Electrophysiological Substrates of Atrial Fibrillation: A Frequency Domain Study of Intra-cardiac Electrograms

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By

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Preface

Over the last decade, there has been a surge of renewed interest and research into the treatment of atrial fibrillation (AF), spurred on by the landmark studies of catheter ablation for AF. Yet despite over 100 years of studying this arrhythmia, it remains incompletely understood and clinical management remains controversial. Nevertheless, invasive and non-invasive treatment of AF continues to advance at pace, resulting in a persisting gap between effective understanding and therapy.

Studying the characteristics of AF electrograms to identify potential substrates and their significance in maintaining AF is an area of research that could help to provide further insight into the mechanisms of AF and could also lead to identification of targets for delivery of therapy via catheter ablation. The studies that are described in this thesis are aimed at contributing to this objective, and involve mainly the use of frequency domain analysis of AF electrograms. The effect of pharmacological and ablative interventions on AF electrograms will also be examined.

The thesis will begin with a brief abstract of the original research studies carried out during my period of research. This will be followed by an introduction of AF, detailing the key concepts and understanding that we have of this arrhythmia to date, as well as the evolution of its treatment, in particular catheter ablation. Full methodology and details of the study results will follow, concluding with a summary chapter.

I would like to take this opportunity to thank all patients who participated in the studies within this thesis. I am also grateful to Drs Ng and Stafford for providing unwavering support and valuable advice throughout my time in research. I would also like to express my appreciation for the patience shown by the Glenfield cardiac

physiologists during signal acquisition and to industry technical engineers, who provided technical help and support for the research. In addition, I would like to thank departmental statistician, Suzanne Stevens for her statistical input, my colleagues in the research group, Drs Jeilan, Kundu and Nicolson for their general support and advice. Finally, special thanks goes to my wife for her support and patience while I work to complete this thesis.

JH Tuan

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Statement of Originality and Involvement

All data collection and analysis carried out for the thesis are entirely my work. The concept and design for each study were formulated between myself, and my supervisors, Dr G.André Ng and Dr Peter Stafford.

Thesis Abstract

The mechanisms responsible for maintenance of AF remain poorly understood. This thesis examines the frequency domain characteristics of AF in order to gain further insights into this arrhythmia. Through a series of studies involving patients undergoing catheter ablation for atrial fibrillation, intra-cardiac electrograms of AF were collected and analysed using Fast Fourier Transform to derive frequency domain parameters of dominant frequency (DF) and organization index (OI). It was found that intravenous flecainide reduced DF of AF, but only an associated increase in OI was predictive of successful return to sinus rhythm. In another study of patients having catheter ablation for persistent AF, a higher OI post-ablation was found to be associated with medium-term freedom of AF, suggesting that OI may be a useful guide to determine the extent of radiofrequency ablation needed. The effects of vagal blockade with atropine were also studied and compared with that of catheter ablation using a stepwise strategy of isolating the pulmonary veins, linear ablation and complex fractionated electrogram ablation, without deliberately targeting ganglionated plexi. This showed that atropine reduced DF and increased OI of AF electrograms, while decreasing mean RR intervals, standard deviation of RR intervals and 5^{th} percentile of RR intervals. The directional changes of all the above parameters mirrored that of catheter ablation, suggesting that vagal blockade and catheter ablation not deliberately aimed at autonomic tissue can have similar effects on the frequency spectrum of AF, probably mediated through modulation of the autonomic tone. The relationship of regional DF and electrogram complexity as assessed by automated measurement of complex fractionated electrogram – mean (CFE-mean) were also compared, pre and post-ablation of the left atrium. There appeared to be only a modest correlation between the two and this was further weakened following ablation, suggesting that these are possibly separate substrate entities.

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List of Abbreviations

AAD	Anti-Arrhythmic Drugs
AF	Atrial Fibrillation
CFE	Complect Fractionanted Electrograms
DF	Dominant Frequency
ECG	Electrocardiogram
FFT	Fast Fourier Transform
LAA	Left Atrial Appendage
OI	Organization Index
PVI	Pulmonary Vein Isolation
PV	Pulmonary Vein
RAA	Right Atrial Appendage
WACA	Wide Area Circumferential Ablation

Abstracts and Publications arising from this Thesis

Tuan J, Osman F, Jeilan M, Kundu S, Mantravadi R, Stafford P, Ng G.A. Increase in Organization Index predicts Atrial Fibrillation termination with Flecainide post-ablation – spectral analysis of intracardiac electrograms. Europace 2010; 12(4):488-93[1]

- Also presented as oral abstract at American Heart Association Annual Scientific Conference, Nov 2008

Tuan J, Osman F, Jeilan M, Kundu S, Mantravadi R, Stafford P, Ng G.A. Spectral Analysis and outcomes after Catheter Ablation for Atrial Fibrillation. Heart Rhythm 2008; 5(5 suppl):S267

- Presented as poster at Heart Rhythm Society, May 2008

Tuan J, Jeilan M, Kundu S, Nicolson W, Stafford P, Ng G.A. Catheter ablation for persistent AF: Changes in Autonomic Tone and Frequency Spectrum mirror that of vagal blockade. Europace 2009;11(Suppl 4):iv9

- Oral presentation at Heart Rhythm Congress, October 2009

Tuan J, Jeilan M, Kundu S, Nicolson W, Chung I, Stafford P, Ng G.A. Regional fractionation and dominant frequency in persistent Atrial Fibrillation: Effects of left atrial ablation and evidence of spatial relationship. Europace 2011; 13(11):1550-56

- Also presented as oral abstract and winner of Young Investigators Prize at Heart Rhythm Congress, October 2009

Please refer to Appendix for more details

Chapter 1
Introduction

1.1 Brief History and Background

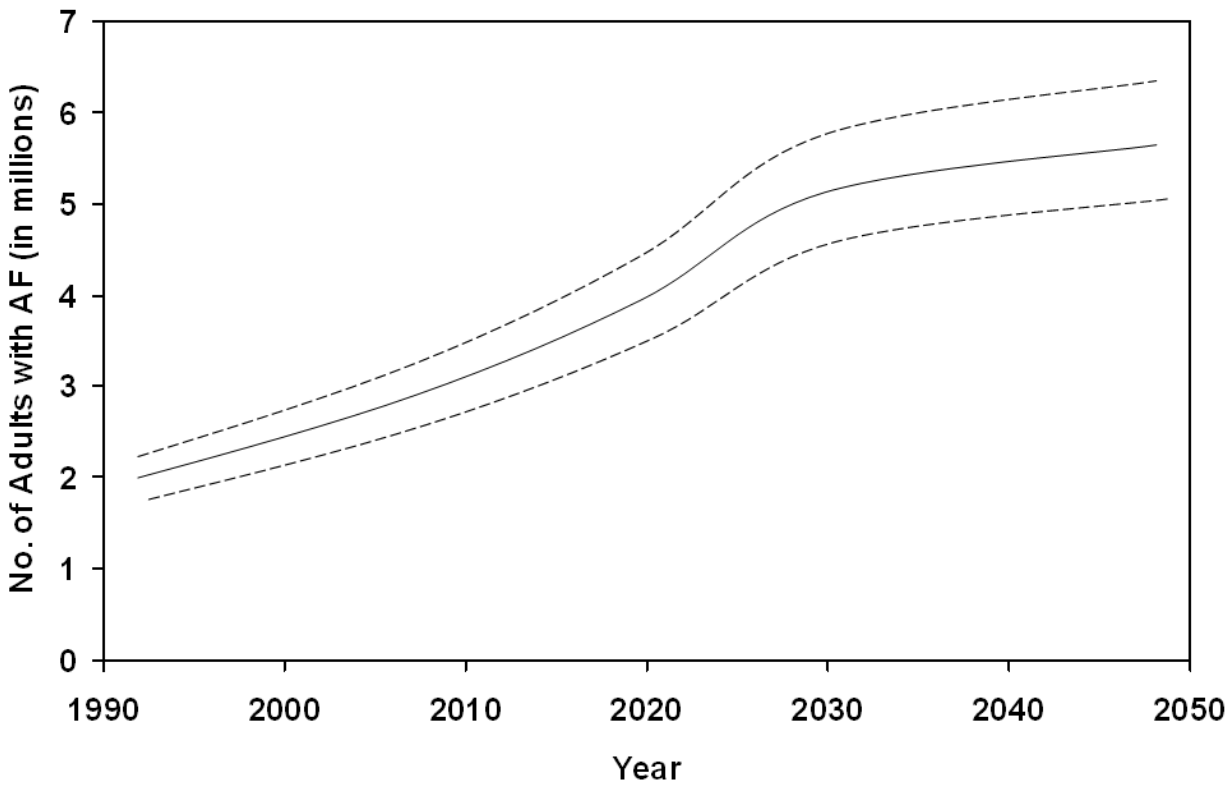
The recognition of the irregular pulse of Atrial Fibrillation (AF) probably dates back several centuries, but the first clear and documented account of the fibrillating atrium was put forward by William Harvey in 1628[2-4], when he also described the circulation of the blood. More than 100 years later, Jean Baptist de Senac made the observation that mitral valve disease was often associated with irregular palpitations.[3] Physiological evidence for AF came from studies using analysis of simultaneously recorded arterial and venous pressures by James McKenzie, who demonstrated absent a-waves during an episode of irregular pulse.[5] It was only the development of the electrocardiogram by Willem Einthoven that provided the first electrocardiographic evidence of AF in 1906. The connection between irregularity of the pulse and clinical AF was subsequently made by Thomas Lewis.[6]

Despite the long history of recognising and treating AF, it is a condition that is still not fully understood. The mechanism of AF continues to be the subject of ongoing research and debate, and its optimal management with both medication and with invasive procedures remain controversial.

1.2 Prevalence, Incidence and Impact on healthcare resources

In modern day clinical practice, AF is acknowledged as the commonest, sustained clinical arrhythmia that is encountered.[7, 8] It occurs in 1-2% of the general population, and its prevalence is expected to double in the next 50 years with an aging population.[9] Previous projections based on a study of adults with a diagnosis of AF at a large health maintenance organization in California, indicated that around 5.6 million adults in the United States will have a diagnosis of AF by the year 2050.[10] (Figure 1.1)

Figure 1.1: Projected number of adults with AF in the United States



Upper and lower curves represent the upper and lower scenarios based on sensitivity analyses. (Adapted from Go et al.[10])

A more recent population study of residents in Minnesota, United States has suggested a trend of increasing incidence of AF (from 3.04 to 3.68 per 1000 person-years) over a 20 year period, leading to an even greater projected prevalence of AF of around 12 million by 2050, possibly rising close to 16 million if further increases in incidence is seen.[11] A European study looking at subjects 55 years and older found prevalence of AF to be 5.5%, rising from 0.7% between the age group of 55-59 years to 17.8% in those more than 85 years of age.[12] In the same study, overall incidence was found to be 9.9 per 1000 person-years. The lifetime risk of AF at age 40 years was estimated to be 26% for men and 23% for women according to a study of subjects from the Framingham Heart Study.[13]

AF is often associated with other significant health problems and is one of the commonest causes for hospital admissions. It has been reported in a Danish study that over the last 20 years, there has been a 60% increase in hospital admissions for AF.[14] Figures in the United States of America have suggested an even more dramatic increase of around 2-3 fold seen over a 15 year period.[15] AF therefore poses a very significant burden to health care resources, with an approximate cost of €3000 annually per patient, and total cost approaching €13.5 billion in the European Union.[8, 16]

