Intrinsic Cardiac Autonomic Nervous System: what do clinical electrophysiologists need to know about the 'heart brain'?

Tolga Aksu[1], Rakesh Gopinathannair[2], Dhiraj Gupta[3], and Dainius Pauza[4]

[1]Kocaeli Derince Egitim ve Arastirma Hastanesi
[2]Kansas City Heart Rhythm Institute
[3]Liverpool Heart and Chest Hospital
[4]Lithuanian University of Health Sciences

February 14, 2021

Abstract

It is increasingly recognized that the autonomic nervous system (ANS) is a major contributor in many cardiac arrhythmias. Cardiac ANS can be divided into extrinsic and intrinsic parts according to the course of nerve fibers and localization of ganglia and neuron bodies. Although the role of extrinsic part has historically gained more attention, the intrinsic cardiac ANS may affect cardiac function independently as well as influence the effects of the extrinsic nerves. Catheter based modulation of the intrinsic cardiac ANS is emerging as a novel therapy for management of patients with brady and tachy arrhythmias resulting from hyperactive vagal activation. However, distribution of intrinsic cardiac nerve plexus in the human heart and the functional properties of intrinsic cardiac neural elements remain insufficiently understood. The present review aims to bring the clinical and anatomical elements of the ICANS together, by reviewing neuroanatomical terminologies and physiological functions, in order to guide the clinical electrophysiologist in the catheter lab, and to serve as a reference for further research.

Intrinsic Cardiac Autonomic Nervous System: what do clinical electrophysiologists need to know about the 'heart brain'?

Authors

Tolga Aksu, MD [1]

Rakesh Gopinathannair, MD, MA [2]

Dhiraj Gupta, MD 4

Dainius H. Pauza, PhD [3]

Affiliations

1 Kocaeli Derince Training and Research Hospital, Kocaeli, Turkey

2 Kansas City Heart Rhythm Institute and Research Foundation, Kansas City, United States

3 Lithuanian University of Health Sciences, Kaunas, Lithuania

4 Liverpool Heart and Chest Hospital, Liverpool, United Kingdom

Correspondence

Assoc. Prof. Dr. Tolga Aksu

Mailing address: Kocaeli Derince Education and Research Hospital, Department of Cardiology, Kocaeli, Turkey

Zip code: 41500

Email: aksutolga@gmail.com

Abstract

It is increasingly recognized that the autonomic nervous system (ANS) is a major contributor in many cardiac arrhythmias. Cardiac ANS can be divided into extrinsic and intrinsic parts according to the course of nerve fibers and localization of ganglia and neuron bodies. Although the role of extrinsic part has historically gained more attention, the intrinsic cardiac ANS may affect cardiac function independently as well as influence the effects of the extrinsic nerves. Catheter based modulation of the intrinsic cardiac ANS is emerging as a novel therapy for management of patients with brady and tachy arrhythmias resulting from hyperactive vagal activation. However, distribution of intrinsic cardiac nerve plexus in the human heart and the functional properties of intrinsic cardiac neural elements remain insufficiently understood. The present review aims to bring the clinical and anatomical elements of the ICANS together, by reviewing neuroanatomical terminologies and physiological functions, in order to guide the clinical electrophysiologist in the catheter lab, and to serve as a reference for further research.

Keywords

Ablation, intrinsic cardiac nerves, ganglionated plexus, left atrium, parasympathetic

Introduction

Cardiac autonomic nervous system (ANS) consists of components from parasympathetic and sympathetic systems (1). As first glance, while parasympathetic activity causes negative chronotropic and dromotropic effects, the sympathetic system primarily affects cardiac contractility and regulates peripheral vasoconstriction (1). Although the two components are usually considered to be antagonistic, autonomic control of the heart is regulated via several levels of feedback loops with a fine balance of sympathetic and parasympathetic signals between the heart and the peripheral and central nervous systems (2).

Structurally, the ANS of any visceral organ is represented by a complex neural plexus formed by extrinsic and intrinsic parts according to localization of postganglionic neurons that provide fibers from the ganglion to the effector organ (3, 4). For the heart, large numbers of neurons are associated with ganglia and their interconnecting nerves on atria and ventricles, and this intrinsic cardiac ANS has been collectively referred to as the heart's "*little brain*"(4-8). Extrinsic and intrinsic cardiac structures in animal models and humans are species-dependent and translating experimental data obtained from animals to humans can be challenging.

Excessive parasympathetic tone may be an important cause of several clinical bradyarrhythmias such as functional atrioventricular block, some forms of sinus node dysfunction, and vasovagal syncope (VVS) (9). Similarly, a causal relationship between intrinsic cardiac ANS and atrial tachyarrhythmias has been demonstrated both in animals and in humans (10-16). Furthermore, electrophysiological effects of ANS may be exacerbated in diseased hearts (11, 12). In recent years, catheter based autonomic neuromodulation has emerged as an important novel therapy for VVS and for atrial fibrillation (AF), and has been shown to have better efficacy when compared with pharmacological therapies (9, 10). However, the full extent of the distribution of intrinsic cardiac nerve plexus on the human heart and the functional properties of human intrinsic cardiac neural elements remain insufficiently understood by many clinical electrophysiologists. In this review, we focus on anatomical and functional characteristics of the intrinsic cardiac ANS in non-human mammals and in humans, and attempt to present neuroanatomical terminologies in an attempt to fill this knowledge gap.

The extrinsic cardiac nervous system

The extrinsic part of cardiac nervous system includes the ganglia and nerves en route to the heart. A

ganglion is a cluster of neuronal cell bodies outside the brain (1). In the ANS, while efferent axons from the central nervous system to the ganglion are known as preganglionic nerve fibers, efferent axons from the ganglion to the effector organ are called postganglionic nerve fibers (1). Although ganglia of the sympathetic division are located within the sympathetic chain or paravertebral ganglia, ganglia of the parasympathetic division which are called as either the intramural or the intrinsic ones are distributed mainly within the epicardial area (3, 17, 18) (Figure 1). Thus, parasympathetic postganglionic fibers tend to be shorter than sympathetic ones. All extracardiac nerves access the epicardial neural plexus as extensions of the extrinsic cardiac plexuses.

