# Integrity of the Ganglionated Plexi Is Essential to Parasympathetic Innervation of the Atrioventricular Node by the Right Vagus Nerve

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**AV Node Innervation by the Right Vagus Nerve.** ***Introduction:*** **Radiofrequency isolation of pulmonary vein can be accompanied by transient sinus bradycardia or atrioventricular nodal (AVN) block, suggesting an influence on vagal cardiac innervation. However, the importance of the atrial fat pads in relation with the vagal innervation of AVN in humans remains largely unknown. The aim of this study was to evaluate the role of ganglionated plexi (GP) in the innervation of the AVN by the right vagus nerve.**

***Methods and Results:*** **Direct epicardial high-frequency stimulation (HFS) of the GP (20 patients) and the right vagus nerve (10 patients) was performed before and after fat pad exclusion or destruction in 20 patients undergoing thoracoscopic epicardial ablation for the treatment of persistent AF. Asystole longer than 3 seconds or acute R-R prolongation over 25% was considered as a positive response to HFS. Prior to the ablation, positive responses to HFS were detected in 3 GPs in 7 patients (35%), 2 GPs in 5 patients (25%), and one GP in 8 patients (40%). After exclusion of the fat pads, all patients had a negative response to HFS. All the patients who exhibited a positive response to right vagus nerve stimulation (n = 10) demonstrated negative responses after the ablation.**

***Conclusion:*** **The integrity of the GP is essential for the right vagus nerve to exert physiological effects of on AVN in humans.** *(J Cardiovasc Electrophysiol, Vol. 28, pp. 432-437, April 2017)*

*atrial fibrillation, autonomic nervous system, AV node fat pads, ganglionated plexi, parasympathetic system, vagus nerve*

## Introduction

The influence of vagal tone on the electrophysiological properties of the atrioventricular node (AVN) has been well documented. However, the integral links between the AVN and cardiac vagal stimulation remain poorly understood in humans. Several studies in animals, including the work conducted by Hou *et al.,*[1] have demonstrated that the vagus nerve exerts its influence on the AVN through the epicardial fat pads that are primarily located on the posterior wall of the left atrium. These fat pads contain ganglionated plexi (GP), which are located close to the antral area of the pulmonary veins.[2] These regions are ablated during epicardial pulmonary vein isolation for the treatment of atrial fibrillation (AF). A number of studies have reported that pulmonary vein isolation for the treatment of AF may be accompanied by transient sinus bradycardia or AVN block, suggesting that radiofrequency (RF) ablation affects vagal cardiac control in humans.[3-5] However, the importance of the fat pads in the vagal innervations of human AVN is not clear. The present study aimed to understand whether fat pad ablation suppresses the vagal effects of the right vagus nerve (RVN) on the AVN based on selective high-frequency stimulation (HFS)[6] of the RVN and GP in a right monolateral thoracoscopic epicardial ablation for the treatment of AF. Our hypothesis was that fat pad ablation will prevent the effect of HFS of the RVN on the AVN conduction properties, thereby demonstrating the importance of these structures in the vagal control of the AVN.

## Methods

### *Patient Selection*

The present study was approved and supervised by the CHU (Centre Hospitalier Universitaire) UCL (Universite Catholique de Louvain) Namur ethics committee. Patients who underwent epicardial ablation for the treatment of persistent AF between October 2006 and April 2008 were included in the present study according the following inclusion and exclusion criteria. The inclusion criteria were as follows: (1) history of at least one attempt of electrical cardioversion for persistent AF,[7] which was defined as continuous AF for more than 7 days and less than 3 years that was unresponsive

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Correction added on February 14, 2017, after first online publication: The clinical implications section has been revised by the author.

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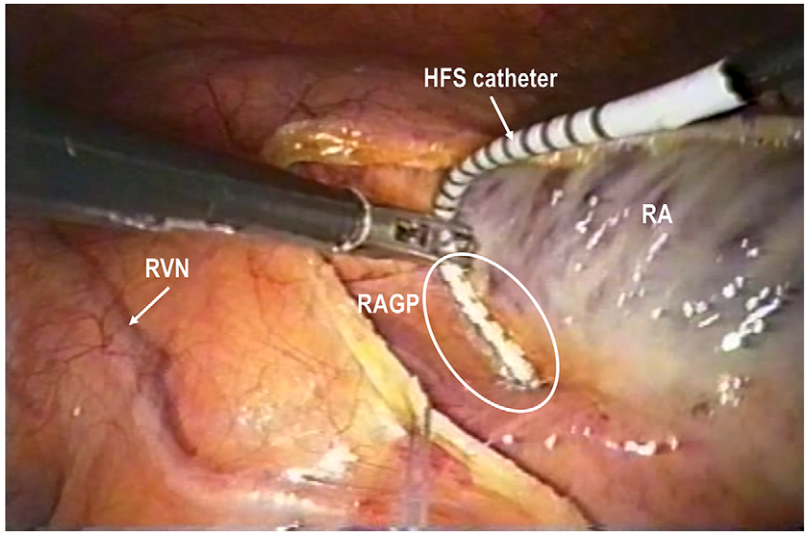