1.3 Clinical Sequelae and Prognosis

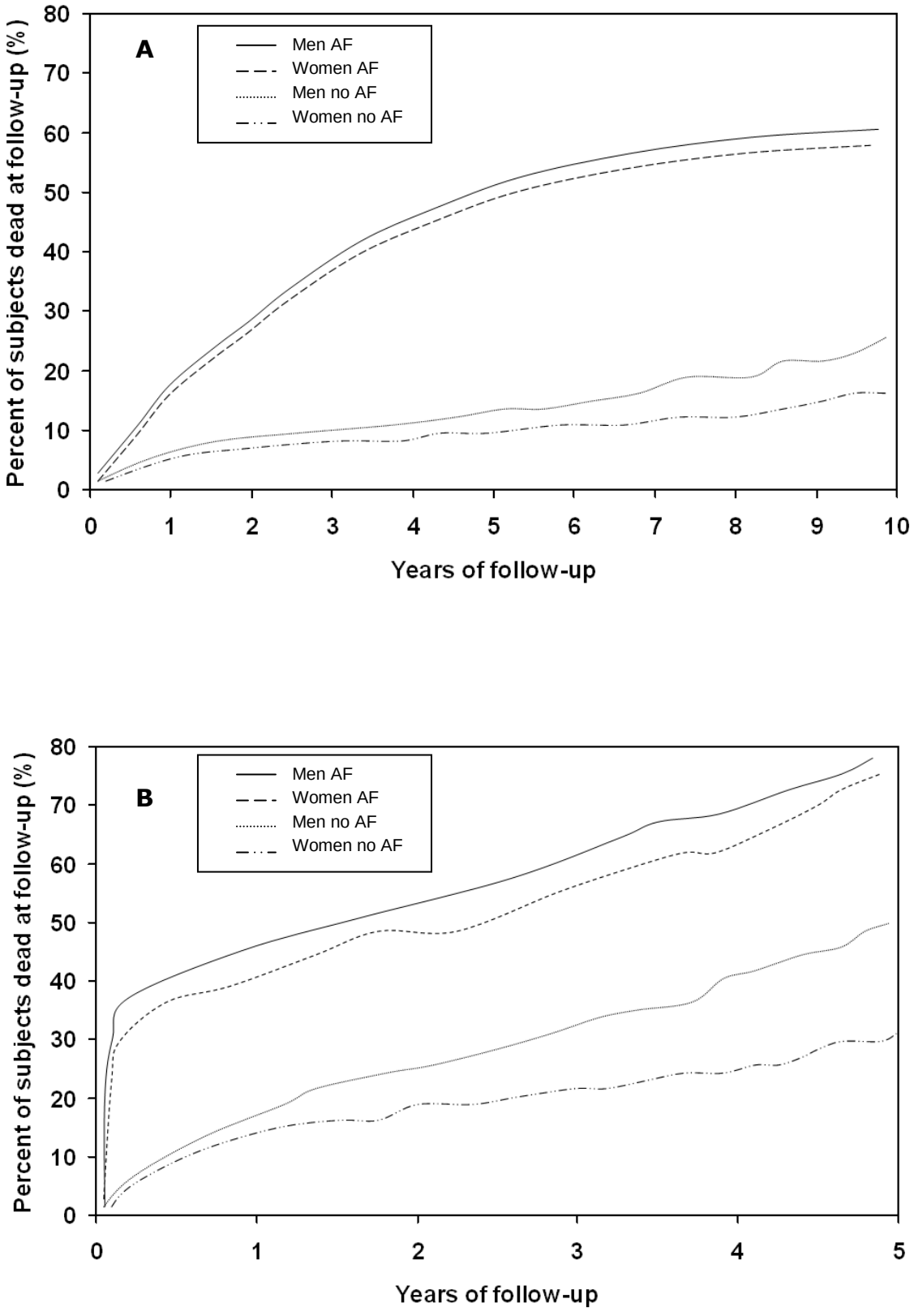
AF is an independent risk factor for stroke,[17-19] with risk going up to almost 5 fold in those over 80 years of age, compared to individuals with no AF.[17] The risk of stroke is also incrementally raised by the co-existence of other conditions such as coronary heart disease, heart failure, hypertension and diabetes. The only effective treatment to reduce the risk of stroke and risk of death from stroke is with anticoagulation

therapy, which is superior to antiplatelet therapy with aspirin but at the cost of increased bleeding risk.[20, 21] Risk stratification scores have been proposed to help quantify stroke risk and guide decision on antithrombotic therapy.[22-24] AF has also been linked with impaired cognitive function and dementia in the absence of any overt stroke.[25-27]

AF results in inefficient atrial contraction and loss of atrial contribution to the cardiac output. If not sufficiently treated, the rapid ventricular rates seen in AF may also give rise to tachycardia induced cardiomyopathy.[28, 29] AF typically gives rise to a variety of symptoms such as palpitations and breathlessness, and is associated with a poorer quality of life compared to healthy controls.[30]

Several studies have also reported an increase in all-cause mortality in patients with AF.[31-34] One of these was from the Framingham Heart Study, which found a 1.5 to 1.9-fold mortality risk in AF patients compared to those in sinus rhythm, after adjusting for pre-existing cardiovascular conditions.[32] In this study, the increase in mortality with AF was seen in both men and women, with AF seemingly diminishing the inherent female survival advantage. (Figure 1.2)

Figure 1.2: Kaplan-Meier mortality curves for subjects aged 55-74 (Graph A) and 75-94 years(Graph B)



Adapted from Benjamin et al.[32]

Data from heart failure trials have also shown AF to be an independent risk factor for mortality and morbidity.[35, 36] The development of AF and congestive heart failure in any temporal order is associated with a poorer prognosis than if either condition existed alone.[37]

1.4 Causal Factors and Associated Conditions

Although AF can occur on its own as an isolated entity (so-called “lone AF”), it is more frequently seen in certain subsets of at-risk individuals and in the setting of other medical conditions. A European survey found that 90% of patients with AF also have other concomitant diseases.[38]

Common causes and associated factors are detailed below:

- **Age**

  Population studies have consistently shown age to be associated with increasing risk of AF.[10, 12, 13, 31]

- **Gender, Race and Genetics**

  Male sex, race: Men are well known to be more susceptible to developing AF.[7, 12, 13, 39] In a survey of over 600,000 men, AF was found to be more prevalent in whites, native Americans, and Pacific Islanders. [40] Data from the Framingham Heart Study also found an increased risk of developing AF in individuals born to parents with a history of AF.[41] Brugada et al had earlier identified a familial form of AF which was inherited in an autosomal dominant fashion, with the disease locus being mapped to chromosome 10.[42] A similar inherited form of AF was also observed in a family by Ellinor et al, who mapped the disease locus in this case to

chromosome 6.[43] This has led to the notion that AF may be an ion channel disorder and further characterization of causative genes is the subject of ongoing research.

- **Hypertension**

Hypertension has long been known to co-exist with AF and is associated with left ventricular hypertrophy, left atrial enlargement and changes in atrial electrophysiology (slowing of conduction velocity[44] and decrease in atrial refractory period[45]), which result in conditions that favour development of AF. [46, 47]

- **Cardiac Disease and Heart Failure**

The presence of congestive heart failure also increases the risk for the development of AF,[39] likely to be due to a combination of increase atrial pressure and associated valvular disease, which on its own is also a risk factor for AF. Interstitial fibrosis affecting electrical conduction in heart failure has been implicated as well.[48] Previous myocardial infarction is also thought to increase the risk of developing of AF in men.[39] Other cardiac conditions associated with AF include congenital heart defects and the cardiomyopathies.

- **Alcohol**

The link between alcohol intake and AF has been demonstrated in a study of over 16,000 subjects which showed that heavy alcohol consumption (35 or more drinks per week) was associated with an increased risk of AF [49]

- **Obesity**

A raised Body Mass Index of >30 has been found to be associated with a 50% increased risk for developing AF, possibly mediated by left atrial dilatation.[50] A closely related finding is that the magnitude of nocturnal oxygen desaturation in obstructive sleep apnoea has been found to be a predictor of AF, independent of body mass index.[51]

- **Pulmonary Disease**

Reduced lung function, such as that seen in patients with chronic obstructive pulmonary disease, is associated with a higher risk of developing AF, with a higher risk of AF hospitalisation than those with normal lung function.[52]

- **Endocrine Disease**

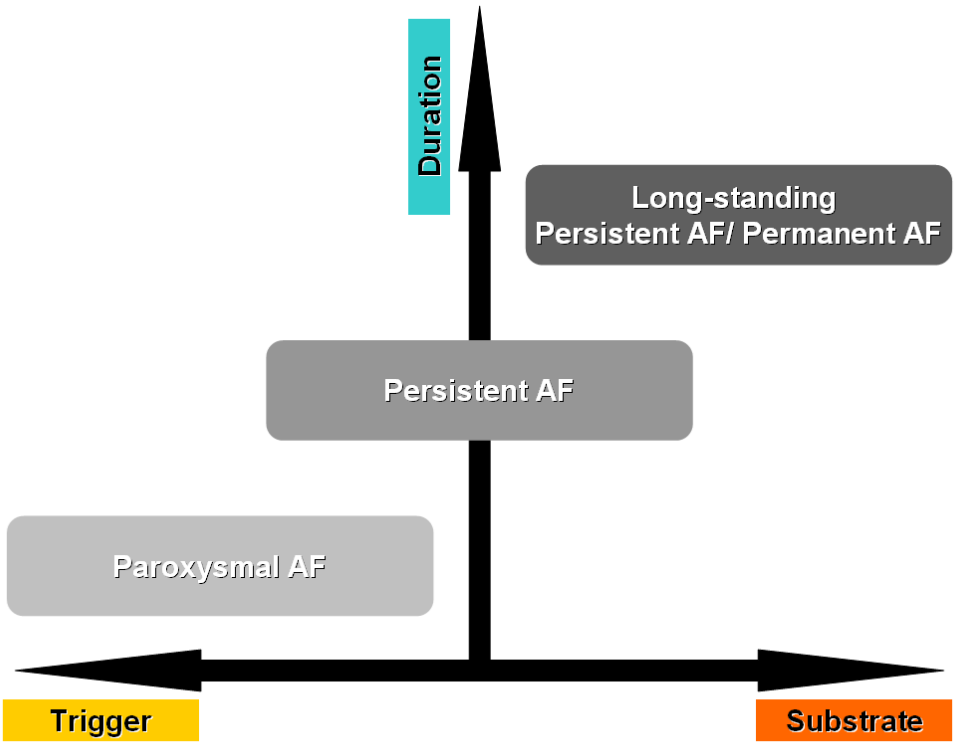
Thyrotoxicosis is a well known cause of AF. Subclinical hyperthyroidism has also been associated with the development of AF.[53] Diabetes often co-exists with AF, and recent data suggests that not only is diabetes a risk factor for AF, the risk increases with longer duration of treated diabetes and worse glycaemic control.[54]

1.5 Classification of AF

According to consensus statements and guidelines from international bodies,[8, 9, 55] AF may be classified as (Figure 1.3):

- ***Paroxysmal AF:*** recurrent AF (≥2 episodes) that terminates spontaneously within 7 days
- ***Persistent AF:*** AF that is sustained for more than 7 days or lasting less than 7 days but requiring pharmacological or electrical cardioversion
- ***Long-standing persistent AF:*** Continuous AF of greater than 1 year in duration
- ***Permanent AF:*** AF where any form of cardioversion has either failed or not been attempted, and where a decision has been taken to accept the presence of AF and not pursue any rhythm control strategy.

Figure 1.3: Classification of AF



Diagramatic representation of the spectrum of AF and relationship with duration, triggers (e.g. ectpoic foci) and atrial substrate (result of electrical and structural remodelling). These will be discussed in greater detail in section 1.6 and 1.7

1.6 Atrial Fibrillation: The self-perpetuating arrhythmia

1.6.1 Electrical remodelling in AF

AF has the inherent ability to perpetuate itself. This has been elegantly demonstrated in a well known study by Wiffels et al[56], who provided experimental evidence for the phrase "AF begets AF". In their study, chronically instrumented goats were maintained in AF with the use of a fibrillation pacemaker. After the first 24 hours of pacing induced AF, a shortening of fibrillation intervals and shortening of the atrial effective refractory period were noted. The ease of AF inducibility increased and an attenuation in physiological rate adaptation was also seen. These electrophysiological changes were completely reversed back to baseline after restoration of sinus rhythm for 1 week. Similar acute reductions in atrial refractoriness have also been demonstrated in human subjects after pacing induced AF.[57]

Yue et al subsequently showed that sustained atrial tachycardia in paced dogs reduced the densities of the transient outward current (I_{to}) and L-type Ca^{2+} current (I_{ca}), and that the latter effect was responsible for decreasing the action potential duration (APD) and APD adaptation to rate.[58] This could therefore be the ionic basis for Wiffels' observation above. A reduction in the Na^{+} current (I_{Na}), conduction velocity and wavelength, as well as increased regional heterogeneity have also been reported in separate publications by the same research group using paced dog hearts.[59, 60] Yue et al went on further to show that prolonged atrial pacing in dogs led to reductions in mRNA concentrations of genes encoding for alpha 1c subunit of L-type Ca^{2+} channels, alpha subunit of Na^{+} channels, and Kv4.3 (potassium channel responsible for I_{to}).[61] These observations mirrored the changes in the corresponding ionic current densities in their earlier studies, thus providing evidence that chronic

atrial tachycardia changes atrial ion channel gene expression, which result in conditions that favour the perpetuation of AF. Similar changes in ionic currents have also been subsequently reported in human AF.[62, 63]

1.6.2 Structural remodelling in AF

Apart from electrophysiological changes, structural remodelling also occurs in AF. The relationship between atrial tissue mass and AF has been recognised as early as 1914.[64] Left atrial dilatation often co-exists with AF and is known to be an independent risk factor for development of AF.[65] On the other hand, left atrial enlargement may also be a result of AF. This may be due to increase in atrial pressure and stretch as a result of loss of atrial contribution to ventricular filling. In patients with newly diagnosed AF, left atrial dimensions (on follow-up of up to 4 years) were found to be larger in those who stayed in AF, compared to those who had paroxysmal AF or no AF recurrence.[66] Follow-up of patients with lone AF showed a significant increase in left and right atrial volume over a 20 month period, having been within normal limits at baseline.[67] According to the multiple wavelet hypothesis proposed by Moe and Abildskov[68], AF is the result of multiple wavelets of re-entrant circuits propagating throughout the atria. The greater the number of wavelets, the more likely the arrhythmia will sustain itself. It can therefore be implied that a larger atrial mass will promote the existence of AF. At a microscopic level, changes such as loss of myofibrils, accumulation of glycogen, changes in mitochondrial structure, fragmentation of sarcoplasmic reticulum and dispersion of nuclear chromatin have been observed in goats with pacing induced AF.[69] These changes have been likened to that seen in myocardial hibernation in the ventricular myocyte. AF is also associated with fibrosis[70, 71] of the atrium and remodelling of connexins (myocyte gap junction proteins) may also play a role, although studies into

this so far have not produced consistent results.[72, 73] Together with macroscopic atrial enlargement, these ultrastructural changes may give rise to increased heterogeneity in atrial conduction and promote maintenance of AF.