The intrinsic cardiac nervous system

The intrinsic cardiac nervous system comprises efferent parasympathetic and sympathetic neurons, afferent (sensory) neurons, and local circuit neurons/interneurons acting via diverse neurotransmitters (Figure 1) (19-24). For many years, studies on the mammalian heart mostly focused on determining the location of epicardial ganglia using histologic examination of heart sections, resulting in the concept of ganglionated plexuses (GPs) consisting of grouping ganglia in different sites (3, 4, 25). However, this somewhat simplistic notion has been challenged by recent observations that epicardial ganglia may be very widely distributed, and their sizes may be extremely variable ranging from those that are only observable with a microscope to those that are easily discernible with the naked eye (26). Furthermore, the hearts of humans and some mammalian species contain more than one and a half thousand ganglia (4, 26, 27). Thus, the actual number and distribution of these ganglia might only be identified on the non-sectioned heart preparations (26). By staining of intrinsic cardiac neural plexus on the whole heart, it has been disclosed that the heart is under neuronal control through one intrinsic epicardiac neural plexus, nerves of which extend to distinct cardiac regions by seven neural pathways or routes, called as epicardiac neural ganglionated subplexi (sGPs) (26-29) (Figure 2). Anatomically, nerves accessing the heart penetrate through the heart hilum at the base of the heart into the epicardium and to the intrinsic ganglia distributed mostly within the epicardium. Then, the postganglionated intrinsic nerves extend towards the specific atrial or ventricular regions - around the sinoatrial node, the roots of caval and pulmonary veins (PVs), and near the atrioventricular node (Figures 1 and 2). The main difference of GP nomenclature from that of sGP terminology is that specific GPs are parts of the larger ganglionated fields of specific sGPs. Since epicardial ganglia are persistently distributed along those sGP nerves, the use of the inclusive term 'ganglia' instead of GPs and sGPs might be more reasonable to define those areas anatomically. Because electrophysiological detection and ablation of*ventricular* sGPs has been practically unexamined so far, the following part of the review will discuss the intrinsic ganglia distributed in the atrial epicardium.

The organization of the intrinsic cardiac autonomic nervous system in non-human mammals

A significantly lower number of nerve cell bodies is evident in structure of sGPs of smaller mammals as compared to larger ones (24-34). Also, while in small mammals, ganglionic cells are usually clustered across the hilum, they are distributed more widely across both atria in larger mammals. Essentially, five atrial sGPs have been defined in non-human mammals (Figure 2): 1) the left dorsal sGP (LDsGP); 2) the middle dorsal sGP (MDsGP); 3) the ventral left atrial sGP (VLAsGP); 4) the ventral right atrial sGP (VRAsGP); and 5) the dorsal right atrial sGP (DRAsGP) (28-31).

The organization of the intrinsic cardiac autonomic nervous system in humans

Although, in general, the morphology of the human sGPs corresponds with that in larger mammals, particularly, with the canines, topographical and quantitative inter-species differences are evident (26, 28, 30, 33, 34). According to GP nomenclature by Armour et al (4) that is commonly used to define the distribution of intrinsic cardiac ganglia in experimental and clinical studies, the following five major and one minor atrial locations are consistently identified in humans: 1)*the superior right atrial GP* located on the postero-superior surface of the right atrium (RA) adjacent to the junction of the SVC and the RA; 2) *the superior left atrial GP* on the postero-superior surface of the left atrium (LA) between the PVs; 3) *the posterior right atrial GP* located adjacent to the interatrial groove; 4)*the posteromedial left atrial GP* on the postero-medial surface of

the LA; 5) *the interatrial septal GP* consisting of fusion and extensions of *the posterior right atrial GP* and *the posteromedial left atrial GP* ; and 6) *the posterolateral left atrial GP* is identified on the postero-lateral surface of the LA. According to Pauza et al (28, 35, 36, 68) who studied the intrinsic nerves and ganglia on the human whole-mount heart preparations, these 5 GPs are densely interconnected by thinner nerves and, therefore should be considered as just distinctive, ganglionated areas of the continuous cardiac ganglionated nerve plexus (Figure 2, Table 1). Since these ganglionated areas have their specific neural fibers accessing through cardiac hilum nerves as well as the nerves that extend from these ganglia towards the specific atrial and ventricular regions, they were referred to by Pauza et al (28) as sGPs. On the human atria, there are discerned (1) *the VRA (anterior right) sGP* which occupies ventral superior right atrial region, ventral side of the root of the SVC, and ventral inferior right atrial region; 2) *the VLA (anterior left) sGP* covers the rather narrow ventral superior left atrial region; 3)*the LD (posterior left) sGP* distributes across the left coronary sulcus, region of dorsal left coronary sulcus, and middle left atrial region regions and contains abundant ganglia; 4*) the MD (posterior middle) sGP* extends on the dorsal superior left atrial region and around the crux cordis; and 5) *the DRA (posterior right) sGP*which resides in the dorsal superior right atrial region, dorsal side of the root of the SVC, and region over the interatrial septum. Innervation routes of these atrial sGPs are provided in Online Supplemental File.

The ligament of Marshall (LOM) or neural fold of the LA is considered part of the intrinsic cardiac ANS. Immunohistochemistry confirmed that it involves a number of branches of the left vagus, specifically *the LDsGP* , and sympathetic nerve fibers (37). Cholinergic nerve fibers originating in the LOM were found to innervate surrounding left atrial structures, including the PVs, left atrial auricle or appendage, and coronary sinus (Figure 3). It becomes the vein of Marshall caudally as it connects with the coronary sinus.

Innervation principles of the sinoatrial and the atrioventricular nodes

In physiological experiments in canine and primates, removal of the epicardial ganglia located at the junction of the inferior vena cava and inferior wall of the LA eliminated the negative dromotropic effects of vagal nerve stimulation without suppressing vagally mediated reductions in heart rate (6, 7, 38, 39). On the contrary, surgical removal of the epicardial ganglia and nerves located around the right PVs attenuated the negative chronotropic response to vagal nerve stimulation without adversely affecting vagal inhibition of atrioventricular conduction (6, 7). In the human heart as well as in the hearts of some other mammalian species, both *the VRAsGP* and *the DRAsGP* supply epicardial nerves to the sinoatrial nodal neural network (Figure 2 and Online Supplemental Figure 1) (40, 41). Although the size and number of ganglia inside these sGPs vary from one heart to another, the largest number and density of epicardial ganglia are usually located at the junction of the SVC with the RA. It is noteworthy that some part of the postganglionated nerves of *the VRAsGP* passing onto the LA invariably extended via a remarkable crest of the ventral surface of the LA. This anatomical distribution should be kept in mind to understand why bi-atrial ablation might be needed for complete denervation of the sinoatrial node in some cases (Figure 4). Despite results of physiological experiments, successive anatomical attempts to determine nerves that coursed towards the canine or ovine atrioventricular node failed as those endocardial nerves are especially tiny (33-35). However, postganglionated nerves from *the LDsGP* , and *the MDsGP* extend towards the interatrial septum and presumably supply the atrioventricular nodal region (Figure 4) (33-35).

