Development of a Multi-channel System for Intrinsic Cardiac Neural Recording

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***Abstract*— Recent clinical evidence suggests that abnormal neural input can contribute to the onset and perpetuation of atrial arrhythmias, such that neural elements have become potential targets for ablation. A better understanding of the influence of the cardiac autonomous nervous system is required to improve therapy. We have developed a multi-channel system to record neural activity simultaneously at different intra and pericardiac locations. The paper presents the specific requirements to be met for recording neuronal extracellular potentials in these repertoires of neurons and the solutions that were adopted.**

I. Introduction

ATRIAL arrhythmias, more particularly atrial fibrillation (AF) have become the most frequently encountered cardiac arrhythmias[1]-[4]. Despite recent progress, there still is a striking need to improve the treatment of AF [5]. Surgical approaches, such as the Maze procedure or catheter ablation [4], [6] target the myocardial tissue. Until recently, neuronal influences were not considered. However, recent clinical evidence suggests that during successful circumferential pulmonary vein ablation to treat AF, there is a concomitant destruction of the distal branches of the vagosympathetic nerve complex [7]. It has also been suggested that selective ablation of pulmonary vein ostial sites associated with autonomic responses can improve the outcome of anti-AF surgery [8].

Classical view of the cardiac neural control describes the regulatory control of the heart by the sympathetic and parasympathetic branches of the cardiac autonomic nervous system (CANS). While the sympathetic efferents tend to increase the cardiac indices, the parasympathetic branch tends to depress them. The sympathovagal balance hypothesis suggests that the activation of one branch is accompanied by a deactivation of the other branch by interactions occurring both at the central and cardiac myocyte levels. Recently, it has been proposed that interaction between the parasympathetic and sympathetic system can also be mediated by a network of intrathoracic and intracardiac interneurons [9], [10]. The intrinsic cardiac nervous system would act as a supplementary level of interaction between the sympathetic and parasympathetic CANS. It is not clear, however, whether such interactions provide positive, negative or mixed feedback between the components of the CANS. A better understanding of the connectivity and interactivity of the components of the CANS is thus necessary to improve the treatment and management of atrial arrhythmias. This requires a device for simultaneously recording extracellular potentials produced by neurons at different locations. The goal of this project is to develop such a device to be used in open-chest patients and canine preparations.

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II. Requirements in recording neural activity

As a starting point, two loci known to be densely populated by cardiac neurons were targeted for recording: the stellate ganglions and the so-called fat pads, on the epicardium of the atria. The parasympathetic efferent neurons originate in medulla, follow the spinal cord, from which they emerge to synapse in the left and right stellate ganglions. The post-ganglionic efferent neurons then project onto the atria and ventricles. In dogs, the stellate ganglions appear typically as 3mm x 4 mm x 3 mm nodes surrounded by a thick layer of conjunctive tissues. At the surface of the atria, ganglionated plexi, which may contain up to 200 neurons [9], [11] can be found in 1-4 mm thick fat pads lying on the epicardium of the atria. For these and other neurons found directly on the epicardium, the electrical activity of the underlying myocardium associated with the cardiac contraction produces large electrical potentials that interfere with the neuronal signals.

For proper recording at these two loci, the data acquisition system should have:

1) Large dynamic range to prevent saturation of the amplifiers when the heart activates,
2) Thin and stiff electrodes to cross the conjunctive tissue barrier without bending,
3) Multiple electrodes to record from many neurons or/and ganglions at the same site,
4) Short electrode leads to place the headstage close to the recording site in order to increase the signal to noise ratio,
5) Multiple headstages to record simultaneously at different locations,
6) Reference electrode close to each set of recording electrodes to reduce artifacts associated with cardiac activation and contraction,
7) Design that prevents electrodes from reaching the myocardium in the fat pads,
8) Low sensitivity to motion artifacts particularly for recording on the myocardium.
9) Optimised filtering to reduce artifacts due to atrial activation and contraction,
10) High sampling rate to discriminate among waveforms associated with different neural units, which action potentials last a few ms,
11) Online signal processing to be able to judge during the experiment whether the activity of the neurons is related to some aspect of cardiac physiology or to determine the interaction between neurons,
12) Simultaneous recording of pressure, ECG, or other physiological signals to correlate with the neuronal activity.

