorded 51% of the time and SCNA was present 41% of the time. By using a generalized linear mixed-effects model, the odds ratio of detecting nerve discharges 15 s prior to FBG/C compared to during the AIVR period for a specific dog was 2.44 (95% CI 1.08–5.51; $p=0.032$) for SGNA and 2.47 (95% CI 1.09–5.61; $p=0.031$) for SCNA. The latency of onset from the beginning of the stellate ganglion and the subcutaneous discharges to the beginning of the FBG/C episode was 16.8 ± 14.3 s and 17.8 ± 18.4 s, respectively. Figure 6A shows an example of FBG/C preceded by SCNA and SGNA.

Nerve activity was integrated 20 s before 50 episodes of FBG/C from 5 dogs and during 50 AIVR control episodes lasting 20 s. Using a linear mixed-effects model, both iSGNA (53.0 (95% CI 42.5–63.6) vs 35.9 (95% CI 29.1–42.7) mV-s; $p=0.0054$) and iSCNA (45.5 (95% CI 37.5–53.5) vs 34.8 (95% CI 28.1–41.4) mV-s; $p=0.0176$) were higher in the 20 s prior to FBG/C than during the 20 s frames of AIVR.

Subcutaneous nerve activity and premature ventricular contractions

Sixty-one PVCs from 6 dogs (9–12 per dog), occurring 22 ± 15 days after surgery were analyzed. Sixty-two percent of isolated PVCs were preceded by SGNA within 15 s and 61% by SCNA. Figure 6B shows an example of isolated PVC associated with SCNA and SGNA. In comparison, during 61 15 s frames of AIVR selected within 22 ± 26 minutes, SGNA was recorded 36% of the time and SCNA was noted 30% of the time. By using a generalized linear mixed-effects model, the odds ratio of having SGNA and SCNA discharges 15 s before isolated PVCs versus during the 15 s periods of AIVR for a specific dog was 4.06 ((95% CI 1.69–9.74); $p=0.0019$) for SGNA and 7.37 ((95% CI 2.75–19.76); $p=0.0001$) for SCNA.

Integrated SGNA and iSCNA 20 s prior to 50 isolated PVCs and during 50 20 s frames of AIVR were calculated in 5 dogs. Using a linear mixed-effects model, both iSGNA (50.5 (95% CI 39.7–61.3) vs 32.2 (95% CI 27.0–37.5) mV-s; $p=0.0019$) and iSCNA (45.2 (95% CI 36.0–54.3) vs 31.6 (95% CI 26.1–37.1) mV-s; $p=0.0058$) were higher in the 20 s prior to isolated PVCs compared to the 20 s control frames of AIVR.

Correlation between stellate ganglion nerve activity and subcutaneous nerve activity

To investigate the correlation between iSGNA and iSCNA, the 150 20 s segments of integrated nerve activity 60 s prior to VT/VF, the 50 20 s segments of integrated nerve activity prior to FBG/C and the 50 20 s segments of integrated nerve activity prior to isolated PVCs along with their respective 20 s AIVR control periods described in the quantitative analysis above were combined for each individual dog. As shown in Table 1, the average Pearson's correlation coefficient for all dogs was 0.65 ± 0.16 ($p<0.0001$ for each dog). When selecting the first 10 30 s frames during each hour of the day on the first available complete recording day from the same 5 dogs, a similar average Pearson's correlation coefficient of 0.73 ± 0.18 ($p<0.0001$ for all dogs) was obtained (Table 1).

Circadian variation in subcutaneous nerve activity

To determine if circadian variation is observed in iSGNA and iSCNA, the first 10 30 s frames during each hour of the day on the first available complete recording day were selected for analysis and combined for all 5 dogs. As demonstrated in Figure 7, both iSGNA and iSCNA exhibited circadian variation ($p<2 \times 10^{-16}$ for both through generalized additive mixed-effects models).

Discussion

The main finding of this study is that subcutaneous nerve discharges were recorded in all dogs studied and that similar to SGNA, SCNA preceded the onset of ventricular arrhythmias and SCD.

Presence of subcutaneous nerve discharges

We⁸ have recently reported that in ambulatory and anesthetized normal dogs, SCNA recorded by bipolar subcutaneous electrodes correlates with the SGNA and can be used to estimate the sympathetic tone. In the present study, the presence of SCNA was demonstrated

in all seven dogs using widely spaced bipolar electrodes placed in the upper trunk. Similar to that in normal dogs,⁸ SCNA in these diseased dogs had similar morphology, onset and duration compared to SGNA, but the recordings were sufficiently different to eliminate significant cross talk between the channels. In addition, SCNA signals were similar to filtered human microneurography signals obtained directly from a peripheral nerve.^{9–11} We therefore postulate that the origin of these discharges is mainly postganglionic sympathetic nerve activity directed to the skin and possibly the skeletal muscles below the hypodermis. The study by Robinson et al demonstrates that apamin injection in the stellate ganglion results in increased signal in the subcutaneous nerves.⁸ These findings suggest that SCNA represents peripheral output of increased sympathetic outflow. However, additional studies utilizing central pharmacological blockade with drugs such as clonidine or stellate ganglion ablation and measuring SGNA and SCNA should be undertaken to further elucidate the specific interplay between SCNA and SGNA.