**Figure 1.** *Procedural view of the high-frequency stimulation of the RAGP. HFS = high-frequency stimulation; RA = right atrium; RAGP = right anterior ganglionated plexi; RVN = right vagus nerve.*

to therapeutic doses of several antiarrhythmic drugs; (2) 18 years of age or older; and (3) able to provide written informed consent and participate in all examinations and follow-ups in the present study. The exclusion criteria were as follows: (1) advanced left ventricular impairment (ejection fraction <35% and/or NYHA functional classification score >II); (2) left atrial thrombus; (3) significant chronic obstructive pulmonary disease (COPD); and (4) a history of cardiac or thoracic surgery.

A total of 20 consecutive patients were finally included in the present study according to these criteria. All of the included patients were able to undergo GP HFS before and after ablation at the same sites. Antiarrhythmic medications were interrupted for over 5 half-lives prior to the ablation except for amiodarone in 5 patients. All of the patients had AF at the beginning of epicardial ablation procedure, and none of these patients were converted to sinus rhythm during the intervention.

### *Operative Techniques*

The epicardial ablation procedure was performed under general anesthesia according to previously reported methods.[8] A double lumen endotracheal tube was used to facilitate unilateral ventilation. The patient was engaged in the supine position with a rotation of 30 degrees to the left. Three working ports were used, including two 10-mm incisions in the fifth and the sixth intercostal spaces and a 5-mm incision in the fourth intercostal space at the anterior axillary line. Insufflation of $CO_2$ was started at 8 mmHg. The pericardial reflection of the superior and inferior caval vein was dissected to access the transverse and oblique sinus. High-frequency stimulation of the GPs and the RVN was performed prior to the ablation (Fig. 1). A multipolar surgical ablation catheter was introduced into the transverse sinus beneath the SVC and guided around the 4 pulmonary veins. Two different sources of monopolar energy were used in the present study, including a Flex 10 microwave ablation device (Guidant Cardiac Surgery, Santa Clara, CA, USA; 65 W and 120 seconds) and a Cobra XL Cooled Surgical Probe (Estech, San Ramon, CA, USA; 65 °C, 60 W, and 150 seconds). High-frequency stimulation of the GPs and the RVN was repeated after the ablation.

### *Influence of the GP on the Atrioventricular Node Conduction Properties*

We evaluated the effects of direct HFS[6] (FIAB Programmable Cardiac Stimulator 8817, PSA Model 3150–200; St. Jude Medical, St. Paul, MN, USA) with the following settings: 20 Hz, 2 milliseconds, and 20 mA on the right anterior (RAGP), the right inferior (RIGP), and the left superior ganglionated plexi (LSGP) using an electrophysiological catheter (Duo-Dec Super LRG Curl, ref: 401904, St. Jude Medical). The left inferior ganglionated plexus was not evaluated in this study because of the considerable difficulty in accessing it using the current unilateral approach. Considering the normal coefficient of variation of R-R intervals in AF,[9,10] a positive response to HFS was defined as an acute prolongation of the R-R intervals in AF patients by over 25% within a minimum of 5 seconds compared with the previous R-R intervals or an asystole of over 3 seconds.[6] After posterior atrial wall isolation and dissection of the GP, HFS of GPs was repeated at the sites where positive responses were obtained prior to the ablation. For negative responses, these sites were extensively explored to validate the results.

### *Vagal-Atrioventricular Node Conduction*

HFS of the thoracic portion of the right vagal trunk was conducted using the same multipolar electrophysiological catheter. The responses to HFS were defined as described above. The vagal nerve was stimulated again after RF ablation.

## Results

The mean age and body mass index of the 20 patients (19 male and 1 female) were 56 ± 9 years and 29 ± 4 kg/m², respectively. The mean AF history of the 20 patients was 7 ± 5 years.