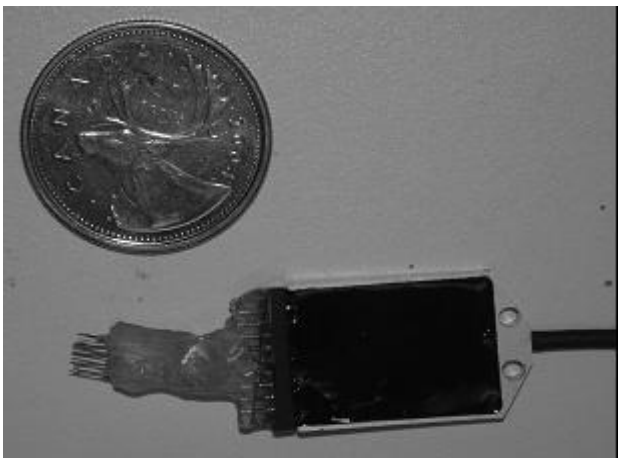

Figure 1 Eight channels headstage (Multi channel systems), with nine tungsten electrodes

III. SYSTEM

Each basic module to record at one site, consists of nine tungsten 2 cm long electrodes coated with epoxy resin with initial impedance of 9–12 MΩ at 1,000 Hz (Frederick Haer, ME, USA). The insulation at the tip of the electrode used as a reference is manually removed to lower its impedance. The electrodes are positioned on a 4-10 mm^2 support (figure 1). Tungsten electrodes are used, even if they are not the optimal for recording from biological tissue, because they are thin and stiff. Theses properties are needed to cross the conjunctive tissue barrier and prevent bending which would change the spatial configuration of the electrode array.

The electrodes are manually curved, crimped and glued in epoxy such that a 1 mm length protrudes from the epoxy. Due to the small mass and volume of the device, applying a small pressure when the array is in contact with the epicardium is enough to make it follow the movement of the myocardium, thus reducing relative movement between electrodes and neurons. This also prevents the friction at the tip of the recording electrodes that can deteriorate the insulation coating.

Each array of nine electrodes is connected to a battery powered 8 channels headstage (MPA-8 Multi Channel Systems, De). Signals from eight electrodes are differentially amplified 10X relative to the reference electrode. The reference electrode is located less than 1 mm apart from the other electrodes. With this configuration, the reference electrode can also record neuronal signals, which complicates signal processing but reduces the artifacts associated with the activation of the heart. Theses artifacts can be up to 3 mV/mm for sites close to the myocardium.

The amplified signals are conditioned by a first order analog high pass filter (-3db @ 1 Hz) to remove the DC component. The signals are then digitalized at 16384 Hz with a 24 bits (+/- 262 mV input dynamic range) analog to digital converter (Active-two, Biosemi Nl). The battery powered A/D converter has a driven right leg circuit to reduce common-mode voltage and a fifth order sinc low pass filter (-3dB @ 3275 Hz). Digitized signals are stored on BDF data format (24 bits version of the European data format). Since the system has a very large dynamic range, the unfiltered signals can be used to measure the electrical activity of the heart and to monitor electrode contact problems. The high pass filtered signals represent the neural activity. On the filtered signal, the noise level of the system measured with low impedance electrodes was less than 3 μV rms. On standard recording, the main source of noise is the thermal noise caused by the high impedance electrodes.

Three optically isolated inputs are included in the system to record other physiological signals. Presently, we record:

1) Arterial pressure on the descending aorta (Kobe pressure transducer)

2) Respiratory pressure (Kobe pressure transducer)

3) Lead II ECG signal (grass electrodes).

These three signals are amplified by an external unit (Nihon Kohden) before being sent to the ADC.

IV. SIGNAL PROCESSING

The digitized signals are processed with a program written in Labview (National instrument, Tx, USA). Second order 300 Hz high pass filtering and threshold peak detection (±6 rms noise level) is applied to each signal. The display of the data is time aligned with a trigger derived

from the ECG, the arterial pressure, the respiratory pressure or the signal from one of the other electrodes. The histogram of the firing times in each channel, with respect to the trigger, can confirm if temporal correlation exists between the channels or with the signal used as trigger.

Additional is done offline using the program Spike (vers. 1.14, Cambridge electronic design UK). At this stage, when needed, portion of the signals included in a ±5 ms window around each activation of the myocardium are removed to eliminate artifacts. Principal component analysis is then done to discriminate between the waveforms associated to different neurons in the signal from each channel.

V. Methodology

Experiments were performed in accordance with the Canadian Council for Animal Care and approved by an institutional animal care committee. Adult mongrel dogs (either sex), weighing 25–40 kg, were anaesthetized with Na thiopental (25 mg/kg iv, supplemented as required), intubated and maintained under positive-pressure ventilation. After surgery, anesthetic was changed to achloralose (25–50 mg/kg iv bolus supplemented with 25 mg/kg iv as required). Following bilateral thoracotomy, the stellate ganglion was exposed and the pericardium incised, exposing the heart. In some instances, atrioventricular block was induced by formaldehyde injection into the AV node to separate atrial from ventricular events. Right ventricular pacing (60 beats/min) was then applied to ensure adequate cardiac input. To induce atrial tachyarrhythmia, bursts of electrical stimuli (1-2 mA, 1-ms duration, and 5 ms pulse interval) were applied to individual pericardiac nerves during the refractory period of the closest atrial region.