Subcutaneous nerve activity, ventricular arrhythmias and sudden cardiac death

Similar to SGNA, SCNA preceded SCD and the majority of ventricular arrhythmias. The association between SGNA and ventricular arrhythmias demonstrated in this study is similar to reports published previously.^{2, 12} Both iSCNA and iSGNA showed progressive increase prior to VT/VF consistent with our prior reports.² Similarly, higher values for iSGNA and iSCNA were noted in the 20 s prior to PVCs and FBG/C compared to control periods of AIVR. These findings further demonstrate that similar to iSGNA, iSCNA is also increased prior to ventricular arrhythmias and SCD.

The latency from the onset of SGNA and SCNA to the development of VT and VF shown in the present study is about 17 s, slightly longer than the 10–15 s latency reported for atrial arrhythmias.¹³ However, the latency is dependent in part on the magnitude of nerve discharge. Large and synchronized sympathetic discharges from the stellate ganglion (the high-amplitude spike discharge activity) may induce VT with very short (less than a few s) latency.²

In our study, episodes of paced rhythm contained pacing artifact contamination in the SCNA channel and episodes of AIVR were used as control periods. Some reports demonstrate that AIVR is associated with increased sympathetic tone;^{14, 15} while others propose that it is associated with increased vagal tone.¹⁶ Our study suggests that at anywhere from 36–59% of AIVR episodes contained SGNA; however we did not measure vagal nerve activity and lack recordings with periods of normal sinus rhythm to compare these results to. As such, conclusions about the autonomic nervous system influences on AIVR cannot be made based on our study.

Relationship between integrated SGNA and integrated SCNA

A good correlation was found between iSGNA and iSCNA with an average Pearson's correlation of 0.73 (range 0.50–0.92). This is similar to values obtained in normal dogs.⁸ It is unclear why some dogs displayed better correlation than others. Because this is a retrospective analysis of a study, which used the subcutaneous electrodes for recording of cardiac rhythm, it is possible that some of the subcutaneous electrodes were located more

caudally within the boundaries of the left lower thorax than others. As the density of sympathetic fibers originating from the stellate ganglion in canines decreases in a cranial to caudal direction with maximum concentration in the upper extremities,⁵ more caudal placement of the left thorax electrode could have resulted in worse correlation between iSGNA and iSCNA. Another possibility is that some dogs had larger percentage of ECG contamination in the subcutaneous channel than others. Alternatively, there may be some variation in the density of synapses between the stellate ganglion and the subcutaneous tissues between animals and for any specific point in the lower thorax, a larger percentage of sympathetic fibers could derive from the thoracic sympathetic chain rather than the stellate ganglion in some canines compared to others. Further studies are needed to determine the optimal location and distance between the electrodes for subcutaneous nerve recordings.

Sources of signals in the subcutaneous space

Sources of recordings from the subcutaneous space could include signals from autonomic nerves, motor and sensory nerves, unfiltered ECG or pacing artifacts and movement artifacts. High pass (150 Hz) filtering eliminates a significant amount of muscle contractions,¹⁷ motion noise¹⁸ and residual ECG signals. When excluding periods during which signal contamination with ECG and/or pacing artifact is pronounced, there is a significant correlation between SCNA, ventricular arrhythmias and SCD and between SCNA and SGNA. In addition, consistent with prior reports observing circadian variation of sympathetic nerve activity in dogs,^{8, 12, 19} both iSGNA and iSCNA showed circadian variation. These collective findings suggest that in the absence of significant ECG and/or pacing contamination, the majority of signals recorded from the subcutaneous space after high pass signal filtering are sympathetic in origin.

Study Limitations

The most significant limitation of this study is that incomplete filtering of ECG and pacing artifact was observed in the subcutaneous channel. The degree of signal contamination appeared to vary between dogs, possibly due to variation in the location of the recording device and its proximity to the recording electrodes. Another limitation is that we only recorded signals from the left stellate ganglion in this study, and that the DSI D70-EEE radio transmitter is designed to record low frequency signals; such as an ECG and signals with frequencies higher than 250 Hz are not detected. However, our recent study has recorded simultaneously from the right stellate ganglion and right thoracic subcutaneous tissues using equipment with wide bandwidth and high sampling rate. The results show that both right SGNA and right SCNA correlated well with the sinus rate.⁸ It is possible that future improvement of recording equipment of the implanted devices will allow more effective use of SCNA for ventricular arrhythmia detection and risk stratification. While high pass filtering at 150 Hz eliminates the majority of muscle contractions and motion artifacts, it also eliminates nerve signals below 150 Hz.²⁰ In addition, we do not have vagal nerve activity recording. Further studies should include vagal nerve measurements and attempt to determine the relationship between vagal nerve activity, SCNA and ventricular arrhythmias.