### *Preablation Results*

A positive response to the GP HFS was obtained from all 3 GPs in 7 patients (35%), 2 GPs in 5 patients (25%; RAGP and

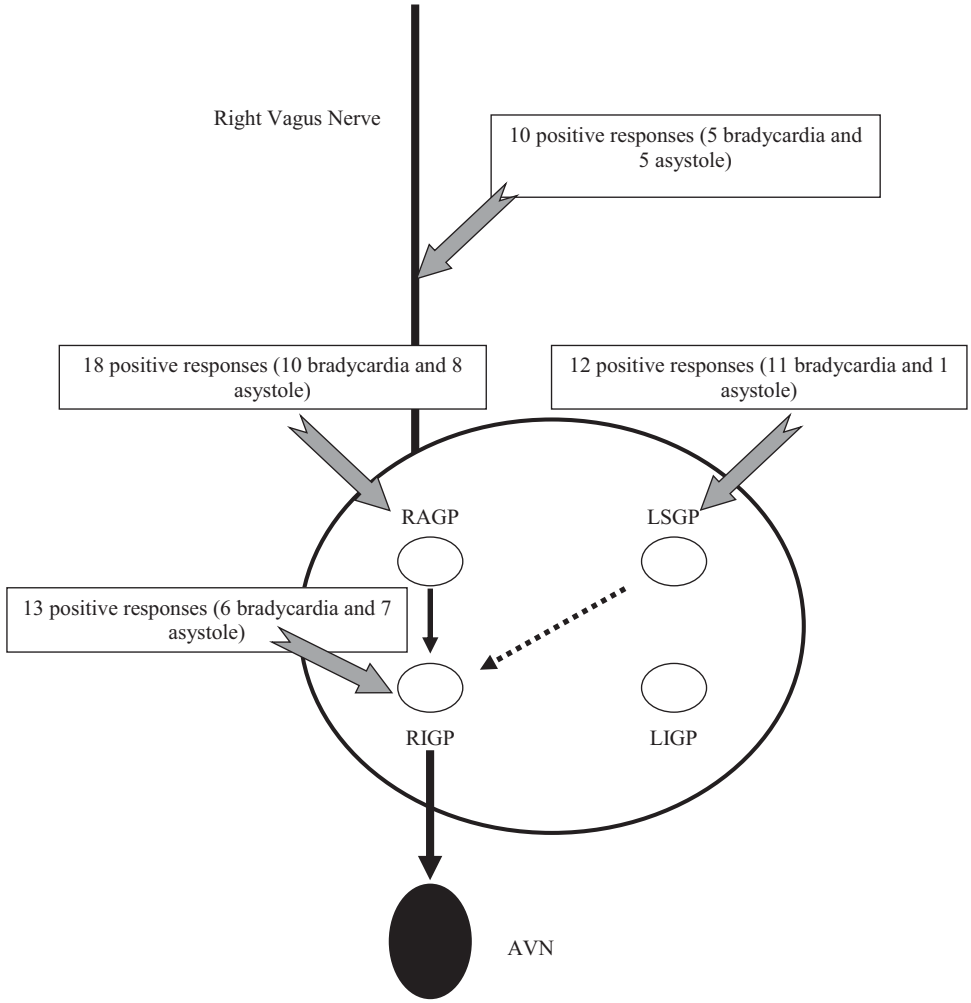


**Figure 2.** *Schematic representation of the posterior wall of the left atrium, the vagus nerve, and the four ganglionated plexi showing the distribution of responses to high-frequency stimulation. The arrows suggest the relative importance of each GP to the other GPs and the AVN (tick lines and dotted lines indicate strong and weak effects, respectively). These arrows must be interpreted with caution because the study protocol does not permit sequential ablation of each GP. AVN = atrioventricular node; RAGP = right anterior ganglionated plexi; LSGP = left superior ganglionated plexi; RIGP = right inferior ganglionated plexi; LIGP = left inferior ganglionated plexi.*

RIGP in 4 patients and RIGP and LSGP in 1 patient), and one GP in 8 patients (25%; RAGP). The type and distribution of the responses are summarized in Figure 2. Figure 3 illustrates the positive responses to HFS in the 3 GPs in one patient. In addition, HFS of the RVN was performed in 10 patients with positive responses.

### *Postablation Results*

After ablation, we were unable to induce any significant positive response to HFS of the GP in the patients who demonstrated a positive response to the HFS prior to the dissection and ablation. In addition, no patients had positive responses to the HFS of the vagus nerve after the epicardial ablation procedure.

## Discussion

In the present study, we evaluated the interactions among the RVN, the GP, and the AVN.