VI. Results

Previous experiments have shown that stimulating the mediastinal nerves that course over the ventral and ventrolateral surfaces of the caudal-most superior vena cava can induce bradycardia followed by AF [12] (figure 2).

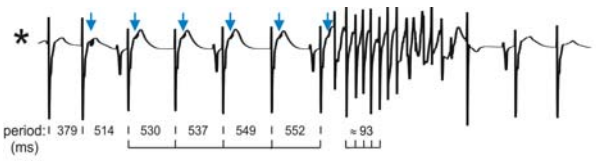

Figure 2. Atrial bradycardia and AF induced by burst stimulation of a right-sided, extrapericardial mediastinal nerve applied during the atrial repolarization phase (blue arrows), as seen from a unipolar electrogram recorded on the right atrium. Atrial activation intervals (in milliseconds, below the trace) increased immediately upon stimulus application.

Figure 3 shows the firing rate of two channels recorded from the right stellate ganglion during the protocol. The firing rate of one channel increases during and shortly after the end of the AF, while the other shows a burst of activity ~20 s after the end of AF. Since the arterial pressure was not changed during or after AF, this suggests the existence of a feedback loop to the stellate ganglion involving cardiac afferent neurons.

Figure 4 shows the result of another measurement from the right stellate ganglion. In this case, the AV node was left intact and recording was done without applying neural stimulation. The figure shows the distribution of the spiking time recorded from two channels, referenced to the activation of heart, measured from the QRS complex of the ECG. The global firing rate was 105 spikes/minute. Waveform analysis has shown that four neurons were picked by one channel. Two of these neurons had a random firing pattern with respect to the QRS (top panel) while two neurons were synchronized with the QRS complex.

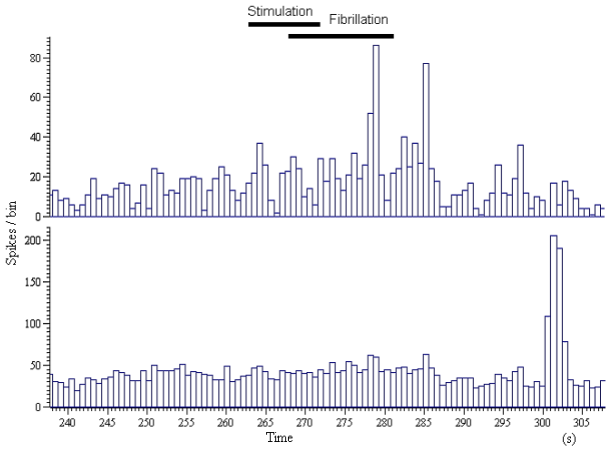

Figure 3 Neuronal activity on two different electrodes measured on the RAGP during the protocol described in figure 2. Each bin represents 1.4 s

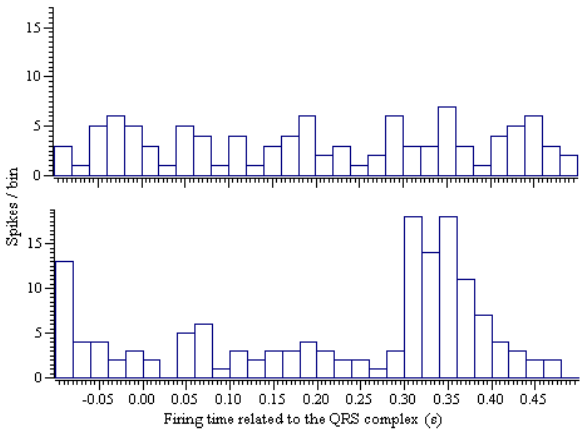

Figure 4 Distribution of the firing time related to the QRS complex for a recording of 2 min on the right stellate ganglion. Top panel: distribution for two neurons with random firing pattern. Lower panel: distribution for two cardiac related neurons. Each bin represents 20 ms.

Figure 5 shows a recording from the right atrial ganglion complex, a fat pad lying on the right atrium where spikes associated with neural firing could reach 250 μV.

Simultaneous spikes were recorded with decaying amplitudes on adjacent electrodes (top circle, in fig.5). In some cases, a temporal delay was repeatedly observed between the activations of adjacent channels (bottom circle, in fig. 5), which suggests that the system could be used to observe the propagation of information between two loci in specific circumstances. Programs to analyze theses aspects will be developed in a near future.