From the discussion above, it can be seen that both electrical and structural remodelling in AF constitute a positive feedback loop that will only lead to one possible outcome: the continued persistence of AF.

1.7 Mechanism of AF – focal vs reentry, have we come full circle?

The mechanism of AF has been the subject of keen research ever since it was discovered. In the last 100 years, several theories of AF mechanism have been put forward by researchers, but up to this day, it remains an area of controversy.

One of the main concepts behind the mechanism of AF and indeed that of most cardiac arrhythmia, is reentry. The observation of reentry was first described by Mayer in 1908[74], when he described circus movement in rings cut from the jellyfish umbrella. He wrote:

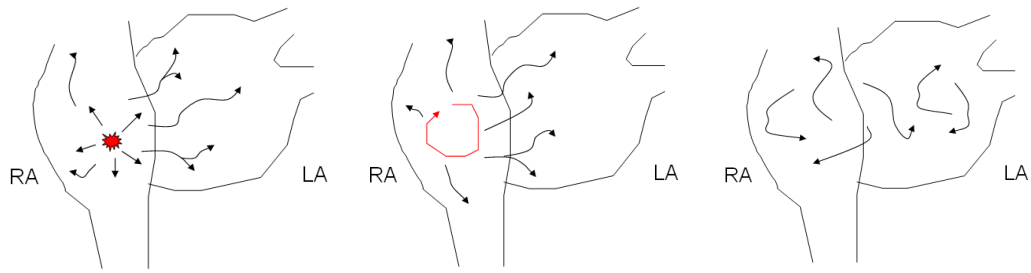
> *"This wave will maintain itself indefinitely, provided the circuit be long enough to permit each and every point of the wave to remain at rest for a certain period of time before the return of the wave through the circuit"*

Further studies of animal hearts by Mines[75, 76] established the importance of unidirectional block as an essential requirement for reentry. He also provided the

beginnings of the concept of wavelength (which is the minimum pathlength that supports reentry) and its relationship with the refractory period and conduction velocity. Studies by Garrey[64] further associated fibrillation with multiple reentry, and also demonstrated the critical mass theory when he showed that AF could not be induced or sustained below a critical atrial mass.

The view that AF may be due to reentry mechanisms was not universally accepted and other researchers suggested that rapid firing of single or multiple atrial ectopic foci are responsible for AF.[77, 78] Lewis initially supported the idea of AF being from a focal ectopic origin[79] but subsequently changed his view to that of single reentry circuit with fibrillatory conduction,[80] as opposed to multiple reentrant circuits originally suggested by Mines and Garrey.(Figure 1.4)

Figure 1.4: Early theories of AF mechanism.



Proposed mechanisms included ectopic focus (left), single reentry with fibrillatory conduction (middle), and multiple reentry (right)

Building on the foundations left by Mines in his original study of reentry, Wiener and Rosenblueth[81] subsequently formulated wavelength (WL) as a product of refractory period (RP) and conduction velocity (CV) [WL = RP X CV], and showed that it critically determined the likelihood of reentry.

The theory of reentry behind AF was once again challenged by Scherf [82, 83] in 1947, when he conducted experiments which showed that injection of aconitine into the atrial wall (either left or right atrial appendage) produced what he described as “auricular tachycardia”, which was subsequently shown to be atrial flutter with episodes of fibrillation as well. Cooling at the site of injection immediately abolished the tachycardia with rapid re-initiation when cooling was stopped. This led to him concluding that atrial flutter and fibrillation are initiated by rapid impulses originating from a focal source and that reentry or circus movement cannot explain his observations.

1.7.1 The Multiple Wavelet Hypothesis

Further experiments carried out by Moe and Abildskov[68] showed that AF induced by either aconitine injection or repeated electrical stimulation of the appendage, was capable of sustaining itself, in the presence of vagal nerve stimulation, even after clamping off the atrial appendage. This led them to conclude that while it is possible that AF may be initiated by focal firing from a single or multiple sites, or by a rapid circulating circus movement (the type that is attributed to Lewis), “true fibrillation” is subsequently self-sustaining and independent of the original trigger. The multiple wavelet hypothesis[68, 84] was put forward to explain this, whereby it was thought that propagation of electrical excitation occurred in a non-uniform manner due to variation in tissue refractory periods, such that depolarisation wave fronts divide into daughter wavelets as they encounter islets of refractory tissue, which then continue to wander randomly around the atria, hence perpetuating AF. Based on this, a larger atrial mass will accommodate a greater number of wavelets and increase the chance of sustaining AF which would appear to match the original theory by Garrey[64] closely. The multiple wavelet hypothesis was subsequently tested in a computer model of AF

and found to agree well.[85] An experimental model of AF by Allessie et al[86] demonstrated that the critical number of wavelets required for the perpetuation of AF ranges between 3-6.

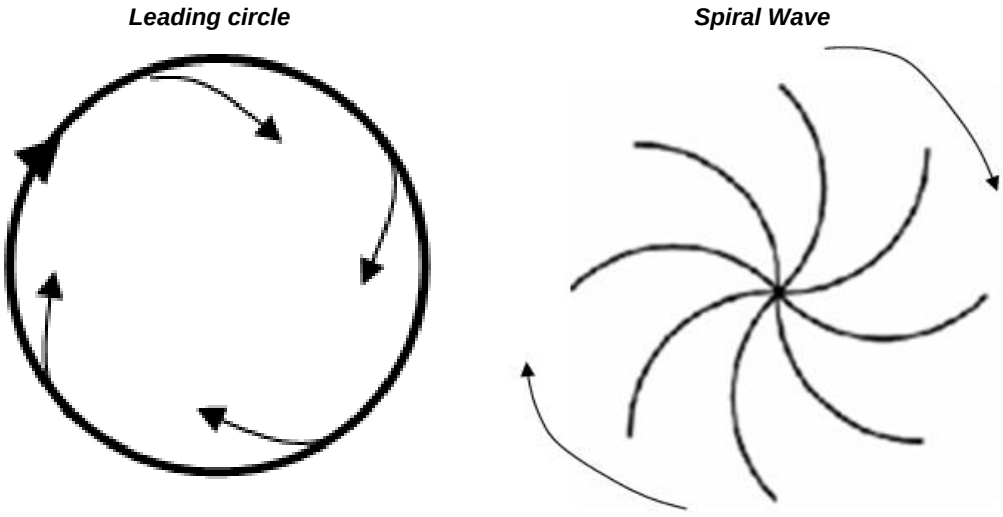
1.7.2 The Leading Circle

The description of reentry by Mines[75] was based on observations in ring-shaped strips of cardiac tissue which assumes the presence of an anatomical obstacle. Further work by Allessie et al[87] led to the proposal of the “Leading Circle” concept. The authors showed that it was possible to induce functional reentry without an anatomical obstacle. A functional reentrant circuit in this case was completely dependent on the electrophysiological properties of the tissue composing the circuit, and naturally established itself as a circuit the size of its wavelength, i.e. the smallest circuit possible, with no excitable gap. This “leading circle” will have the most rapid activation rate compared to its peripheries and hence will control overall rate. The centre of the core in this case is activated by centripetal wavelets form the leading circle, keeping it refractory (Figure 1.5). The number of leading circles a tissue can contain is dependent on its dimensions relative to that of the circuits which in turn is predominantly determined by its wavelength (product of conduction velocity and refractory period). In the context of AF, a larger wavelength will mean a decrease in the capacity to accommodate multiple circuits, which will mean increased likelihood of termination. The significance of this has made wavelength a potentially useful tool to evaluate the efficacy of drugs used in the treatment of AF.[88-90]

1.7.3 Spiral Waves

Subsequent work by Panfilov[91] and Winfree[92] proposed spiral wave activity as the mechanism by which self-sustaining rhythms are maintained in excitable media such as the heart, hence challenging the leading circle concept as the cause of functional reentry. Spiral waves, also called rotors or vortices, rotate around a core where isochrones converge and conduction velocity is slow (Figure 1.5). The size of the core is determined by the curvature of the wave front and by the refractory period. The core of a spiral wave is excitable but non-excited, and there remains an excitable gap in front of the leading edge of the wave, unlike that of the leading circle. Polymorphic and monomorphic patterns of activation can arise due to drifting spirals and anchored spirals respectively, suggesting that the behaviour of the core or rotor may play a role in determining the appearance of the arrhythmia.[93, 94]

Figure 1.5: Leading Circle reentry vs Spiral wave reentry.



The leading circle has a core that is continuously excited by centripetal waves and has no excitable gap, while the spiral wave has an excitable core that is not excited. An excitable gap is present unlike the leading circle.

Three-dimensional spiral waves would be expected to occur across the thickness of the myocardium and these are known as scroll waves. The early studies looking into spiral waves were based on models of monomorphic and polymorphic ventricular tachycardia. Subsequent experimental evidence has also been presented showing presence of spiral waves in AF. Using optical mapping of sheep atria, Skanes et al[95] demonstrated evidence of spatiotemporal periodicity during AF, with good correlation between the dominant frequency of the periodic activity and that of the atrium globally. The sources of spatiotemporal periodicity were predominantly located in the left atrium, and were shown be stationary rotors on the anterior wall of the left atrial appendage in their model of AF. Conversely, there was usually no periodic activity in the right atrium and complex patterns of activation were seen, giving rise to irregular activity on the electrograms. An extension of this work by Mandapati et al[96] which included more extensive contact mapping of the atria, demonstrated the characterization of spatial distribution of dominant frequencies (DF) which allowed identification of sites with rapid periodic activity. The sites with the highest DF were typically located close to the pulmonary veins and posterior left atrium. Optical mapping in the same study revealed the presence of stable rotor activity at highest DF sites, suggesting that stable, localized reentry in the form of a rotor could be the mechanism of AF that was seen. These findings would agree well with an earlier report by Schuessler et al[97], where it was found that multiple reentrant circuits seen in early onset of induced AF, stabilized to a small, single and stable reentrant circuit when AF became sustained, suggesting that AF could be more “focal” than previously thought. From the above studies, Jalife et al went on to propose that sustained AF is due to periodic activity from re-entrant sites in the form of rotors, and that wavefronts emanating from these could divide into wavelets when encountering anatomical or functional obstacles, and these can go on to generate new rotors.[98, 99]

This hypothesis would seem to share some common ground with Lewis' view[80] that AF was due to circus movement consisting of a single re-entrant circuit and fibrillatory conduction.

While the rotor hypothesis has gained increasing acceptance over the recent years, this does not exclude the existence of the other proposed mechanisms. For example, the revolutionary use of catheter ablation to treat AF has shed further light on the role of focal activity in AF. By mapping intra-cardiac electrograms in patients with paroxysmal AF, Jais et al[100] found rapid focal activity predominantly from the left atrium, which was responsible for producing a surface ECG pattern of AF. Haissaguerre et al[101] subsequently showed in a larger series of patients, that ectopic activity responsible for triggering AF were mainly located in the pulmonary veins, and that catheter ablation of these sites was able to eliminate AF on medium term follow-up.

In summary, the mechanism of AF is still not well understood despite extensive research. The theory of AF has rotated between all the 3 main described mechanisms over the last 100 years with no clear final consensus. It is possible that there are different forms of AF, and that all the described mechanisms may be involved at different stages of the arrhythmia history. In the presence of continued electrical and mechanical remodelling, a final common equilibrium may eventually be reached over time irrespective of the initial trigger or mechanism.

1.8 Autonomic nervous system and AF

The influence of the autonomic system on the heart has been known for a long time. In 1921, Loewi carried out experiments on frog hearts to show that electrical

stimulation of the vagus nerve resulted in the production of a chemical substance that was capable of slowing the heart rate. This substance which he called “Vagusstoff”, was later shown to be acetylcholine. This eventually led to him being awarded the Nobel Prize in Physiology or Medicine. We now know that acetylchoine is the main neurotransmitter of the parasympathetic nervous system, while epinephrine and norepinephrine are neurotransmitters of the sympathetic nervous system.