### *Interactions Between the GPs and the AV Node*

Prior to the ablation, the positive responses to HFS from the RAGP were higher than that from the RIGP and LSGP (Fig. 2). The different responses to HFS among these GPs could be explained by the following reasons. First, certain GPs were difficult to access using the unilateral right thoracoscopic approach with the pacing catheter. Second, a larger dissection area was required to reach the RIGP and LSGP than to reach the RAGP. The dissection of the pericardial reflections and the surgical techniques could have interrupted certain efferent innervations of these GPs.

A negative response to HFS was obtained in all of the GPs after ablation. This observation is supported by previous studies reported by Quan *et al.*[11] and Shah *et al.*,[12] who demonstrated that the RIGP selectively innervated the AVN in humans. Although other GPs were not tested in these 2 studies, Hou *et al.*[1] identified a large interactive network among different GPs in dogs and showed that this network serves as an "integrated center" of the cardiac autonomic innervation.

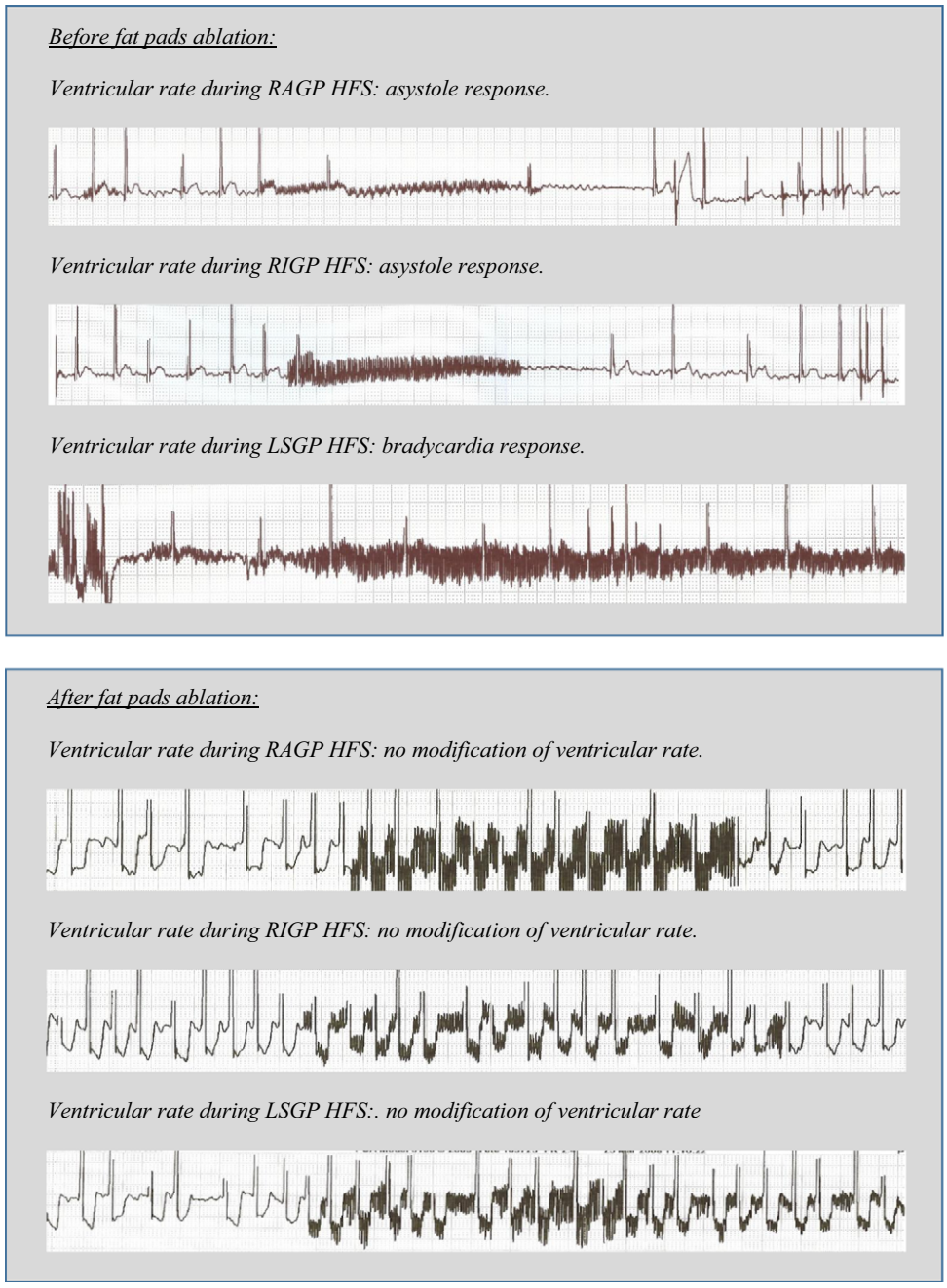


**Figure 3.** *Illustration of the different responses to HFS as a function of the site of stimulation in one patient before and after ablation. RAGP = right anterior ganglionated plexi; LSGP = left superior ganglionated plexi; RIGP = right inferior ganglionated plexi; HFS = high-frequency stimulation. [Color figure can be viewed at wileyonlinelibrary.com]*

These results are consistent with our findings that HFS of the RAGP and LSGP could also influence the AVN in addition to the RIGP. Even if the relative role of the different GPs must be interpreted with caution because of the absence of sequential ablation of the different GPs in this study, the higher relative asystolic response (53%) under direct HFS stimulation of the RIGP than under direct HFS stimulation of the LSGP (9%) and the RAGP (44%) could suggest that the RIGP was the last GP in the human GP network. Moreover, the influence of the RAGP on the AVN appears to be more important than its influence on the LSGP (Fig. 2), which is similar to the results of Hou *et al.*[1]

### *Interactions Between the RVN and the AVN*

A positive response from the AVN to HFS of the RVN was obtained in 10 patients prior to the ablation, although such responses were not obtained in any patient after the ablation. The absence of any alteration in the ventricular rate in response to HFS of the RVN after the ablation of GP suggests that the RVN is not directly connected to the AVN and that the integrity of the GP is required to produce vagal effects on the AVN. The right unilateral thoracoscopic approach did not allow for HFS of the left vagus nerve. Nevertheless, our findings for the RVN are consistent with

the previous observations of the absence of a direct pathway between both the right and left vagus nerves and the AVN.[1]

Thus, we identified the functional neural pathway between the RVN and the AVN and demonstrated that the integrity of the GP is a mandatory interlink.