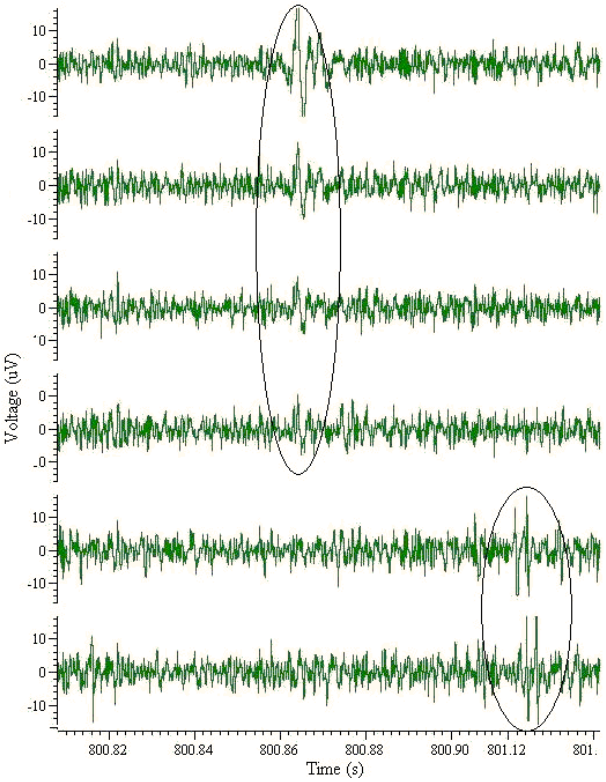

Figure 5 Neuronal activities measured on the right atrial ganglionic complex. Upper circle: firing of neurons with decaying amplitude with distance. Lower circle: neurons with a delay in firing time upon two channels

VII. Conclusion and Perspectives

The device we have developed allows multi-channel recording over several sites of the CANS to analyze neural activity. In parallel with this project, our group has initiated a protocol to measure neuronal activity of the different component of the CANS by functional MRI, which could give neuronal information on a macroscopic level. These results will be compared with the results obtained with this system. When completed and approved, we expect to use this system for clinical studies in patients undergoing open-heart surgery. The long-term objective of our work is to develop a system that could be mounted on a catheter to target specific areas for ablation.

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References

[1] W. B. Kannel, and P. A. Wolf, "Epidemology of Atrial Fibrillation" in *Atrial Fibrillation: Mechanisms and Management,* Raven Press Publishers, New York, pp. 81-92, 1992

[2] AS Go, EM Hylek, and KA Phillips, Y Chang, LE Henault, JV Selby, DE Singer, "Prevalence of diagnosed atrial fibrillation in adults", *Jama*, Vol. 285, pp. 2370-2375, 2001

[3] PA Wolf, JBM Mitchell, CS Baker, WB Kannel, RB D'Agostino, "Impact of atrial fibrillation on mortality, stroke and medical costs", *Arch. Intern. Med.*, Vol.160, pp.49-57, 2000

[4] RG Hart, and JL Halperin, "Atrial fibrillation and stroke: concepts and Controversies", *Stroke*,Vol.32, pp.803-808, 2001

[5] PD Pacifico, and A Henry, "Ablation fo atrial fibrillation: are cures really achieved", *J. Am. Coll. Cardiol*., Vol.43, pp.1940-1942, 2004

[6] B Finta, DE Haines, " Catheter ablation therapy for atrial fibrillation", *Cardiol Clin*, Vol.22, pp.127-145, 2004

[7] C Paponne, V Santinelli, F Magusso et al., "Pulmonary vein denervation enhances long term benefit after circumferential ablation for paroxysmal atrial", *Circulation*, Vol.109, pp.327-334, 2004

[8] M.Platt and R. Mandapati, "Limiting the number and extent of radiofrequency applications to terminate atrial flutter and subsequently prevent its inducibility", *Heart Rhythm*, Vol.1 Supp 1, p. S11, 2004

[9] JA Armour, DA Murphy, BX Yuan, S Macdonald, DA Hopkins, "Gross and microscopic anatomy of the human intrinsic cardiac nervous system", *Anat. Record*., Vol. 247, pp.289-298, 1997

[10] RC Arora, M Waldmann, DA Hopkins and JA Armour, "Porcine intrinsic cardiac ganglia",*Anat Rec A Discol Mol Cell Evol Biol*, Vol. 271, pp.249-258, 2003

[11] BX Yuan, JL Ardell, DA Hopkins, AM Losier, JA Armour, "Gross and microscopic anatomy of the canine intrinsic cardiac nervous system", *Anat Rec,* Vol .239(1), pp. 75-87, 1994

[12] JA Armour, LP Richer, P Page, A Vinet, T Kus, M Vermeulen, Nadeau R, Cardinal R., "Origin and pharmacological response of atrial tachyarrhythmias induced by activation of mediastinal nerves in canines", *Auton Neurosci*. 118:68-78,2005