In the context of clinical arrhythmia, it was Coumel et al who first described the observation of paroxysmal AF secondary to vagal over-activity.[102] This subsequently led to the description of vagal and sympathetic forms of AF; the former being prevalent in normal hearts, and the latter occurring in the presence of heart disease.[103]

Anatomical studies have demonstrated the presence of clusters of autonomic tissue i.e ganglionated plexi (GP) located at specific sites around the human heart, which include locations near the pulmonary veins, predominantly on the posterior surfaces of the atria.[104, 105] Although a bradycardic response is often described when stimulating GP, they have been shown to contain both sympathetic and parasympathetic elements,[106] which are thought to have a synergistic effect in initiating AF.[107, 108] A recent study by Arora et al, found that parasympathetic fibres and muscarinic receptors were preferentially located in the posterior left atrium and that cholinergic blockade with topical tropicamide in this region attenuated ERP shortening during vagal stimulation, and almost completely eliminated vagally-induced AF, thereby highlighting the potential importance of this region.[109]

Stimulation of the vagus nerve has been known to increase the atrial rate in atrial flutter in the early days of AF research by Lewis et al.[110] In 1972, Armour et al[111] showed it was possible to induce both atrial and ventricular tachycardias by local stimulation of cardiac nerves. Vagal stimulation or exogenous acetylcholine produces a negative chronotropic effect on the SA node and a negative dromotropic effect on the AV node.[112, 113] At the atrial level, vagal stimulation reduces atrial effective refractory period[114, 115] and also increases spatial variability in refractoriness,[116] thereby making conditions favourable for starting and sustaining AF. Sympathetic stimulation also results in reduction in atrial effective refractory periods[114, 117], and is thought to play a role in AF as well.[107, 118] Muscle sleeves of thoracic veins can also develop automaticity and triggered activity with sympathetic stimulation, which could in turn trigger AF.[119]

An early ablation study on dogs in 1995,[120] found that catheter radiofrequency ablation of the atrium based on the maze procedure[121] markedly attenuated the shortening of the atrial effective refractory period with vagal stimulation, and also made AF non-inducible by rapid pacing. Schauerte et al[118] described application of high frequency stimulation (during the atrial refractory period) at pulmonary vein and superior vena cava sites to produce local parasympathetic stimulation. This resulted in triggering of rapid atrial ectopic beats leading to induction of AF. These effects were blunted by beta-blockade and abolished by atropine, implying involvement of both sympathetic and parasympathetic pathways. An earlier study by the same research group showed that radiofrequency ablation of local parasympathetic tissue successfully abolished vagally mediated AF in dogs.[115] Adaptation of ablation techniques to target autonomic tissue in human subjects with AF have also shown encouraging results.[122, 123] This will be discussed in greater detail in Chapter 5.

There is increasing evidence that atrial ectopy responsible for induction of AF under autonomic influence, is possibly due to triggered firing from early afterdepolarizations.[108, 124, 125] Parasympathetic stimulation shortens action potential duration, producing earlier repolarization, and sympathethic stimulation causes a longer and larger intracellular calcium release. This drives Na^+/Ca^{2+} exchange across the cell membrane (ratio of 3 Na^+ ions in and 1 Ca^{2+} ion out), resulting in a net inward current, hence producing early afterdepolarizations and triggered firing.[125]

1.9 Medical Therapy for AF

There are 3 treatment aims in AF which include 1) rate control of AF, 2) rhythm control by maintaining sinus rhythm as much as possible, and 3) reducing the risk of thromboembolism. How aggressively each aim is pursued is dependent on severity of symptoms, presence of haemodynamic compromise or heart failure, and other associated comorbidities.

1.9.1 Antithrombotic therapy

In general, the risk of stroke and thromboembolism is increased by 4-5 fold in AF, irrespective of whether AF is paroxysmal or persistent.[126] This risk will vary depending on the existence of other associated risk factors and risk scoring systems have been developed to help guide antithrombotic therapy.[22-24]

Meta-analyses[127, 128] have demonstrated that warfarin significantly reduced risk for stroke, systemic thromboembolism, and also all-cause mortality compared to placebo. Compared to aspirin, warfarin is more efficacious at reducing stroke and thromboembolism risk.

1.9.2 Rate vs Rhythm Control

In the absence of spontaneous resolution of AF, sinus rhythm can be successfully restored by using anti-arrhythmic medication or DC cardioversion. Long term maintenance of sinus rhythm is, however, difficult to achieve and AF recurrence rates can exceed 50% in the first month post DC cardioversion.[129] Concurrent treatment with class III anti-arrhythmic agents such as amiodarone improves the efficacy of DC cardioversion.[130]

Several studies have also compared treatment using a rate control approach with rhythm control. The AFFIRM trial (AF Follow-up Investigation of Rhythm Management)[131] found no significant differences in stroke or mortality between rate and rhythm control strategies. More patients were hospitalized, and there were also more drug-related adverse effects in the rhythm control group. In fact, a non-significant trend for increased mortality was seen in the rhythm control group. Further analysis of the data suggested that the presence of sinus rhythm was actually associated with a lower risk of death, and that any benefit of anti-arrhythmic drugs were offset by their association with increased mortality. [132] The smaller RACE (Rate Control vs Electrical Cardioversion) study also found rate control to be not inferior to rhythm control strategy for composite endpoint of death and other associated morbidity.[133] Similarly, the recent AF-CHF (The AF and Congestive Heart Failure) trial found no difference in mortality in patients with history of heart failure and AF when randomized to rate or rhythm control.[134] The optimal intensity of rate control has not been clearly defined, although recent evidence suggests that a lenient rate-control strategy (resting heart rate <110 bpm) is as effective as strict rate control (resting heart rate <80 bpm, during moderate exercise <110 bpm).[135]

In terms of quality of life assessment, follow-up analysis of the AFFRIM cohort[136] as well as the RACE cohort[137] of patients found no overall improvements in quality of life measures between the 2 treatment strategies, although maintenance of sinus rhythm in the RACE study (rather than assigned treatment strategy) was associated with improvement in quality of life. The PIAF[138] (Pharmacologic Intervention in AF) and STAF[139] (Strategies of Treatment of AF) studies also found no difference between rhythm and rate control. However, analysis of patients maintaining sinus rhythm on anti-arrhythmic medication in 2 other randomized double-blinded studies did show improvement in quality of life measures and exercise capability.[140, 141]

1.9.3 Upstream Therapy

As mentioned previously, AF is associated with structural changes in the atrium which includes interstitial fibrosis.[48, 142] This could be a contributing cause or a consequence of AF. Atrial stretch and enlargement in AF can activate the renin-angiotensin-aldosterone system[143] which is known to be involved in myocardial fibrosis seen in hypertensive heart disease, cardiomyopathy and myocardial infarction.[144] Blockade of this pathway could in theory, inhibit conditions favouring the development of AF. Indeed, large scale trials assessing the use of angiotensin II receptor blockers have found a reduced incidence of AF in patients with left ventricular hypertrophy[145] and symptomatic heart failure[146] when treated with this class of drugs. The anti-inflammatory effect of statins and the influence of fish oil on vagal activity and ion channels are also thought to have beneficial effects on atrial remodelling in the long term, and may play a role in reducing the likelihood of developing AF. However, clear evidence for the use of these agents is lacking at the moment, and they remain the subject of ongoing research.

1.10 Ablation therapy for AF

1.10.1 Background

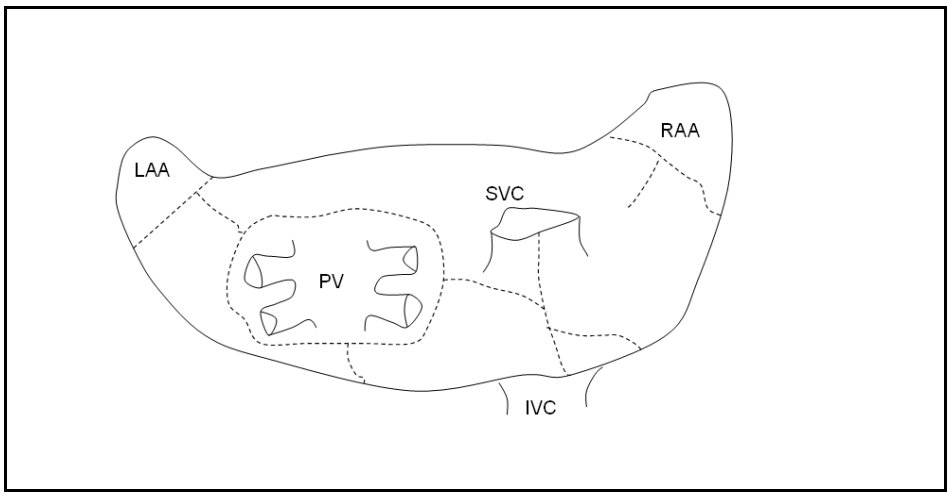
The use of anti-arrhythmic medication is limited by pro-arrhythmic risks and extra-cardiac side effects, most notably seen in Amiodarone and some class I agents. Findings from some of the studies discussed earlier,[132, 140, 141] suggest some potential benefit of maintaining sinus rhythm, which may be offset by harmful effects of anti-arrhythmic drugs. This could therefore imply that non-pharmacolgical approaches to treating AF may yield more satisfactory results, if associated risks can be minimized.

1.10.2 Surgical AF ablation

Surgical ablation of AF was first attempted on human subjects by Cox et al and was reported in 1991.[121] The technique essentially involved cut-and-sew incisions in both atria, thereby compartmentalizing the atria and reducing the contiguous atrial mass available to participate in fibrillation. (Figure 1.6) This was also known as the Maze procedure and early versions of the procedure were associated with impaired heart rate response to exercise and loss of atrial transport, which led to further revisions of the technique. In 123 patients (around 50% with chronic AF) undergoing a mixture of Maze I, II and III procedures, 66% were in sinus rhythm, 33% were atrial paced and 1% were in AF on follow-up of at least 3 months post-procedure.[147] Although there was a reducing trend of requirement for permanent pacing with each revision of the procedure, 25% of patients who had the Maze III procedure, still needed implantation of a permanent pacemaker. Subsequent success rates improved to 98% free of AF after almost 6 years follow-up.[148] Due to the complexity of the procedure and need for cardiopulmonary bypass, the Maze procedure is infrequently carried out alone and tended to be performed only in conjunction with other reasons for heart surgery.

A meta-analysis of the Maze procedure in addition to mitral valve surgery found evidence (although weak) indicating that the Maze procedure, compared to control, led to a reduction in stroke risk, increased likelihood of maintaining sinus rhythm, increased need for permanent pacing, and increased risk of post-operative bleeding unless radiofrequency energy was used to create the lesions.[149]

Figure 1.6: The Cox-Maze III procedure



Cut-and-sew lesions as indicated by the dashed lines. View from posterior aspect of atria. (LAA: left atrial appendage, PV: pulmonary veins, RAA: right atrial appendage, SVC: superior vena cava, IVC: inferior vena cava

1.10.3 Catheter ablation of AF

Catheter ablation for AF by radiofrequency energy was subsequently developed to offer a less invasive option to the treatment of AF. This was initially tested in animals and case reports of catheter ablation of human AF soon followed.[150] Early reports had focused on right atrial linear ablation, but it was later found that adding left atrial ablation to right atrial ablation provided further benefit in patients with paroxysmal AF.[151] A further study of patients with frequent paroxysmal AF found that the surface ECG pattern of AF could be caused by focal firing within the atria and that these could be eliminated successfully by ablation.[100] This eventually led to the landmark study by Haissaguerre et al which was published in 1998, where it was found that

over 90% of atrial ectopy responsible for triggering paroxsymal AF were located in the pulmonary veins, and that radiofrequency ablation of these foci resulted in freedom from AF off anti-arrhythmic drugs in around 60% of patients after 8 months of follow-up.[101] A further study by Chen et al[152] a year later found that over 80% of patients were free of AF (off medication) 6 months after ablation of pulmonary vein ectopic foci, although subsequent transoesophageal echocardiography revealed significant pulmomary vein stenosis in 42% of ablated pulmonary veins. Although pulmonary veins are the most commonly described sites for ectopy-induced AF, it is also known that ectopy could originate in the left atrial posterior wall, superior vena cava, crista terminalis, ligament of Marshall, coronary sinus ostium and atrial septum.[153]

The discovery of focal sites triggering AF which were amenable to catheter ablation led to a new era in the treatment of AF. Similarly encouraging results were also reproduced by other ablation centres and the procedure itself continued to evolve with increasing operator experience. The original procedure put forward by Haissaguerre et al involved segmental ostial isolation of ectopic foci near the pulmonary vein ostia or at sites electrically connecting the left atrium to the pulmonary vein in the absence of ectopy.[154, 155] Pappone et al described the use of 3D electroanatomical mapping of the left atrium to create contiguous linear lesions in both atria and also circumferential encirclement of each pulmonary vein ostium, with freedom of AF after 9 months follow-up documented in up to 85% of patients (62% drug-free).[156, 157] In a larger series of patients (n=251), Pappone et al showed that 85% and 68% of patients with paroxysmal and permanent AF respectively were free of AF 10 months after ablation using his described technique above of circumferential ablation of the pulmonary veins.[158]

A subsequent comparison between the 2 techniques in patients with paroxysmal AF found circumferential pulmonary vein ablation using 3D electroanatomical mapping to be superior than segmental ostial isolation in terms of freedom of AF.[159] In this study, the authors made some modifications to circumferential pulmonary vein ablation by encircling both ipsilateral veins together and adding linear ablation to the posterior atrium and mitral valve isthmus to reduce the risk of developing atrial flutter. Another study examining this showed better outcomes with segmental ostial isolation however, although in this study, the patients in the circumferential ablation group did not have posterior atrial linear ablation, and atypical atrial flutter was observed significantly more frequently in this group.[160] Although not originally a specified end-point, the importance of ensuring complete isolation of pulmonary veins using the circumferential vein ablation approach was also highlighted by Ouyang et al.[161]

1.10.4 Atrial tachycardia and flutter

One of the potential problems with ablating the pulmonary veins and left atrium is the development of iatrogenic atrial tachycardia or flutter, which occurs acutely in around 10% of patients.[162] The mechanism of atrial tachycardias is thought to be mainly due to focal micro-reentry at ostia of pulmonary veins, although non-reentrant mechanisms may also be involved.[163] Macro-reentrant circuits can result from creation of isthmuses and conduction gaps during ablation. The additional creation of linear ablation lesions to achieve conduction block across the posterior, anterior and roof of left atrium as well as mitral valve isthmus, have been described and shown to reduce the risk of developing iatrogenic atrial arrhythmia post-ablation.[164-166] The addition of linear ablation to segmental ostial pulmonary vein isolation was also shown to improve chances of maintaining sinus rhythm in patients with persistent

AF.[167] Concurrent cavotricuspid isthmus ablation in patients with both AF and typical atrial flutter may also further improve outcomes.[168]

1.10.5 Trigger vs Substrate ablation

Apart from ablating and isolating triggers of AF such as pulmonary or non-pulmonary ectopic foci, modification of the substrate of AF maintenance is also thought to be important, particularly in persistent or long-standing forms of AF.