Theoretical background of intrinsic cardiac autonomic nervous system modulation for vasovagal syncope and atrial fibrillation

As the most common type of syncope, VVS is characterized by an abrupt dysregulation of the ANS to maintain adequate blood pressure and/or heart rate for cerebral perfusion (42). Central or peripheral triggers initiate three well-defined efferent responses: a cardioinhibitory response due to parasympathetic overactivity manifested by persistent bradycardia or prolonged pauses; a vasodepressor response due to sympathetic withdrawal manifested by significant hypotension; and a mixed response manifested by co-existing bradycardia and hypotension (Online Supplemental Figure 2) (42). Theoretically, neuromodulation of the intrinsic cardiac ANS works by preventing the parasympathetic efferent arm of the reflex arc in cardioinhibitory VVS and in mixed VVS types with a predominant cardioinhibitory response. The clinical

efficiency of catheter based neuromodulation in patients with VVS has been reported by several groups (43-49), with freedom from syncope of between 80% and 100%.

After demonstration of the PV myocardial sleeves as the main foci for initiation of paroxysmal AF, studies have focused on the difference in electrophysiological properties of PVs and the adjacent LA myocardium. Experimental studies have shown that stimulation of epicardial ganglia causes both sympathetic and parasympathetic activation and PV myocytes are more prone to effects of local autonomic nerve stimulation compared to LA myocytes (13-16, 50-54). While parasympathetic effect shortens the action potential duration, norepinephrine enhances the calcium transient in the PVs. The disparity between the short action potential duration and the enhanced calcium transient results in early afterdepolarization and triggered firing in the PVs (Online Supplemental Figure 2). Scherlag et al (50) showed that stimuli applied to PVs could not induce AF unless there was also simultaneous stimulation of the adjacent epicardiac ganglia. Furthermore, blockade of muscarinic cholinergic receptors by atropine suppressed triggered firing from the PVs. Although experimental data have shown potential importance of intrinsic cardiac ANS, clinical studies investigating the role of adding ANS modulation to PV isolation in patients with AF have shown conflicting results (52-54). After demonstration of that high-frequency stimulation (HFS) at the LOM may cause parasympathetic responses characterized by significant slowing of atrioventricular nodal conduction and AF induction in animal and human experiments, the role of the vein of Marshall ethanol infusion has been investigated in patients with AF (55, 56). In the VENUS Randomized Clinical Trial, addition of vein of Marshall ethanol infusion to catheter ablation, compared with catheter ablation alone, increased the likelihood of remaining free of AF or atrial tachycardia at 6 and 12 months. (57). A similar experience was confirmed in recently published Marshall-Plan study (58).

Detection ofintrinsic cardiac autonomic nervous system during electrophysiological study

Three different approaches have been used for clinical identification of intrinsic cardiac ANS in atria in the catheter laboratory: 1) HFS; 2) electrogram analysis; and 3) empirical anatomic ablation (9, 10).

High-frequency stimulation

Animal experiments demonstrated that electrical stimulation of left atrial sites areas caused 2 types of response in different parts of the LA: a vagal response (VR), defined as a significant prolongation of the PR and/or RR intervals, and a normal response characterized by the absence of any effect or nonsignificant changes on the PR or RR intervals (55). Theoretically, demonstration of a positive VR differentiates autonomic innervation sites than normal atrial myocardium. Because each epicardiac ganglia contains both parasympathetic and sympathetic neural elements, response to HFS may change according to duration of application. While shorter applications stimulate parasympathetic fibers, if HFS is delivered more than 2–5 seconds, sympathetic fibers may be stimulated, which may subsequently mitigate the parasympathetic response (55, 59).

According to the sites exhibiting a VR to HFS and fractionated electrogram characteristics during AF, 5 major epicardiac GPs were identified by Nakagawa et al (55): 1) *the right anterior GP* ; 2)*the right inferior GP* ; 3) *the left superior GP* ; 4)*the left inferior GP* ; and 5) *the Marshall ligament GP* . Figure 5 demonstrates the anatomical distribution of these 5 GPs (55). Interestingly, in spite of using a quite similar HFS protocol, Kim et al (60) found a more scattered distribution although the VR sites were located mainly on the posterior wall of the LA. We believe that one reason for this discrepancy may be that HFS may stimulate not only epicardial ganglia but also nerves that extend from epicardial ganglia towards the atrial regions. Indeed, despite its strong theoretical background, a HFS-based strategy has not demonstrated an advantage over empirical anatomic ablation in patients with AF as well as VVS (61, 62).

Electrogram analysis

By using fast Fourier transform analysis, Pachon et al (63) defined*compact* and*fibrillar atrial potentials* during sinus rhythm. Because fibrillar potentials demonstrated fragmented and heterogeneous conduction properties, authors speculated that fibrillar potentials may be originated from incursions of neural and vas-

cular structures and be used to detect autonomic innervation sites. Lellouche et al (64) analyzed electrogram characteristics based on VRs during radiofrequency application and demonstrated that the best single predictor of VR during radiofrequency application was the presence of at least 4 electrogram deflections at the ablation site. In our initial work, epicardiac ganglia sites were detected through a combination of fast Fourier transform analysis of electrograms, and HFS (65). In accordance with Lellouche's observations, all the electrograms on the radiofrequency ablation sites demonstrated a fragmented pattern. Because the better resolution afforded by usage of higher high-pass filters allowed better appreciation of electrogram fragmentation (63), in our subsequent work, we used band-pass filter settings of 200-500Hz instead of conventional band-pass filter settings of 30-500Hz during sinus rhythm and targeted all fragmented electrograms in regions which are anatomically consistent with autonomic innervation sites (46) (Table 1). Indeed, this simpler electroanatomical mapping-guided strategy demonstrated an identical success rate in preventing prodromal symptoms and syncope recurrence as compared to the previous combined approach. Figure 5 demonstratesthe anatomical distribution of GPs according to our definition method (49).

Empirical anatomic ablation

This strategy can be used in 2 different ways: adjunctive to electrogram analysis or HFS (44) or both; or as a true stand-alone strategy, via isolated right atrial or left atrial approaches (48, 62). Sun et al (48) compared HFS-guided and empirically defined ablation approaches in 57 VVS cases. No statistical differences were found between HFS-guided and anatomical-guided ablation groups in either freedom from syncope or recurrent prodromes.

Future implications

The optimal approach to modulate the intrinsic cardiac ANS, the number of sGPs to be targeted and the long-term consequences of these therapies remain unclear. It is important to note that intrinsic cardiac ANS modulation can be associated with procedural risks, either in terms of procedural complications or the potential for off target effects like enhanced susceptibility to ventricular arrhythmias (66). Given the very complicated and variable distribution of epicardial ganglia and sGPs in humans, further characterization of the hierarchy of sGPs and specific atrial sGPs that ought to be targeted for arrhythmia therapy will be crucial. Until we get more data from prospective randomized studies, it is reasonable to proceed with caution in ablating these structures.