## Clinical Implications

During AF ablation, the GPs close to the ostia of the veins sometimes undergo unintended ablation. The imbalance of the cardiac ANS has been known to play a significant role in the genesis and perpetuation of arrhythmia. Because the cell bodies of intrinsic parasympathetic innervation of the heart are mainly located on the atrial GP, ablation of these GPs affects atrial and ventricular intrinsic innervation. The impact of GP ablation on atrial and ventricular arrhythmias appears to have opposite effects.

### *Atrial Level*

A meta-analysis performed at the atrial level demonstrated a positive benefit of GP ablation combined with PV isolation on AF recurrence compared with GP ablation or PV isolation alone (at short and relatively long-term intervals). However, this issue remains controversial because incomplete ablation of the GP can increase the vulnerability of the atria to AF 13–15 and denervation is likely transient. 14 The acute or chronic impacts of GP ablation on the denervation of AVN in humans have not been previously studied.

After ablation of persistent AF, one of the most troublesome complications is the development of atrial tachycardia with high ventricular response. 16 Nayebpour et al. 17 demon-strated that the impact of the parasympathetic tone on the AVN conduction properties is particularly important at rapid rates. This study suggested that the ablation of the GP that led to parasympathetic denervation of the AVN could play a role in the high ventricular rate response of atrial tachycardia after AF ablation. Therefore, our study provides new insights on that particular mechanism.

### *Ventricular Level*

At the ventricular level, the imbalance between the sympathetic and parasympathetic intrinsic cardiac autonomic nervous system increased the incidence of ventricular arrhythmias and facilitated ventricular fibrillation, especially in ischemic cardiac disease. In patients with ischemic cardiac disease, ablation of these GPs must be performed with caution because of the increased risk of ventricular arrhythmias.

## Limitations

The minimally invasive right-sided thoracoscopic approaches applied in the present study prevented sequential ablation of the GPs and HFS of the left vagus nerve. Therefore, we were not able to precisely evaluate the specific interactions among the GPs and analyze the role of the left vagus nerve.

Our results indicate that further studies should be performed to evaluate the long-term influence of GP ablation on the electrophysiology of the AVN.

## Conclusions

The right-sided unilateral thoracoscopic approach applied here to perform epicardial PV isolation was also used to explore the role and influence of the GP on the complicated vagal innervation of the heart.

The functional neural pathway between the RVN and the AVN was identified, and the integrity of the GPs represents a mandatory interconnected network.

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