1.10.5.1 Linear ablation

This can be achieved by further compartmentalizing the atrium similar to the surgical Maze procedure, by means of creating linear lesions as discussed above, which also serves to reduce the risk of developing atrial tachycardia or flutter. The endpoint of non-inducibility after pulmonary vein isolation has been suggested to help select patients with paroxysmal AF to have further linear ablation.[169] As mentioned above, additional linear ablation to the roof and mitral valve isthmus improves outcomes compared to pulmonary vein isolation alone, in patients with persistent AF.[167]

1.10.5.2 Complex fractionated electrograms

There has also been interest that sites containing fractionated or fragmented electrograms have a role in persistent AF. Konings et al showed that fragmented AF electrograms are found at pivot points and zones of slow conduction, which could represent sites of wavelet reentry facilitating the maintenance of AF.[170] On this basis, Nademanee et al[171] carried out catheter ablation on 121 patients with paroxysmal and persistent AF, specifically targeting complex fractionated electrograms (CFE) in both atria, without attempting to isolate the pulmonary veins. In his study, CFEs were

defined as atrial electrograms which are fractionated (composed of 2 deflections or more) and/or consisted of continuous baseline deflections with prolonged activation complex. Termination of AF was seen in 95% of patients and 91% of patients were free of arrhythmia and symptoms at 1 year follow-up, with the majority needing only 1 ablation procedure. Using the same technique of ablation in a larger cohort of over 600 patients with paroxysmal, persistent and permanent forms of AF, Nademanee et al[172] showed maintenance of sinus rhythm in about 80% of patients over a mean follow-up period of over 2 years. However, despite this initial optimism, the evaluation of CFE ablation by other authors have not reproduced the same success rates, and there also appears to be conflicting evidence about its value as an adjunct to routine pulmonary vein isolation.[173-177]

1.10.5.3 Dominant Frequency (DF) Analysis

There is evidence from animal studies that AF exhibits characteristics of spatiotemporal periodicity and that the identification of high DF sites could reflect underlying rotor activity which may be important targets for ablation.[95, 96] Sanders et al[178] carried out DF mapping of human AF and found that patients with paroxysmal AF were more likely to have DF sites within the pulmonary veins, while atrial DF sites were more prevalent in those with permanent AF. In addition, it was also noted that ablation at sites with high DF was associated with increased likelihood of terminating AF. A left-to-right atrial frequency gradient is seen in patients with paroxysmal AF[179] and pulmonary vein isolation has been shown to eliminate this gradient[180], highlighting the potential role of the pulmonary veins in maintenance of AF. By using a strategy of mapping and ablating maximal DF sites (in real-time) followed by circumferential pulmonary vein isolation, Atienza et al found that 88% and 56% of paroxysmal AF and persistent AF patients respectively, were free of AF (mean follow-

up of 9 months).[181] In this study, a proportion of the maximal DF sites were not ablated due to technical or safety reasons and on analysis of follow-up results, ablation of maximal DF sites was found to be an independent predictor of freedom from AF.

A reduction in atrial DF measured from coronary sinus electrograms as well as from different sites within the left atrium can be seen after circumferential pulmonary vein or electrogram based ablation.[182] A critical decrease in DF by ≥ 11% has been proposed to be associated with maintenance of sinus rhythm after ablation of persistent AF.[183]

1.10.5.4 Ganglionated Plexi Ablation

The autonomic nervous system has been implicated in the initiation and maintenance of AF as discussed previously. Ganglionated plexi (GP) are autonomic tissue located on the epicardial surface of the heart, typically near pulmonary veins and also posterior surfaces of the atria. GPs are thought to act as integration centres that modulate the autonomic influence from the autonomic nervous system and have been proposed as potential targets for ablation. Po et al[184] carried out GP ablation together with pulmonary vein antral ablation by using high frequency stimulation to first identify locations of GP. Single procedure success rate was found to be around 86% in patients with paroxysmal AF after mean follow-up of 22 months. A purely anatomically based approach of ablating sites expected to contain GP (without the use of high frequency stimulation) has also been described. In a study of patients with paroxysmal AF, this technique resulted in maintenance of sinus rhythm in 71%, with the use of implantable recorder for rhythm monitoring for 12 months.[185] The results however in patients with long-standing persistent AF are less encouraging

(38% single procedure success rate if only GP locations were targeted).[186] A randomized comparison between anatomical GP ablation and high frequency guided ablation of GP in patients with paroxysmal AF found the former to confer better outcomes.[187]

1.10.6 Outcomes following AF ablation

There are numerous non-randomized studies publishing outcome data of catheter ablation for AF from individual centres. In general, single procedure success ranged between 38 to 78% in patients with paroxysmal AF, 22 to 45% in persistent AF, and 16 to 84% with mixed types of AF.[55] Repeat ablation procedures can lead to further improvements in success rates. Interpretation of outcome data from these studies is difficult due to difference in methods of AF recurrence surveillance, duration of follow-up, techniques of ablation used, definition of success, variable use of anti-arrhythmic drugs, difference in patient population and varying degrees of AF ablation experience from different centres. It is therefore not difficult to see how the reported success rates can vary so greatly. What is consistent from these studies however, is that persistent or long-standing persistent forms of AF are less easily treated with catheter ablation.

A worldwide survey of 8745 patients in 2005, found that 52% became asymptomatic off anti-arrhythmic drugs, and 24% became asymptomatic while on previously ineffective anti-arrhythmic drugs after almost 1 year of follow-up after catheter ablation of AF.[188] The overall complication rate was in the region of 6%. An update of the same survey in 2010, which included 16309 patients, found that 70% became asymptomatic off anti-arrhythmic drugs, and 10% became asymptomatic while on

previously ineffective anti-arrhythmic drugs after 18 months of follow-up. Overall success rates were significantly better in paroxysmal AF (83%) than persistent AF (75%) and long-lasting persistent AF (72%). Major complications occurred in 4.5% of cases (Table 1.1).[189] These findings indicate an improvement in outcomes with advancement in ablation technology and operator skill.

Table 1.1: Complication rates from the most recent worldwide survey

Complication	**Rate (%)**
Death	0.15
Tamponade	1.31
Pneumothorax	0.09
Haemothorax	0.02
Sepsis, abscess or endocarditis	0.01
Permanent diaphragmatic paralysis	0.17
Femoral pseudoaneurysm	0.93
Arteriovenous fistula	0.54
Valve damage requiring surgery	0.07
Atrio-oesophageal fistula	0.04
Stroke	0.23
Transient ischaemic attack	0.71
PV stenosis requiring intervention	0.29
Total	4.54

Adapted from Cappato el al.[189]

There are a number of randomized control trials comparing catheter ablation of AF with anti-arrhythmic drug therapy (Table 1.2).[190-196] All have pointed towards ablation being superior to anti-arrhythmic drug therapy in terms of freedom from AF. Additionally, some of these studies have also shown more marked improvements in symptom scores and quality of life measures in the ablation group. Systematic

reviews of the available data suggest that catheter ablation has greater efficacy at reducing AF recurrence and also causes less complications or side effects, compared to anti-arrhythmic drug therapy.[197, 198]

Table 1.2: Comparison of catheter ablation vs AAD outcomes

Authors	Year	No. (Ablation vs AAD)	Type of AF	Ablation strategy	Drugs used for AAD*group	Follow-up (mths)	Outcome (Ablation vs AAD)	Other measures
Krittayaphong et al[190]	2003	15 vs 15	PAF and persistent AF	SOPVI, linear ablation of RA	amiodarone	12	79% vs 40% freedom from AF	Improvement in symptom scores and QOL measures in ablation group only
Wazni et al[191]	2005	33 vs 37	PAF and persistent AF	SOPVI	flecainide, propafenone, sotalol, amiodarone	12	13% vs 63% recurrence of AF	Reduced hospitalization and higher QOL scores in ablation group
Pappone et al[193]	2006	99 vs 99	PAF	CPVA	flecainide, propafenone, sotalol, amiodarone	12	86% vs 22% free from arrhythmia	Fewer hospitalizations in ablation group
Stablile et al[192]	2006	68 vs 69	PAF and Persistent AF	CPVA, MVI, CTI	flecainide, propafenone, sotalol, amiodarone, verapamil, beta blockers, disopyramide	12	44% vs 91% had recurrence	
Oral et al[194]	2006	77 vs 69	Long-standing persistent AF	CPVA, MVI, roof line, posterior left atrium	amiodarone, DCCV	12	74% vs 58% in sinus rhythm	Decrease in symptom severity if sinus rhythm is maintained
Jais et al[195]	2008	53 vs 59	PAF	SOPVI, linear ablation, CTI	flecainide, propafenone, sotalol, amiodarone, disopyramide, dofetilide, cibenzoline	12	89% vs 23% no recurrence	Symptom scores, exercise capacity and QOL measures better in ablation group
Wilber et al[196]	2010	106 vs 61	PAF	CPVA, linear ablation, CFE, CTI	dofetilide, flecainide, propafenone, sotalol, quinidine	9	66% vs 16% free from treatment failure	Better QOL measures and symptom scores in ablation group

AAD = Anti-Arrhythmic drugs, SOPVI = segmental ostial pulmonary vein isolation, CPVA = circumferential pulmonary vein ablation, CTI = cavotricuspid isthmus, MVI = mitral valve isthmus

All study patients had drug-refractory AF, except for Wazni et al, where ablation was tested as first line treatment.

1.10.7 Structural Remodelling following AF ablation

As discussed previously, increased atrial size can be both a contributing cause and also an effect of the process of AF. Studies examining the effects of ablation have shown that left atrial dimensions are reduced following AF ablation.[199-201] This may be due to a combination of maintenance of sinus rhythm and also scar formation after ablation. The effect of ablation on left atrial transport is however less clear and there appears to be conflicting data on this.[201, 202]

Catheter ablation of AF has also been shown to result in remodelling of the left ventricle. Increases in left ventricular ejection fraction, and decrease in diastolic and systolic dimensions were demonstrated in a study of patients with congestive heart failure undergoing ablation of AF.[203] A separate study demonstrated an improvement in both left ventricular systolic and diastolic function after restoration of sinus rhythm by catheter ablation of AF.[200]

1.10.8 Advancements in ablation technology

Since the pioneering work on catheter ablation of AF presented by Haissaguerre et al in 1998, there has been rapid development of technology to assist and improve ablation techniques. These included the use of non-fluoroscopic 3D navigation within the atria,[204] and also the use of robotic navigation.[205, 206] Several studies have evaluated different energy sources with novel catheter designs. Natale et al reported on the successful use of a through-the-balloon delivery of ultrasound energy for isolation of pulmonary veins.[207] Ablation using high intensity focused ultrasound[208] and cryoablation via balloon catheters[209] have also been reported. Endoscopic visualization to assist laser energy delivery through a balloon catheter is another

technology that is being evaluated.[210, 211] More recently, multi-electrode ablation catheters capable of delivering duty-cycled radiofrequency energy have been described and found to show early promising results.[212, 213]

1.11 Summary

The mechanisms of AF remain incompletely understood. Maintenance of sinus rhythm has not been convincingly shown to confer advantages compared to a rate control strategy, possibly due to adverse effects of anti-arrhythmic medication used to pursue a rhythm control strategy. By inference, catheter ablation for AF, which has the potential of achieving maintenance of sinus rhythm with minimal or no use of anti-arrhythmic medication, may possibly result in net benefit. To clearly define its place in the treatment of AF, large-scale randomized control studies are necessary to assess the long-term efficacy of catheter-based AF ablation and its effects on symptoms and quality of life, as well as morbidity and mortality, compared to conventional drug treatment. The CABANA study[214] (Catheter Ablation vs Anti-arrhythmic drug therapy for AF) is one such study which is currently still enrolling patients.

While the outcomes for catheter ablation of paroxysmal AF are on the whole satisfactory, persistent or long-standing AF ablation continues to pose a significant challenge. Success rates are generally lower than that of paroxysmal AF and procedures are often prolonged, complicated and technically challenging. The study of intracardiac electrogram characteristics of persistent AF can help further our understanding of this enigmatic arrhythmia. The rest of the thesis will examine this in detail, predominantly with the use of frequency domain analysis. A series of studies involving human subjects undergoing catheter ablation of AF will be outlined in the chapters that will follow.