Conclusions

From an electrophysiologic perspective, although differences in nomenclature exist between cardiac neuroanatomists and cardiac physiologists, the route of innervation to the heart tends to be fairly consistent and should be defined by the cardiac plexus, the major ganglionated part of which is found in the atrial epicardium. Understanding of these specific anatomical principles of the innervation of the heart may facilitate to create a framework for modern therapies to directly target autonomic function.

Funding: There is no finding

Disclosures: All authors declare that there is no financial relationship.

References

1. Hall JE. Guyton and Hall Textbook of Medical Physiology (12th ed.). Philadelphia, PA: Elsevier Saunders, 2011.

2. Shivkumar K, Ajijola OA, Anand I, et al. Clinical neurocardiology defining the value of neuroscience-based cardiovascular therapeutics. J Physiol 2016;594:3911-3954.

3. Janes RD, Brandys JC, Hopkins DA, Johnstone DE, Murphy DA, Armour JA. Anatomy of human extrinsic cardiac nerves and ganglia. Am J Cardiol 1986;57:299–309.

4. Armour JA, Murphy DA, Yuan BX, Macdonald S, Hopkins DA. Gross and microscopic anatomy of the human intrinsic cardiac nervous system. Anat Rec 1997;*247* :289–298.

5. Ellison JP, Hibbs RG. An ultrastructural study of mammalian cardiac ganglia. J Mol Cell Cardiol 1976;*8:* 89–101.

6. Ardell JL. Anatomy and function of mammalian intrinsic cardiac neurons. In: Armour JA, Ardell JL, eds. Neurocardiology. New York, NY: Oxford University Press; 1994:95–114.

7. Randall WC, Ardell JL, O'Toole MF, Wurster RD. Differential autonomic control of SAN and AVN regions of the canine heart: structure and function. Prog Clin Biol Res 1988;275:15-31.

8. Billman GE, Hoskins RS, Randall DC, Randall WC, Hamlin RL, Lin YC. Selective vagal postganglionic innervation of the sinoatrial and atrioventricular nodes in the non-human primate. J Auton Nerv Syst 1989;26:27-36.

9. Aksu T, Guler TE, Yalin K, Mutluer FO, Ozcan KS, Calò L. Catheter Ablation of Bradyarrhythmia: From the Beginning to the Future. Am J Med Sci 2018;355:252-265.

10. Aksu T, Güler TE, Mutluer FO, Oto MA. Vagal denervation in atrial fibrillation ablation: A comprehensive review. Anatol J Cardiol 2017;18:142-148.

11. Opthof T, Coronel R, Vermeulen JT, Verberne HJ, van Capelle FJ, Janse MJ. Dispersion of refractoriness in normal and ischaemic canine ventricle: effects of sympathetic stimulation. Cardiovasc Res 1993;27:1954-1960.

12. Opthof T, Dekker LR, Coronel R, Vermeulen JT, van Capelle FJ, Janse MJ. Interaction of sympathetic and parasympathetic nervous system on ventricular refractoriness assessed by local fibrillation intervals in the canine heart. Cardiovasc Res 1993;27:753-759.

13. Choi E-K, Shen MJ, Han S, et al. Intrinsic cardiac nerve activity and paroxysmal atrial tachyarrhythmia in ambulatory dogs. Circulation 2010;121:2615–2623.

14. Shen MJ, Choi EK, Tan AY, et al. Patterns of baseline autonomic nerve activity and the development of pacing-induced sustained atrial fibrillation. Heart Rhythm 2011;8:583–589.

15. Uradu A, Wan J, Doytchinova A, et al. Skin Sympathetic Nerve Activity Precedes the Onset and Termination of Paroxysmal Atrial Tachycardia and Fibrillation. Heart Rhythm 2017;14:964–971.

16. Patterson E, Po SS, Scherlag BJ, Lazzara R. Triggered firing in pulmonary veins initiated by in vitro autonomic nerve stimulation. Heart Rhythm 2005;2:624-631.

17. Hopkins DA, Bieger D, de Vente J, Steinbusch WM. Vagal efferent projections: viscerotopy, neurochemistry and effects of vagotomy. Prog Brain Res 1996;107:79–96.

18. Hanna P, Rajendran PS, Ajijola OA, et al. Cardiac neuroanatomy-Imaging nerves to define functional control. Auton Neurosci 2017;207:48-58.

19. Baluk P, Gabella G. Some parasympathetic neurons in the Guinea-pig heart express

aspects of the catecholaminergic phenotype in vivo. Cell Tissue Res 1990;261;275–285.

20. Arora RC, Waldmann M, Hopkins DA, Armour JA. Porcine intrinsic cardiac ganglia. Anat Rec A Discov Mol Cell Evol Biol. 2003 Mar;271(1):249-58.

21. Hardwick JC, Mawe GM, Parsons RL. Evidence for afferent fiber innervations of parasympathetic neurons of the Guinea-pig cardiac ganglion. J Auton Nerv Syst 1995;53:166–174.

22. Armour JA. Potential clinical relevance of the 'little brain' on the mammalian heart. Exp Physiol 2008;93:165-176.

23. Navickaite I, Pauziene N, Pauza DH. Anatomical evidence of non-parasympathetic cardiac nitrergic nerve fibres in rat. J Anat 2021;238:20-35.

24. Pauza DH, Rysevaite-Kyguoliene K, Vismantaite J, et al. A combined acetylcholinesterase and immunohistochemical method for precise anatomical analysis of intrinsic cardiac neural structures. Ann Anat 2014;196:430-440.

25. Yuan BX, Ardell JL, Hopkins DA, Losier AM, Armour JA. Gross and microscopic anatomy of the canine intrinsic cardiac nervous system. Anat Rec 1994;239:75-87.

26. Pauza DH, Pauziene N, Pakeltyte G, Stropus R. Comparative Quantitative Study of the Intrinsic Cardiac Ganglia and Neurons in the Rat, Guinea Pig, Dog and Human as Revealed by Histochemical Staining for Acetylcholinesterase. Ann Anat 2002;184:125-36.

27. Pauza DH, Skripka V, Pauziene N, Stropus R. Anatomical study of the neural ganglionated plexus in the canine right atrium: Implications for selective denervation and electrophysiology of the siuoatrial node in dog. Anat Rec 1999;255:271-294.

28. Pauza DH, Skripka V, Pauziene N, Stropus R. Morphology, distribution, and variability of the epicardiac neural ganglionated subplexuses in the human heart. Anat Rec 2000;259:353-382.

29. Rysevaite K, Saburkina I, Pauziene N, Noujaim SF, Jalife J, Pauza DH. Morphologic pattern of the intrinsic ganglionated nerve plexus in mouse heart. Heart Rhythm 2011;8:448-454.