Conclusions

In the absence of significant ECG/pacing artifact contamination of the nerve recording channels, SCNA measured from the thorax could be used to estimate the left SGNA and predict the susceptibility to ventricular arrhythmias in a canine model of SCD.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Abbreviations

AIVR	accelerated idioventricular rhythm
ECG	electrocardiogram
FBG/C	frequent bigeminy or couplets
HASDA	high-amplitude spike discharge activity
LABDA	low-amplitude burst discharge activity
PVC	premature ventricular contraction
SCNA	subcutaneous nerve activity
SGNA	stellate ganglion nerve activity
VF	ventricular fibrillation
VT	ventricular tachycardia

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Clinical Perspectives

Sympathetic tone is important in cardiac arrhythmogenesis. Because the sympathetic nerve structures, such as stellate (cervicothoracic) ganglion are located deep in the thoracic cavity, it is difficult to directly record from those structures to determine the sympathetic tone. Heart rate variability and microneurography techniques have been used to assess sympathetic tone in patients. However, technical difficulties have prevented routine use of those methods for arrhythmia prediction and risk stratification.

Subcutaneous tissues contain sympathetic nerve fibers that originate from the stellate ganglion. We found that it is possible to record the subcutaneous nerve activities with implanted electrodes and radiotransmitters in ambulatory dogs. In addition, we found that the subcutaneous nerve activities correlate well with the stellate ganglion nerve activity, and that the subcutaneous nerve activities precede the onset of ventricular tachycardia and fibrillation. Because subcutaneous tissues are much more accessible than the thoracic cavity, the ability to directly record sympathetic nerve activities from the subcutaneous tissues may provide a new approach to estimate the sympathetic tone. These methods can be translated into clinical practice by incorporating them in the implantable loop monitors, pacemakers or implantable cardioverter-defibrillators with high sampling rate and wide bandwidth. The nerve signals and the electrocardiographic signals can be separated by different filtering settings. The data can be analyzed to determine whether or not subcutaneous nerve activities precede the onset of spontaneous ventricular arrhythmias and sudden death. These clinical studies may lead to improved arrhythmia prediction in patients with implanted devices.

Figure 1. Stellate ganglion nerve activity (SGNA) and subcutaneous nerve activity (SCNA) in association with ventricular tachycardia (VT) from two different dogs

In the first dog (Panel A), unfiltered (raw) signal (bottom panel) shows both the ECG and the high frequency SCNA. The ECG was nearly completely removed by high pass filtering as shown in the SCNA channel. In a second dog (Panel B), unfiltered (raw) signal (bottom panel) has large contamination by the ECG. High pass filtering did not completely remove the ECG (downward arrows). Increase in nerve activity over the baseline however is still evident (upward arrow points at onset of SCNA) preceding VT. The SGNA and SCNA channels are filtered at 150 Hz high pass; the electrocardiogram (ECG) is filtered at 30 Hz low pass. The raw signal is unfiltered. Units for SGNA, SCNA and ECG are displayed in mV.

Figure 2. Subcutaneous nerve activity (SCNA) and stellate ganglion nerve activity (SGNA) precede ventricular fibrillation (VF) and sudden cardiac death

A: Simultaneous firing of SGNA and SCNA is observed 40 s prior to VF. Downward arrows point to low-amplitude burst discharge activity (LABDA). Similar to Figure 1B, the subcutaneous channel in this animal is heavily contaminated by ECG artifacts; however, presence of nerve discharges is still apparent. **B:** Continuous tracing from panel A 40 seconds later shows initiation of VF (upward arrow) and large discharges in the stellate and subcutaneous channels (indicated by the asterisks) following VF. **C:** 10 s tracing from the boxed panel in B demonstrating the onset of VF in greater detail. Units for SGNA, SCNA and ECG are displayed in mV. p=p waves.