Chapter 2
Methodology

2.1 Introduction

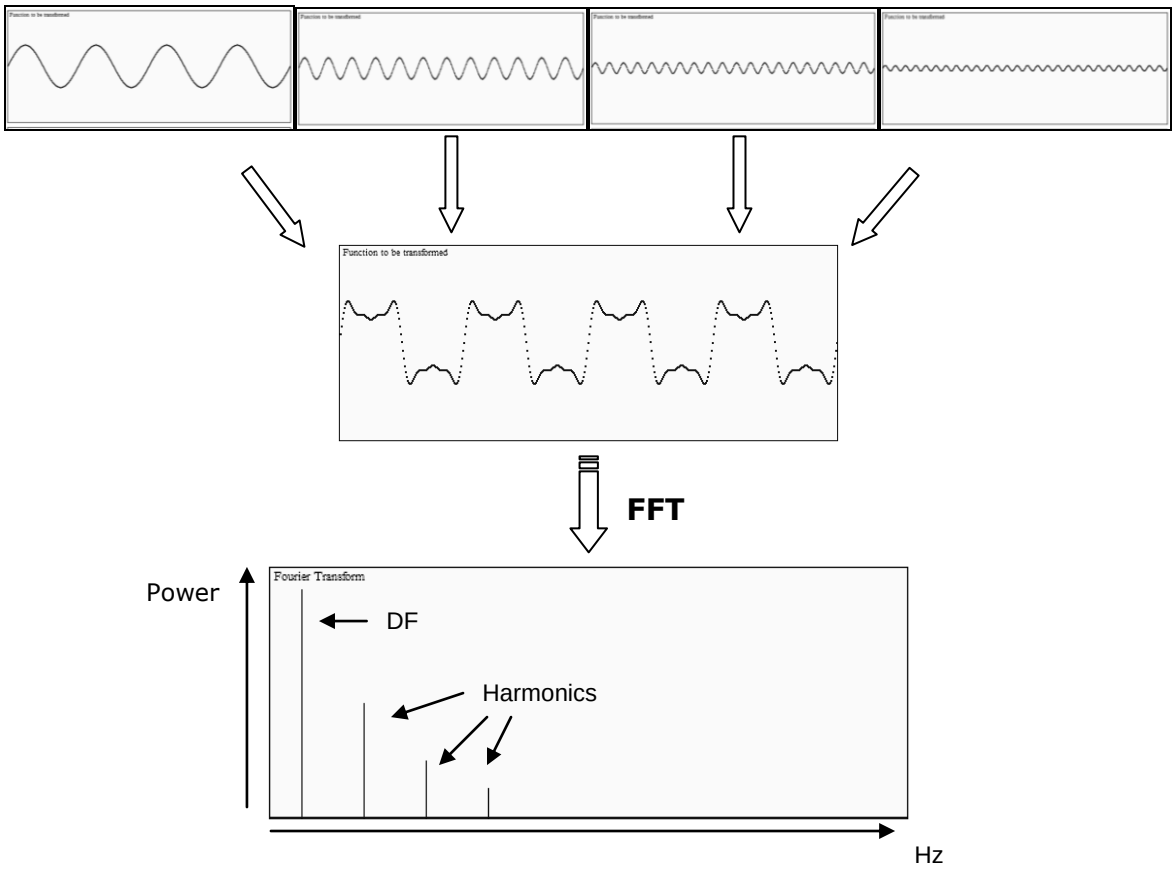
The atrial activation in AF is typically thought to be random and chaotic. There is however, evidence of spatial organization[215] and also of "linking" of wavelet propagation demonstrated by studies on human AF,[216] suggesting that the process of AF may not be as disorganized as previously thought. Early studies of AF electrograms using frequency domain analysis showed the presence of discrete dominant frequency peaks in the power spectra,[217-219] which could indicate regions of atrial activation at a similar rate. The interest in this subsequently led to application of frequency domain analysis to electrograms from high resolution optical mapping, where it was found that spatiotemporal periodic activity was present during AF.[95] Several other studies have since followed on to study the complex spatiotemporal characteristics of AF by utilising frequency domain analysis.[96, 178, 179, 220-222]

2.2 Fourier Transform

Fourier transformation is a mathematical operation that allows a continuous signal, which is made up by the summation of sinusoidal waveforms, to be broken down into its constituent frequencies. A computationally efficient way of doing this is by using the Fast Fourier Transform (FFT). One of the requirements for performing FFT is that the number of time samples used is a power of 2 (e.g. 512, 1024, 2048, 4096). The resultant frequency spectrum produced will lie between 0 Hz to half of the sampling frequency. The frequency resolution is determined by dividing the sampling frequency by the number of points (or time samples) recorded. A signal that is perfectly periodic, will contain a dominant frequency (DF) (the sinusoidal waveform with the largest amplitude) and harmonic frequencies (from the remaining sinusoidal

waveforms constituting the signal) at multiples of the dominant frequency. (Figure 2.1)

Figure 2.1: Summation of sine waves giving rise to a cumulative square wave



FFT produces a power spectrum showing the component frequencies consisting of the DF and its harmonic frequencies.

2.2.1 Aliasing

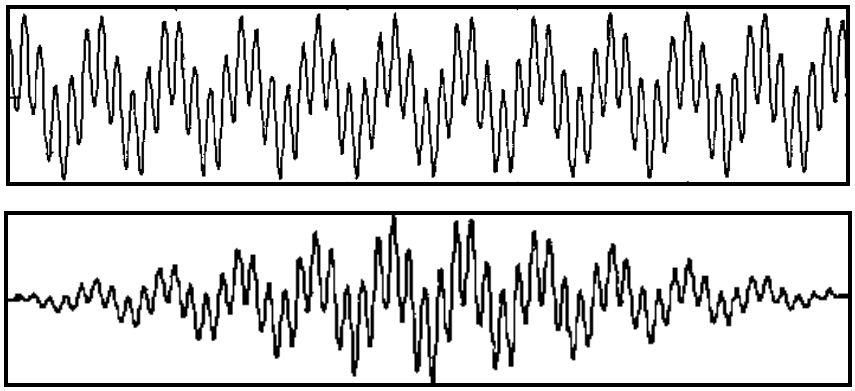
To avoid any errors from under-sampling a signal of interest , the sampling frequency must be at least twice the maximum frequency of the signal, as stated by the Nyquist criteria.[223] Distortion or errors in the frequency spectrum from an inadequate sampling rate is known as aliasing. Apart from increasing the sampling rate, aliasing can be countered by pre-processing the signal via a low pass filter for example, to cut off any unwanted frequencies above a certain range.

Frequency domain analysis works best at reflecting periodicity of a signal when it looks like a sine wave. To ensure optimal analysis of an intracardiac electrgram by FFT, a number of pre-processing steps are necessary.[215, 224, 225] These include bandpass filtering at (e.g. 40 - 250 Hz) to further accentuate the signal, rectification (by using absolute values) to obtain a monomorphic waveform, low pass filtering at 20 Hz to restrict analysis to frequencies within the physiological spectrum of activation, and application of “windowing” functions to counter the effect of spectral leakge which is discussed below.

2.2.2 Spectral Leakage

In the presence of frequency components that are not integer multiples of the sampling period, spectral leakage will be seen with finite length sampling. This is due to abrupt cut-off of signals at the edges of the sampling interval, resulting in inaccurate representation of the frequency spectrum. To minimize this effect, “anti-leakage” windows are applied, which essentially involves modification of the original signal by multiplying a windowing function to attenuate the edges of the signal to zero (Figure 2.2). This results in sharpening and accentuation of the peaks in the frequency spectrum, hence reducing any potential errors in identifying the dominant peak and its harmonics. Several windowing functions exist, which offer varying balance between reducing spectral leakage at the expense of loss of spectral detail. Popular choices include both the Hanning and Hamming window functions, which have been described in the frequency analysis of human AF.[178, 226, 227]

Figure 2.2: Window Function



Original signal (top) and appearance after application of a Hamming window function (bottom) which attenuates the signal at the edges of the segment.

2.2.3 Dominant Frequency and Organization Index

Once all pre-processing steps have been carried out, the signal can be subjected to FFT to obtain a frequency power spectrum. In the context of analysing AF, the DF of the electrogram is the frequency with the tallest peak in the power spectrum,[95, 228] which is also the definition used in the studies carried out for this thesis. Multi-site sampling of the atria will allow for the identification of areas of maximal DF, which can be defined as sites demonstrating high frequency relative to adjacent sites, with a decreasing frequency gradient of ≥ 20%.[178] If the electrogram signal is adequately periodic and sinusoidal in morphology, the DF will closely approximate the rate of local atrial activation. Several studies have previously demonstrated good correlation between cycle length of AF and DF of the signal,[95, 227, 229] although a recent study was unable to replicate this.[230] Differences in types of electrograms sampled and method of frequency domain analysis between the studies may have accounted for this. Indeed, Ng et al[227] found good correlation between DF and AF activation rates measured in the time domain, but this relationship was weakened by increasing variation in signal amplitude and rate. In this study, averaging consecutive segments of DF reduced this distortion.

Additionally, frequency domain analysis also allows for assessment of temporal organization and regularity of AF. This can be measured by examining the representation of the areas under the DF and its harmonics across the whole power spectrum. This is based on the concept that the more periodic and regular a signal is, the greater the concentration of the power (or area) under the DF and its harmonics. Using frequency domain analysis, an increase in the indices of organization have been found to be associated with increasing likelihood of termination of AF by DC shock,[220] burst pacing[221] and via ablation.[229] The method of assessing electrogram organization in the studies performed for this thesis is calculated as the ratio of the power under the DF and its harmonics (0.75 Hz) band, to the total power of the resulting power spectrum. As will be demonstrated later, (Figure 2.3) the FFT software displays this as a regularity index ("RI") but organization index (OI) is the preferred terminology used for this thesis and hence "OI" will be used to replace "RI".

2.2.4 Spatiotemporal Stability

The spatiotemporal stability of electrograms in the frequency domain has been demonstrated in both animal[95, 96] and human studies of AF.[178, 229, 231] However, very recently published work using prolonged 5 minute recordings and multi-electrode contact mapping of AF has disputed this observation, finding temporal variability in sequential DF maps.[232] Despite there being more evidence in the literature supporting stability of DF, it remains important to acknowledge that there could be potential limitations in this technique and further studies into this are warranted. Previous studies have also suggested recording electrograms over at least 5s to increase the accuracy of fractionation and DF.[233, 234]

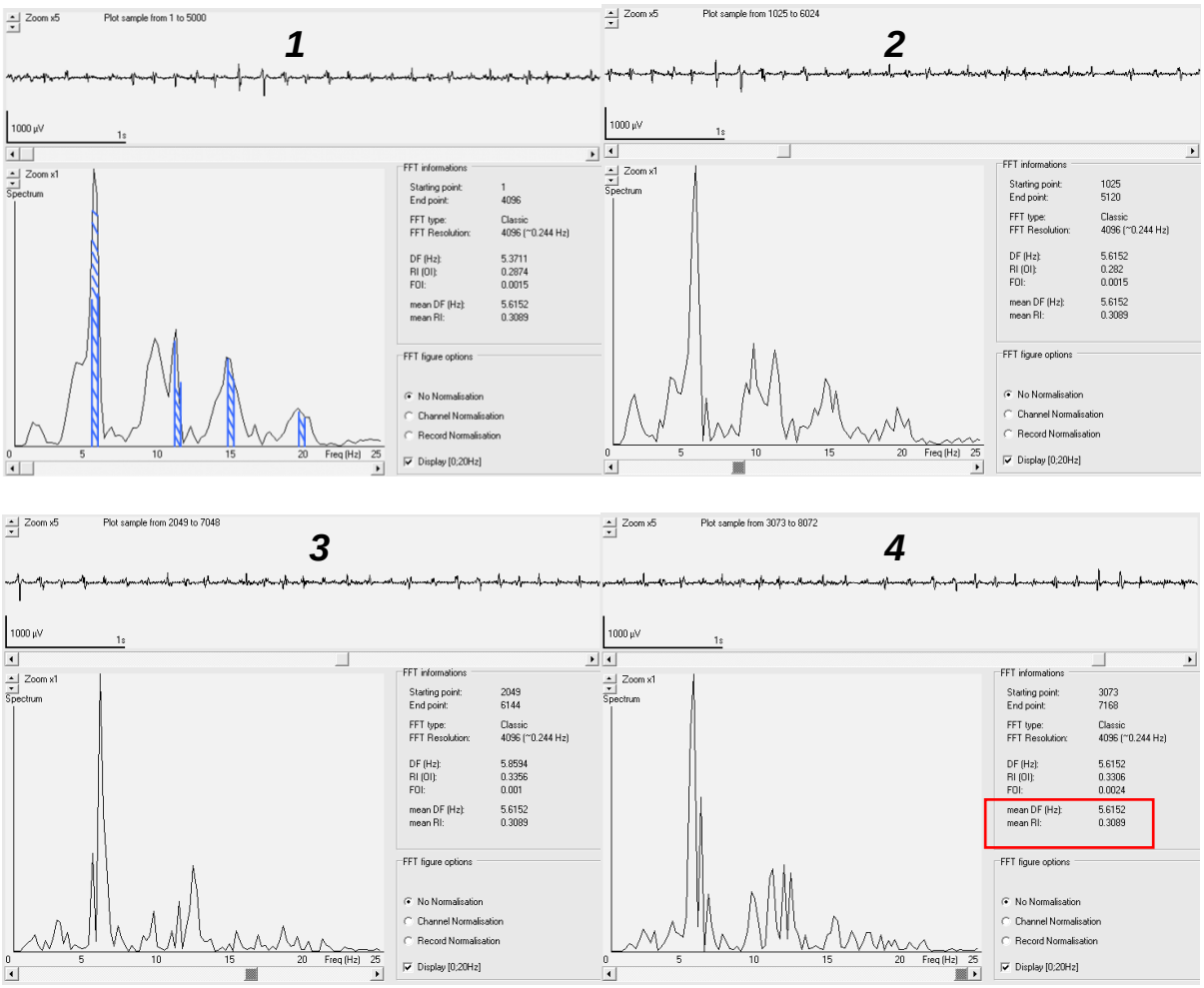
2.3 Frequency Domain Analysis Specific to the Thesis

In this thesis, the same FFT formula and processing methods are used for all electrograms recorded (bipolar in all cases) using FFT computation software version 3.2.0.0 (Bard Electrophysioplogy, Lowell, MA, United States). More detailed description of the specific analysis of the frequency spectrum carried out for each study can be found in the individual chapters. Broadly speaking, recording and pre-processing steps used are:

1. Sampling frequency of 1000 Hz
2. Signal acquisition filter of 30-250 Hz
3. Power processing (rectification)
4. Application of Hamming window
5. Bandpass filtering at 1-20 Hz

The duration of electrogram analysis is adapted and varied according to the conditions of each study. A 4096 point FFT is used in all cases. At a sampling rate of 1000 Hz, this gives a frequency resolution of 0.24 Hz. A 1024 point sliding window is used to detect and track any changes in the electrograms over the full duration of each electrogram recording, so that DF and OI values (as defined earlier) are obtained at each slide/shift in the window. The average of these DF and OI values for each location are then used for final data analysis (Figure 2.3).