30. Ai J, Epstein PN, Gozal D, Yang B, Wurster R, Cheng ZJ. Morphology and topography of nucleus ambiguus projections to cardiac ganglia in rats and mice. Neuroscience 2007;149:845–860.

31. Batulevicius D, Pauziene N, Pauza DH. Architecture and age-related analysis of the neuronal number of the guinea pig intrinsic cardiac nerve plexus. Ann Anat 2005;187:225-43.

32. Saburkina I, Gukauskiene L, Rysevaite K, et al. Morphological pattern of intrinsic nerve plexus distributed on the rabbit heart and interatrial septum. J Anat 2014;224:583-593.

33. Batulevicius D, Skripka V, Pauziene N, Pauza DH. Topography of the porcine epicardiac nerve plexus as revealed by histochemistry for acetylcholinesterase. Auton. Neurosci 2008;138:64–75

34. Saburkina I, Rysevaite K, Pauziene N, et al. Epicardial neural ganglionated plexus of ovine heart: anatomic basis for experimental cardiac electrophysiology and nerve protective cardiac surgery. Heart Rhythm 2010;7:942–950.

35. Pauza DH, Skripka V, Pauziene N. Morphology of the intrinsic cardiac nervous system in the dog: a whole-mount study employing histochemical staining with acetylcholinesterase. Cells Tissues Organs 2002;172:297-320.

36. Pauza DH, Pauziene N, Tamasauskas KA, Stropus R. Hilum of the heart. Anat Rec 1997;248:322-324.

37. Ulphani JS, Arora R, Cain JH, et al. The ligament of Marshall as a parasympathetic conduit. Am J Physiol Heart Circ Physiol 2007;293:H1629-35.

38. Chiou CW, Eble JN, Zipes DP. Efferent vagal innervation of the canine atria and sinus and atrioventricular nodes. The third fat pad. Circulation 1997;95:2573–2584.

39. Fee JD, Randall WC, Wurster RD, Ardell JL. Selective ganglionic blockade of vagal inputs to sinoatrial and/or atrioventricular regions. J Pharmacol Exp Ther 1987;242:1006–1012.

40. Pauziene N, Pauza DH, Stropus R. Morphology of human intracardiac nerves: an electron microscope study. J Anat 2000;197:437–459.

41. Saburkina I, Pauza DH. Location and variability of epicardiac ganglia in human fetuses. Anat Embryol 2006;211:585–594.

42. Writing Committee Members, Shen WK, Sheldon RS, Benditt DG, et al. 2017 ACC/AHA/HRS guideline for the evaluation and management of patients with syncope: A report of the American College of

Cardiology/American Heart Association Task Force on Clinical Practice Guidelines and the Heart Rhythm Society. Heart Rhythm 2017;14:e155-e217.

43. Pachon JC, Pachon EI, Cunha Pachon MZ, Lobo TJ, Pachon JC, Santillana TG. Catheter ablation of severe neurally meditated reflex (neurocardiogenic or vasovagal) syncope: cardioneuroablation long-term results. Europace. 2011 Sep;13(9):1231-42.

44. Yao Y, Shi R, Wong T, et al. Endocardial autonomic denervation of the left atrium to treat vasovagal syncope: An early experience in humans. Circ Arrhythm Electrophysiol 2012;5:279-286.

45. Sun W, Zheng L, Qiao Y, et al. Catheter Ablation as a Treatment for Vasovagal Syncope: Long-Term Outcome of Endocardial Autonomic Modification of the Left Atrium. J Am Heart Assoc 2016;5:e003471.

46. Aksu T, Guler TE, Mutluer FO, Bozyel S, Golcuk SE, Yalin K. Electroanatomic-mapping guided cardioneuroablation versus combined approach for vasovagal syncope: a crosssectional observational study. J Interv Card Electrophysiol 2019;54:177–88.

47. Hu F, Zheng L, Liang E, et al. Right anterior ganglionated plexus: the primary target of cardioneuroablation. Heart Rhythm 2019;16:1545-1551.

48. Calo L, Rebecchi M, Sette A, et al. Catheter ablation of right atrial ganglionated plexi to treat cardioinhibitory neurocardiogenic syncope: a long-term follow-up prospective study. J Interv Card Electrophysiol. 2020 Aug 6. doi: 10.1007/s10840-020-00840-9.

49. Aksu T, Guler TE, Bozyel S, Yalin K, Gopinathannair R. Usefulness of post-procedural heart rate response to predict syncope recurrence or positive head up tilt table testing after cardioneuroablation. Europace 2020;22:1320-1327.

50. Scherlag BJ, Yamanashi W, Patel U, Lazzara R, Jackman WM. Autonomically induced conversion of pulmonary vein focal firing into atrial fibrillation. J Am Coll Cardiol 2005;45:1878-1886.

51. Po SS, Scherlag BJ, Yamanashi WS, et al. Experimental model for paroxysmal atrial fibrillation arising at the pulmonary vein-atrial junctions. Heart Rhythm 2006; 3:201-208.

52. Pokushalov E, Romanov A, Shugayev P, et al. Selective ganglionated plexi ablation for paroxysmal atrial fibrillation. Heart Rhythm 2009;6:1257-1264.

53. Katritsis DG, Giazitzoglou E, Zografos T, Pokushalov E, Po SS, Camm AJ. Rapid pulmonary vein isolation combined with autonomic ganglia modification: a randomized study. Heart Rhythm 2011;8:672-8.

54. Berger WR, Neefs J, van den Berg NWE, et al. Additional Ganglion Plexus Ablation During Thoracoscopic Surgical Ablation of Advanced Atrial Fibrillation: Intermediate Follow-Up of the AFACT Study. JACC Clin Electrophysiol 2019;5:343-353.

55. Nakagawa H, Scherlag BJ, Patterson E, Ikeda A, Lockwood D, Jackman WM. Pathophysiologic basis of autonomic ganglionated plexus ablation in patients with atrial fibrillation. Heart Rhythm 2009;6: S26-S34.

56. Lin J, Scherlag BJ, Niu G, et al. Autonomic elements within the ligament of Marshall and inferior left ganglionated plexus mediate functions of the atrial neural network. J Cardiovasc Electrophysiol 2009; 20:318-24.

57. Valderrabano M, Peterson LE, Swarup V, et al. Effect of Catheter Ablation With Vein of Marshall Ethanol Infusion vs Catheter Ablation Alone on Persistent Atrial Fibrillation: The VENUS Randomized Clinical Trial. JAMA 2020;324:1620-1628.

58. Derval N, Duchateau J, Denis A, et al. Marshall bundle elimination, Pulmonary vein isolation, and Line completion for ANatomical ablation of persistent atrial fibrillation (Marshall-PLAN): Prospective, single-center study. Heart Rhythm 2020:S1547-5271(20)31218-2.