Figure 3. Increased stellate ganglion nerve activity (SGNA) and subcutaneous nerve activity (SCNA) precede ventricular fibrillation (VF) and sudden cardiac death in another dog
A: Low amplitude burst discharge activity in the stellate ganglion is evident 40 s prior to VF. At the same time, SCNA is intermittently quiescent and discharges resume 30 s before VF (downward arrows). **B** is continuous 40s after panel A, showing initiation of VF and massive stellate and subcutaneous discharges after VF onset (asterisks). **C:** Magnification of the boxed area of panel B showing the beginning of VF (upward arrow). The SGNA and SCNA channels are filtered at 150 Hz high pass; the ECG is filtered at 30 Hz low pass. Units for SGNA, SCNA and ECG are displayed in mV. ECG=electrocardiogram. p=p waves.

Figure 4. Examples of stellate ganglion nerve activity (SGNA) and subcutaneous nerve activity (SCNA) prior to the onset of ventricular tachycardia (VT) and in association with accelerated idioventricular rhythm (AIVR)

A: Prolonged low amplitude burst discharge activity (LABDA) recorded from the stellate ganglion and the subcutaneous tissues are present prior to the onset of several short runs of VT. **B:** LABDA associated with episodes of AIVR not preceding VT in the same canine. The maximum amplitudes of SGNA and SCNA are lower in this frame compared to **A**. Units for SGNA, SCNA and ECG are displayed in mV. ECG=electrocardiogram. The SGNA and SCNA channels are filtered at 150 Hz high pass; the electrocardiogram (ECG) is filtered at 30 Hz low pass. The raw signal is unfiltered.

Figure 5. Integrated stellate ganglion nerve activity (SGNA) and integrated subcutaneous nerve activity (SCNA) in mV-s prior to the onset of ventricular tachycardia (VT), ventricular fibrillation (VF) and accelerated idioventricular rhythm (AIVR)

Progressive increase in both integrated SGNA and integrated SCNA is noted from 20 s periods of AIVR (n=50) to 20 s intervals measured 60 s, 40 s and 20 s before initiation of VT (n=49) and VF (n=1) in 5 dogs. *P<0.05 vs AIVR; †P<0.05 vs - 60 s; ‡P<0.05 vs AIVR. Red bars signify the mean values and the upper and lower 95% confidence intervals. Numerical values underneath the x-axis represent the mean and upper and lower 95% confidence intervals of integrated SGNA and SCNA in mV-s. INA=integrated nerve activity.

Figure 6. Examples of stellate ganglion nerve activity (SGNA) and subcutaneous nerve activity (SCNA) prior to the onset of frequent couplets and a premature ventricular contraction in two different canines

A: Low-amplitude burst discharge activity (LABDA) is noted in the stellate and the subcutaneous channel in association with frequent couplets. **B:** Two LABDA episodes and one episode of high-amplitude spike discharge activity (HASDA, arrow) with amplitude of 1.1 mV recorded from the stellate ganglion and two episodes of LABDA from the subcutaneous tissues with lower amplitude are associated with the occurrence of a premature ventricular contraction in another canine. The SGNA and SCNA channels are filtered at 150 Hz high pass; the ECG is filtered at 30 Hz low pass. Units for SGNA, SCNA and ECG are displayed in mV. ECG=electrocardiogram.

Figure 7. Circadian variation in integrated stellate ganglion nerve activity (SGNA) and integrated subcutaneous nerve activity (SCNA) in mV-s

Both SGNA (A) and SCNA (B) exhibit circadian variation ($p < 2e-16$ for both). Solid lines represent mean integrated SGNA (A) and SCNA (B) averaged for the first ten 30 s segments of each hour over 24 hours and combined in 5 dogs. Dotted lines represent 95% upper and lower confidence intervals.

Table 1

Pearson's correlation coefficients between integrated stellate ganglion nerve activity (SGNA) and integrated subcutaneous nerve activity (SCNA).

Dog number	Pearson's Correlation *	P value	Pearson's Correlation †	P value
1	0.48	<.0001	0.67	<.0001
2	0.64	<.0001	0.50	<.0001
3	0.80	<.0001	0.91	<.0001
4	0.83	<.0001	0.92	<.0001
5	0.52	<.0001	0.66	<.0001
Average	0.65		0.73	

* Values for 20 s intervals of integrated SGNA and SCNA 60 s prior to ventricular tachycardia (n=49; 7–12 per dog) and ventricular fibrillation (n=1), 20 s prior to frequent bigeminy or couplets lasting for 10 or more consecutive beats (n=50; 9–12 per dog), 20 s prior to premature ventricular contractions (n=50; 8–12 per dog) and during their respective 20 s frames of accelerated idioventricular rhythm used as control (n=150) combined for each individual dog in five dogs. Total number of integrated nerve activity frames=400. Values for dog #1 were adjusted in 4 episodes of VT for the presence of unfiltered ECG artifacts.

† Values for 30 s intervals of integrated SGNA and SCNA during the first 10 frames of each hour over a 24-hour period combined for each dog in the same five dogs (total number of integrated nerve activity frames=1200; 240 per dog).