Figure 2.3: Technique of frequency domain analysis



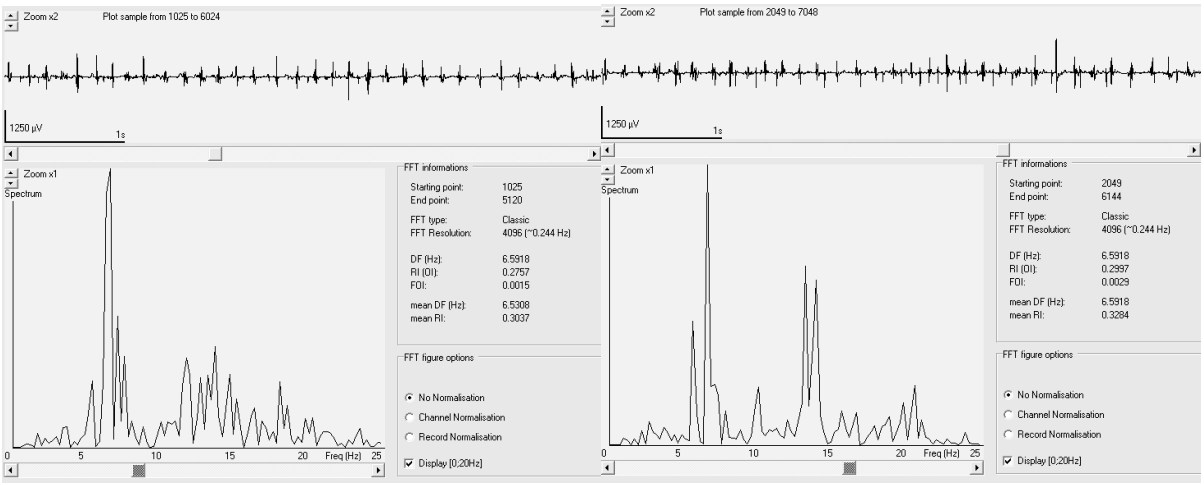
The above example is an 8s electrogram which is subjected to a 4096 point FFT from the start to the end (panels labelled 1-4 sequentially) to obtain DF and OI at each 1024 point slide or shift of the FFT window.The resultant mean DF and OI over the entire segment is highlighted in the red box. [in all cases, "mean RI" displayed in the result is defined as the mean OI in this thesis; this is calculated as the ratio of power under the DF and its harmonics (0.75 Hz band, blue shaded areas in panel 1) to total power of resulting power spectrum (unshaded area under curve)].

2.3.1 Test of Temporal Stability

Intra-cardiac electrograms collected from the studies described in the latter chapters are predominantly from signals recorded from decapolar catheters placed in the coronary sinus. This has the advantage of being able to maintain a stable position over long periods of time to allow for recording of electrograms from the same site. To test for temporal stability of frequency domain parameters (DF and OI) used in the

studies, electrograms were analysed from all the 5 recording bipoles of the coronary sinus catheter and compared at the start and end of a 10 minute period of recording. DF and OI obtained (over 10s) from each of the 5 bipoles were averaged and compared after discarding signals with far-field artefact. This analysis was carried out in 5 patients with persistent AF. No significant differences were seen between baseline and after 10 minutes (Figure 2.4), suggesting temporal stability of DF and OI. [DF 5.7 ± 0.4 Hz vs 5.8 ± 0.5 Hz (p = 0.20) and OI 0.27 ± 0.02 vs 0.26 ± 0.01 (p = 0.63) respectively]

Figure 2.4: Test for temporal stability of DF and OI



Examples of FFT of electrograms (from the same position in the coronary sinus, CS9-10) recorded at baseline (left) and 10 minutes later (right) showing comparable mean DF and OI (DF = 6.5 Hz and 6.6 Hz, OI = 0.30 and 0.33 respectively).

2.3.2 Time Domain vs Frequency Domain Analysis

Analysis of atrial electrograms in the time domain analysis essentially involves the detection of deflections and measuring the interval between deflections over time (e.g. AF cycle length, complex fractionated electrograms). The complex, variable and sometimes continuous electrogram signals seen in AF can make this assessment difficult. Manual or visual analysis of electrograms in the time domain can be tedious

and subjective, and automated detection algorithms may not be able to fully accommodate the beat-to-beat variation in electrogram characteristics. Frequency domain analysis provides a more objective assessment of electrogram activity and can be easily and rapidly applied to complex signals using pre-determined signal processing steps. Apart from obtaining the DF of the signal, additional useful information such as organization or regularity of the signal can also be conveniently assessed at the same time by measuring parameters such as the OI.

At the same time however, it is also important to recognise that there are also limitations with the technique of frequency domain analysis. It is dependent on signal quality and is also vulnerable in some cases to varying characteristics of the electrogram, similar to time domain analysis. Although its spatiotemporal stability may remain under scrutiny, we have found our technique to be stable from the same recording position. If care is taken to ensure consistent acquisition of good quality signals together with strict application of pre-processing steps, FFT has the potential to be a valuable tool for rapid, real-time assessment of AF characteristics.

Chapter 3
Increase in Organization Index predicts Atrial Fibrillation termination with Flecainide post-ablation – spectral analysis of intracardiac electrograms

3.1 Introduction

Despite their widespread use in the treatment of atrial fibrillation (AF), the mechanism by which class Ic anti-arrhythmic drugs terminate AF is not well understood. Early studies on the effects of flecainide[89] and propafenone[90] in canine AF suggested that termination of AF was the result of a rate-dependent increase in atrial effective refractory period, which led to an overall increase in wavelength. However, more recent work on goat AF failed to show this relationship, but instead, found that widening of the temporal excitable gap to be consistently associated with AF termination with flecainide, as was the case with several other anti-arrhythmic drugs used in the study.[235] These studies utilised different AF induction protocols in different animals, and also employed different methods of measuring atrial refractory periods, which make direct comparison difficult. Moreover, none of these proposed theories of pharmacological AF termination have been adequately tested in human AF, the mechanism of which remains incompletely understood.

Recent studies of intracardiac signals during catheter ablation for AF in humans have suggested that prolongation of AF cycle length[236] and increased organization[229] of AF are associated with return to sinus rhythm (SR). Whether these observations can also reliably predict pharmacological termination of human AF is unclear. To gain greater understanding into the mechanism of AF termination using class Ic anti-arrhythmic drugs, we used flecainide to assess its effect on time and frequency domain characteristics of human AF, using intracardiac signal analysis. We hypothesized that flecainide will prolong AF cycle length, reduce the dominant frequency of AF, as well as increasing its organization prior to termination.

3.2 Methods

Patients who were given flecainide during catheter ablation for AF were studied. All documented AF during the ablation procedures were either spontaneous or already pre-existent. No programmed AF induction was carried out in any patient. Paroxysmal AF is defined as AF that terminates spontaneously within 7 days, while persistent AF is defined as AF which is sustained beyond 7 days or lasting less than seven days but requiring pharmacological or electrical cardioversion. All anti-arrhythmic therapy was discontinued for more than 5 half-lives prior to the procedure.

AF ablation was carried out using bilateral femoral venous access. Under fluoroscopic guidance, a deflectable decapolar catheter and a quadripolar catheter were positioned in the coronary sinus and His position respectively. A circular pulmonary vein mapping catheter and a deflectable, irrigated tip ablation catheter were advanced into the left atrium after transseptal puncture. Standard segmental ostial pulmonary vein isolation alone (n=13) was performed in patients with paroxysmal AF. More extensive ablation was carried out in patients with persistent AF or when paroxysmal AF failed to terminate with segmental ostial pulmonary vein isolation alone. This consisted of either 1) segmental ostial pulmonary vein isolation with linear ablation, followed by complex fractionated electrogram ablation (n=10), or 2) wide area circumferential ablation followed by linear ablation (n=3). In all cases, left atrial anatomical geometry was created using EnSite NavX^{TM} electroanatomical mapping (St. Jude Medical Inc, St Paul, Minnesota USA). Patients who remained in AF at the end of the ablation procedure (either in AF at the start or developed AF during ablation) were given flecainide. This was administered at a dose of 2 mg/kg body mass (maximum of 150 mg) as intravenous infusion over 10 minutes, followed by observation for 10 minutes for any change in cardiac rhythm.

3.2.1 Time and frequency domain analysis

Intra-cardiac electrograms were recorded on a 30-250Hz filter with sampling frequency of 1 kHz (Labsystem Pro, Bard Electrophysiology, Lowell, MA, USA). Time and frequency domain analyses were carried out offline. Analysis was carried out on bipolar AF electrograms recorded from the decapolar catheter in the coronary sinus at the end of catheter ablation for AF. All electrograms with good signal quality on the decapolar catheter were analysed (signal:noise amplitude ratio of > 2). Readings were taken prior to flecainide infusion, as well as after the full dose of flecainide had been given. In the event of return to SR after flecainide infusion, electrograms just prior to rhythm change were analysed. Cycle length (CL) measurements were carried out using built-in autodetection software (Labsystem Pro, Bard Electrophysiology, Lowell, MA, USA) and also verified manually. Mean CL was calculated over a 10s recording period. The same 10s segment was exported and analysed using Fast Fourier Transform with a spectral resolution of 0.24Hz (4096 points), after processing with a Hamming window. A 1024 point sliding window was used to give the mean DF and OI of the signal. The DF and OI of all suitable coronary sinus electrograms across all 5 bipoles were averaged for each patient and used for analysis.

3.2.2 Statistical analysis

All continuous variables are expressed as mean +/- standard error of mean. Normally distributed data were analysed using paired and unpaired Student's t-test as appropriate. Non-parametric data were analysed by Mann Whitney or Wilcoxon signed rank test. Categorical data were analyzed using Chi squared or Fisher's exact test. Logistic regression was carried out to identify independent predictors of

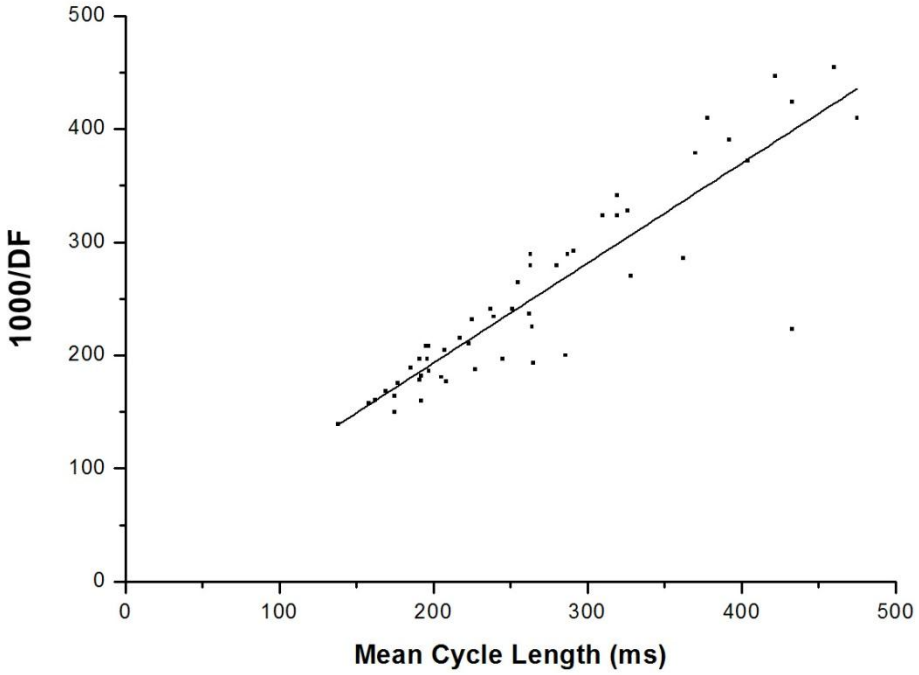
outcome after flecainide infusion and Receiver operator characteristic analysis was used to evaluate the relevant parameter. A p value of <0.05 was considered statistically significant.