59. Po SS, Nakagawa H, Jackman WM. Localization of left atrial ganglionated plexi in patients with atrial fibrillation. J Cardiovasc Electrophysiol 2009;20:1186-1189.

60. Kim MY, Sikkel MB, Hunter RJ, et al. A novel approach to mapping the atrial ganglionated plexus network by generating a distribution probability atlas. J Cardiovasc Electrophysiol. 2018 Dec;29(12):1624-1634.

61. Pokushalov E, Romanov A, Shugayev P, et al. Selective ganglionated plexi ablation for paroxysmal atrial fibrillation. Heart Rhythm 2009;6:1257-1264.

62. Sun W, Zheng L, Qiao Y, et al. Catheter Ablation as a Treatment for Vasovagal Syncope: Long-Term Outcome of Endocardial Autonomic Modification of the Left Atrium. J Am Heart Assoc 2016;5:e003471.

63. Pachon M JC, Pachon M EI, Pachon M JC, et al. A new treatment for atrial fibrillation based on spectral analysis to guide the catheter RF-ablation. Europace 2004;6:590-601.

64. Lellouche N, Buch E, Celigoj A, et al. Functional characterization of atrial electrograms in sinus rhythm delineates sites of parasympathetic innervation inpatients with paroxysmal atrial fibrillation. J Am Coll Cardiol 2007;50:1324-1331.

65. Aksu T, Golcuk E, Yalin K, Guler TE, Erden I. Simplified cardioneuroablation in the treatment of reflex syncope, functional AV block, and sinus node dysfunction. Pacing Clin Electrophysiol 2016;39:42-53.

66. Jungen C, Scherschel K, Eickholt C, et al. Disruption of cardiac cholinergic neurons enhances susceptibility to ventricular arrhythmias. Nat Commun 2017;8:14155.

Figure legends

Figure 1

A schematic representation of the cardiac autonomic nervous system in mammalians including the humans. The diagram demonstrates how preganglionic sympathetic axons arise from the spinal cord, synapse with the second sympathetic neurons in the sympathetic chain or intrinsic cardiac ganglia, and proceed as the postganglionic sympathetic axons innervating the diverse cells in the heart (marked in green). The vagus nerve preganglionic axons primarily arise from the dorsal vagal and ambiguus nuclei and synapse with the second parasympathetic neurons within epicardial ganglionated nerve plexus (in brown). Cardiac sensory neurons localize in the dorsal root and vagus nerve sensory ganglia that involve mostly nitrergic axons spread to the heart, while the first four thoracic dorsal root ganglia give rise to the axons containing substance P (SP) and calcitonin gene related peptide (CGRP). The hilum of heart concentrates all nerves reaching this organ. Epicardial ganglionated nerve plexus encloses numerous nitrergic neuronal somata with neuronal nitric oxide (nNO) synapsing the neurons within the brain stem and spinal trigeminal nuclei (mark in blue). Please notice the noradrenaline (NA) and adrenaline synthetizing small intensively fluorescent (SIF) cells adjacent to ganglia. Despite epicardial localization of ganglionated nerve plexi, there is a highly dense network of sensory and efferent nerve fibers in myocardial and endocardial levels.

Figure 2

Comparative drawings of the posterior (A) and anterior (B) views of epicardial ganglionated subplexi (sGPs) in different mammals. Scheme summarizing the descriptions of the disposition, course, and innervation areas of sGPs highlighted in different colors. The dotted and shadowed areas represent the main locations of intracardiac ganglia. While topography of sGPs is generally similar, structural organization of these sGPs varies with the species. Intrinsic ganglia from small mammals are discretely located within the limits of heart hilum on the heart base which is demarcated by dashed lines, in contrast to larger mammals and humans, where these ganglia become progressively scattered and widely distributed. The left dorsal sGP (LDsGP) is labeled in red, middle dorsal (MDsGP) in yellow, dorsal right atrial (DRAsGP) in pink, ventral right atrial (VRAsGP) in blue, the ventral left atrial (VLAsGP) in green, left coronary (LCsGP) in brown, and the right coronary (RCsGP) in magenta, respectively. Other abbreviations: Ao-aorta; IVC-inferior vena cava;

LAA-left atrial appendage; LAzV-left azygos vein; LIPV-left inferior pulmonary vein; LSPV-left superior pulmonary vein; PT-pulmonary trunk; RAA-right atrial appendage; RIPV-right inferior pulmonary vein; RSPV-right superior pulmonary vein; SVC-superior vena cava.

Figure 3

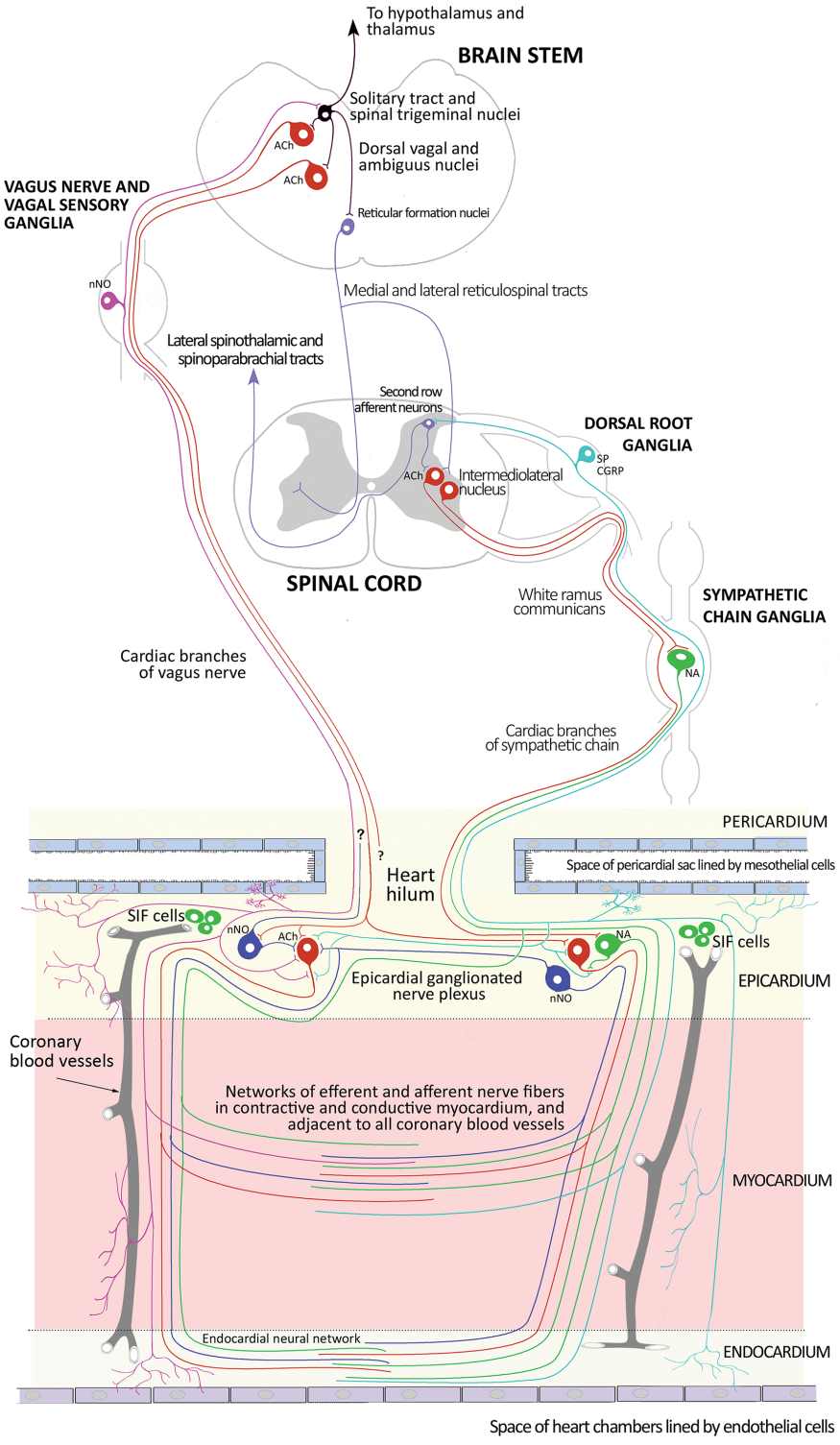
Macrophotographs of the newborn (A) and the adult (B) pressure inflated human left atrium stained histochemically for acetylcholinesterase from the inferior (A) and the lateral (B) views. Note in panel A the parallel epicardial nerves of the LDsGP that extend from and inside the ligament of Marshal (LOM) on the lateral wall of left atrium towards the inferior side of left atrium (arrows) supplying the walls of left pulmonary veins (PV) by thinner ganglionated nerves. White arrows in panel B point to epicardial nerves of the LDsGP that extend on lateral side of the left atrium as the LOM or the left atrial neural fold. Moreover, note the thin epicardial nerves branching from the MDsGP and LDsGP in panel A as well as from LDsGP in panel B that proceed towards the PVs. Dotted line in panel A marks the reflection of pericardium into epicardium on roots of the left PVs. Abbreviations are same as in figure 2.

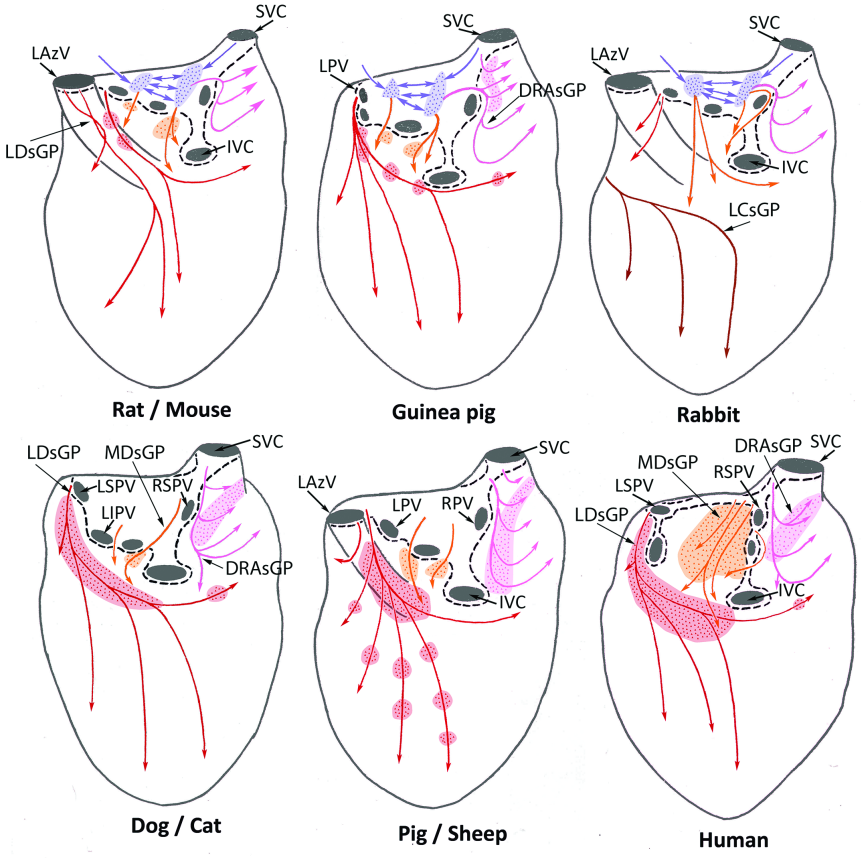
Figure 4

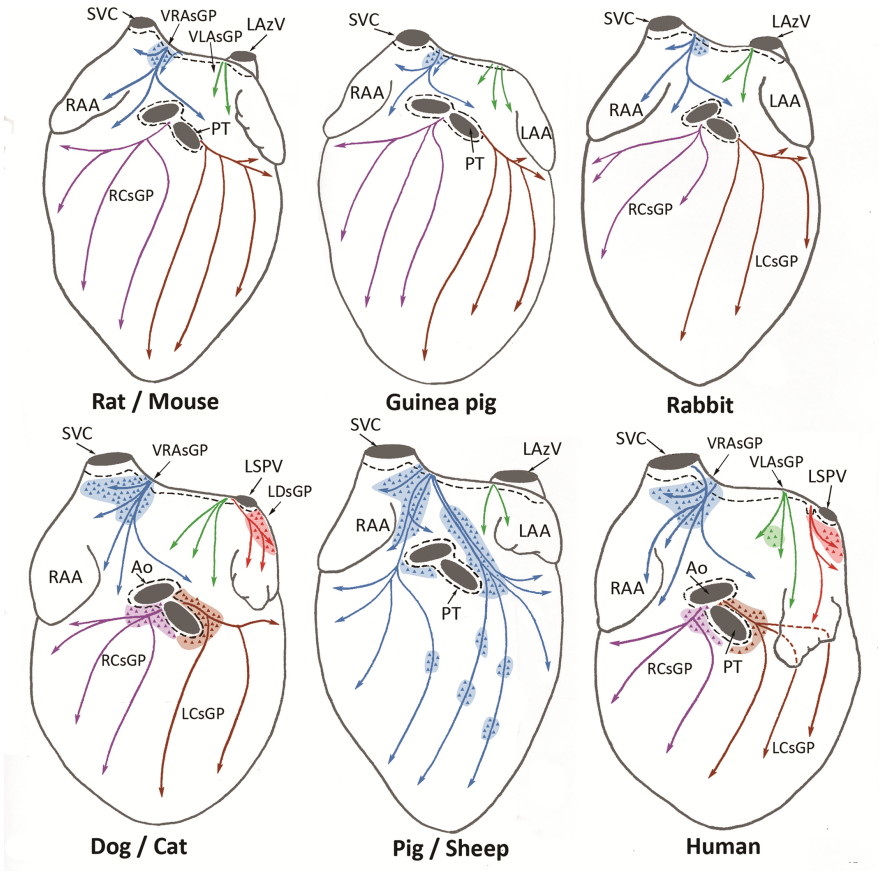
Schematic drawing demonstrates the location, course, and innervation regions of the epicardial ganglionated subplexi from anteroposterior (A) and posteroanterior (B) views on 3D mapping system. Red dotted lines indicate distribution of sGPs. While VRAsGP and DRAsGP supply the sinoatrial node (SN), innervation of the atrioventricular node (AVN) is provided by LDsGP and MDsGP. The limits of heart hilum are demarcated by white dotted lines. CS-coronary sinus; His-His bundle; LOM-Ligament of Marshall. Other abbreviations are same as in figure 2.

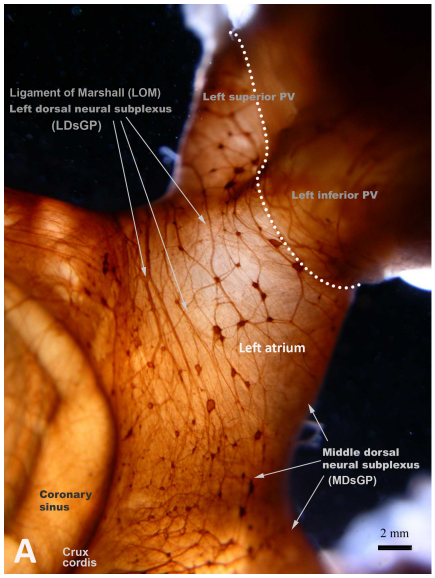
Figure 5

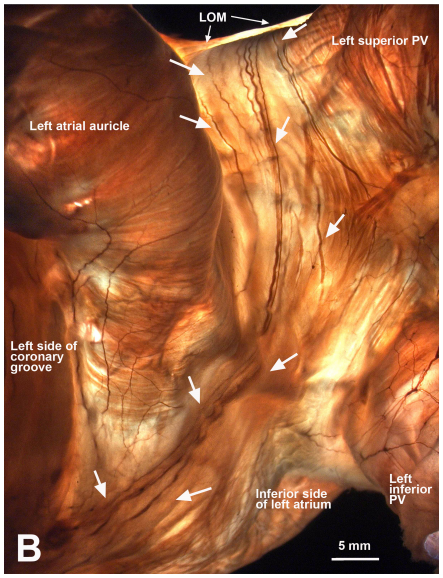
Schematic view of distribution of ganglionated plexi (GPs) based on the presence of fragmented bipolar endocardial atrial electrograms (FEGM) during sinus rhythm (red spheres) or positive response to high-frequency stimulation (HFS) during atrial fibrillation (white spheres) according to the relevant literature. Please compare localizations of GPs according to the nomenclature of Armour (4) with distribution of sGPs of Pauza (28) in Figure 4. CS-coronary sinus; LIGP-left inferior GP; LSGP-left superior GP; PMLGP-posteromedial left GP; RIGP-the right inferior GP; RSGP-right superior GP; VOM-vein of Marshall. Other abbreviations are explained in legend to figure 2.

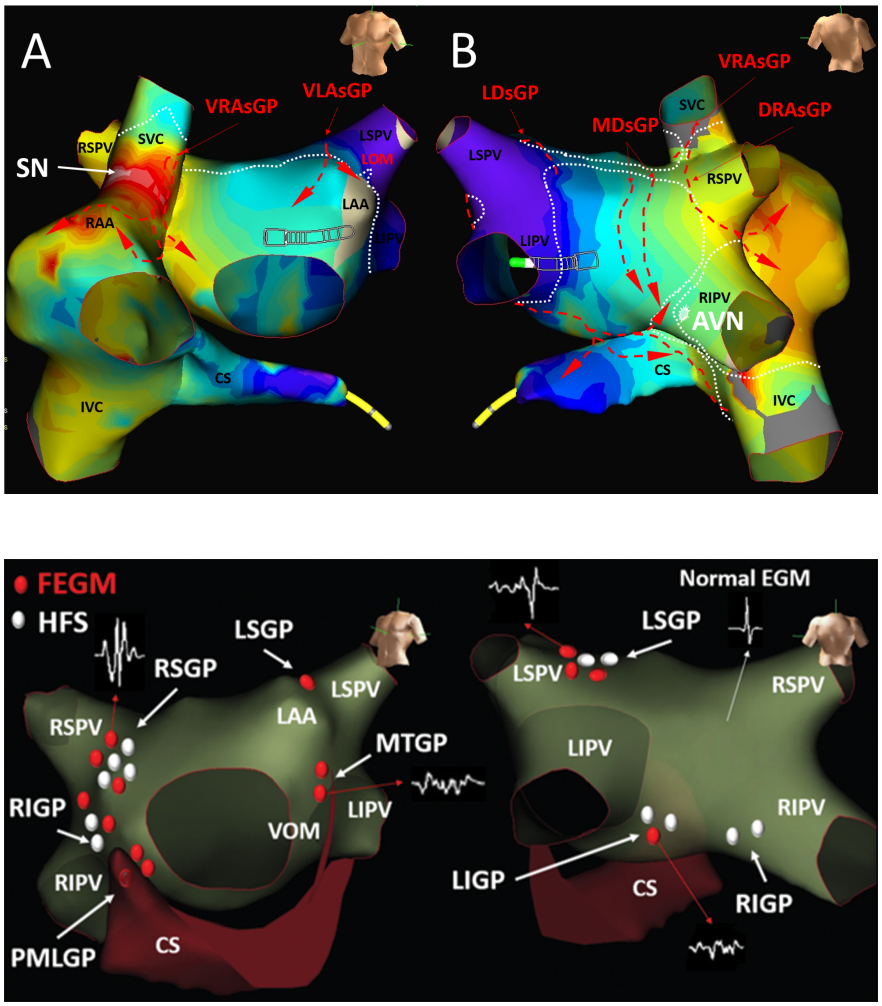












Hosted file

`Table 1.pdf` available at https://authorea.com/users/311249/articles/508788-intrinsic-cardiac-autonomic-nervous-system-what-do-clinical-electrophysiologists-need-to-know-about-the-heart-brain