3.3 Results

A total of 26 patients were included in the study. Sixteen (62%) had paroxysmal AF and 10 (38%) had persistent AF. Four of the patients with paroxysmal AF were in AF at the start of the procedure while the remaining 12 developed AF spontaneously during the procedure. All patients with persistent AF started the procedure in AF. Seven (27%) converted to SR with flecainide (6 paroxysmal AF, 1 persistent AF). Average time to cardioversion with flecainide was 252 ± 36s after initiating the infusion. Taking all patients together, mean CL increased from 211 ± 9 ms to 321 ± 17 ms ($p < 0.0001$) after flecainide. Mean DF decreased from 5.2 ± 0.2 Hz to 3.6 ± 0.2 Hz ($p < 0.0001$). The mean OI remained unchanged [0.33 ± 0.02 before and 0.32 ± 0.02 after flecainide ($p = 0.90$)]. The inverse of the DF (1000/DF) revealed a strong linear relationship with mean CL (Pearson correlation coefficient = 0.90, $p < 0.0001$) (Figure 3.1).

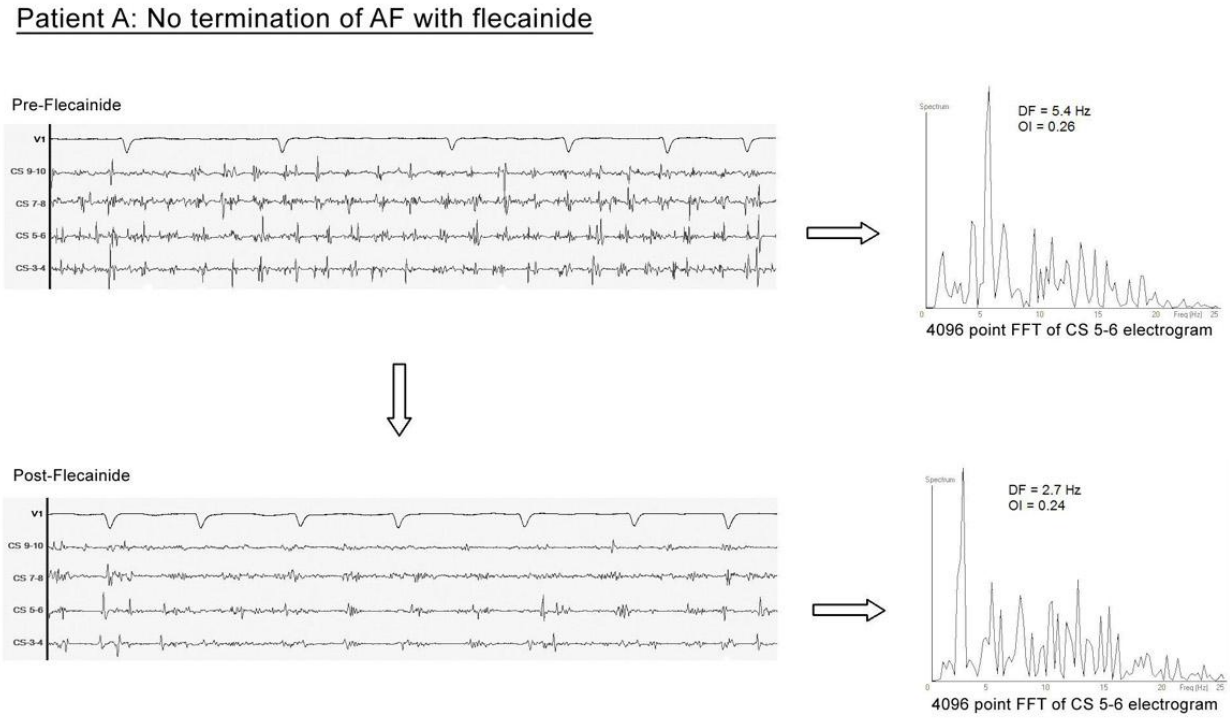
Representative examples of intracardiac recordings and frequency spectrum analysed for this study are shown in Figure 3.2 and Figure 3.3. Figure 3.2 shows data on the ECG (lead V1) and intracardiac electrograms before and after intravenous flecainide in a patient whose AF did not terminate with flecainide together with the corresponding spectral data from FFT analysis, whilst Figure 3.3 shows the same analysis in a patient whose AF converted to SR with flecainide.

Figure 3.1: 1000/DF vs mean Cycle Length



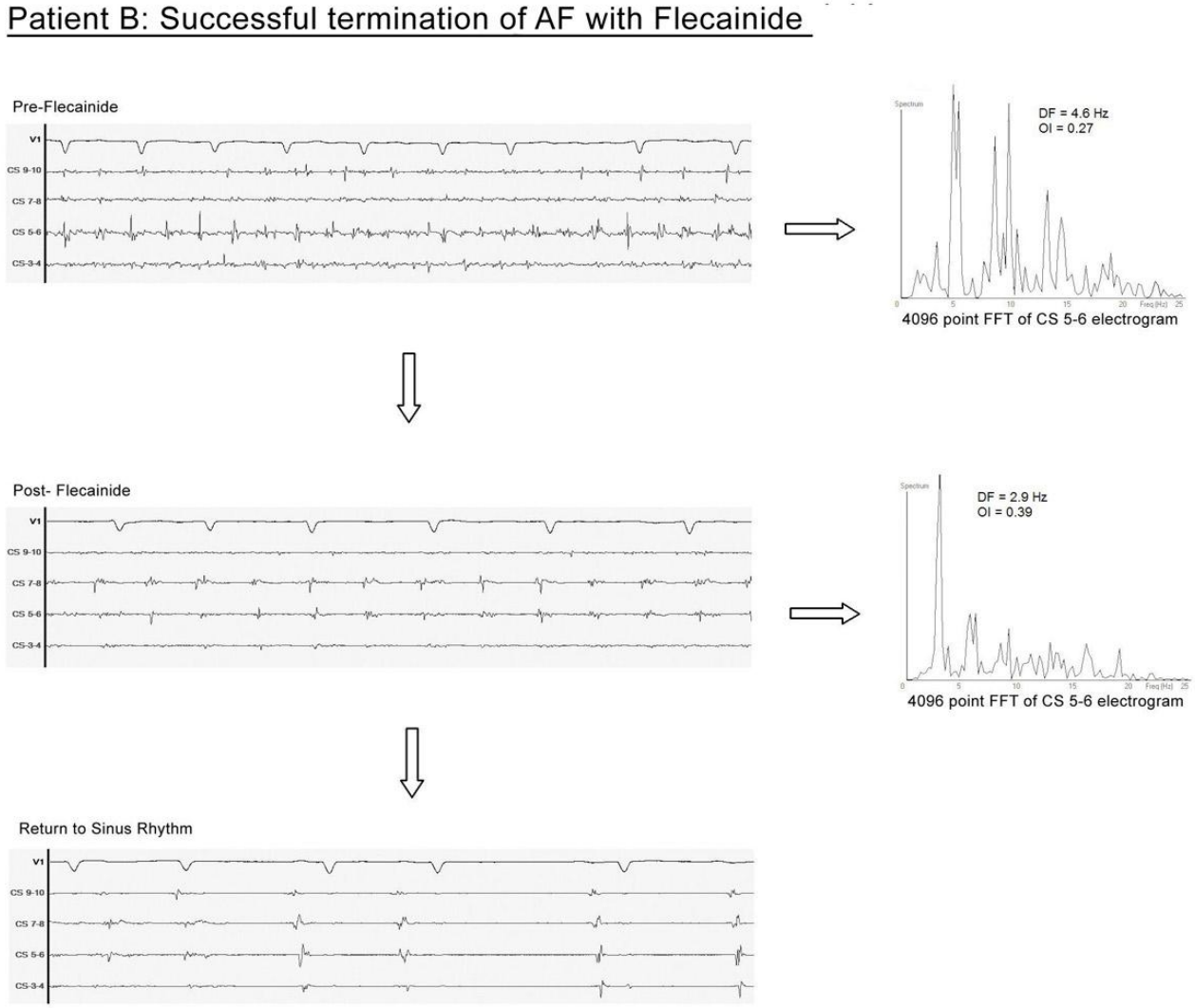
Plot of mean cycle length during atrial fibrillation and the inverse of corresponding dominant frequency (1000 / DF) in all patients with and without intravenous flecainide showing a linear relationship between the 2 parameters (Pearson correlation coefficient = 0.90, $p < 0.0001$).

Figure 3.2 Example of no termination of AF after flecainide



Typical example of a patient (Patient A) in whom AF did not terminate with intravenous flecainide. The left hand panels show ECG lead V1 and intracardiac electrograms before and after flecainide (both at paper speed 50mm/s). The right hand panels show the corresponding frequency spectra with dominant frequency decreasing from 5.4 Hz to 2.7 Hz, but organization index showed little change (0.26 pre- and 0.24 post-flecainide) with flecainide.

Figure 3.3: Example of termination of AF after flecainide



Typical example of a patient (Patient B) in whom AF terminated with intravenous flecainide. The left hand panels show ECG lead V1 and intracardiac electrograms before and after flecainide and after returning to sinus rhythm (all at paper speed 50mm/s). The right hand panels show the corresponding frequency spectra with dominant frequency decreasing from 4.6 to 2.9 Hz and organization index increased from 0.27 to 0.39 before return to SR.

Comparing patients who converted to SR after flecainide, with those who remained in AF, OI post-flecainide was 0.41 ± 0.04 vs 0.29 ± 0.02 (p = 0.013) and relative change in OI was 29 ± 13 vs -3.9 ± 6% (p = 0.016) respectively. No significant differences were noted in the mean CL and DF post-flecainide and similarly, no significant differences were seen in the relative change in CL and DF in the 2 groups. Mean CL post-flecainide was 335 ± 34 vs 315 ± 20 ms (p = 0.61), mean DF was 3.2 ± 0.3 vs 3.8 ± 0.2 Hz (p = 0.20), relative change in CL was 76 ± 24 vs 48 ± 8% (p = 0.16) and relative change in DF -37 ± 9 vs -25 ± 4% (p = 0.21), when comparing patients who returned to SR after flecainide with those who stayed in AF respectively. There were no significant differences in the CL, DF or OI between the 2 groups prior to flecainide administration. Detailed characteristics of the study patients can be found in Table 3.1. Logistic regression analysis identified that a greater relative increase in OI (p = 0.04), a higher OI post-flecainide (p = 0.03) and SR at start of procedure (p = 0.03) to be independently associated with successful reversion to SR with flecainide. Receiver operator characteristic curves indicate that an OI of > 0.33 and a relative increase in OI of > 14.8%, independently have a 71% sensitivity and 79% specificity for identifying those who will return to SR after flecainide administration [Area under curve 0.82, (p = 0.01) and 0.77 (p = 0.03) respectively].

Table 3.1: Patient Characteristics

	Successful Termination of AF with flecainide (n=7)	Failed Termination of AF with flecainide (n=19)	P value
Age	48 ± 9	55 ± 11	P = 0.14
Sex	6 Male (86%)	14 Male (74%)	P= 0.65
Paroxysmal AF	6 (86%)	10 (53%)	P= 0.19
Persistent AF	1 (14%)	9 (47%)	
Hypertension	4 (57%)	8 (42%)	P = 0.67
Ejection Fraction (%)	52 ± 5	54 ± 3	P = 0.28
Left atrial dimension (cm)	4.1 ± 0.6	4.3 ± 0.6	P= 0.66
Rhythm at start of procedure:			P = 0.026
- SR	6 (86%)	6 (32%)	
- AF	1 (14%)	13 (68%)	
Ablation Strategy:			P = 0.53
- PVI Alone	4	9	
- PVI + CFE ablation	3	7	
- WACA	0	3	

Total ablation times (s)	2963 ± 334	3424 ± 426	P = 0.52
CL pre-flecainide	202 ± 20	214 ± 10	P = 0.53
CL post-flecainide	335 ± 34	316 ± 20	P = 0.61
Absolute change	134 ± 38	101 ±16	P = 0.37
% Change in CL	76 ± 24	48 ± 8	P = 0.16
DF pre-flecainide	5.4 ± 0.5	5.1 ± 0.2	P = 0.59
DF post-flecainide	3.2 ± 0.3	3.8 ± 0.2	P = 0.20
Absolute change	-2.2 ±0.6	-1.33 ± 0.2	P = 0.12
% Change in DF	-37 ± 9	-25 ± 4	P = 0.21
OI pre-flecainide	0.33 ± 0.04	0.32 ± 0.03	P = 0.90
OI post-flecainide	0.41 ± 0.04	0.29 ± 0.02	P = 0.013
Absolute change	0.08 ± 0.04	-0.03 ± 0.02	P = 0.017
% Change in OI	29 ± 13	-3.9 ± 6	P = 0.016

PVI = pulmonary vein isolation (segmental ostial), CFE = complex fractionated electrograms, WACA = wide area circumferential ablation, CL = cycle length, DF = dominant frequency, and OI = organization index

3.4 Discussion

This study shows, using spectral analysis of intracardiac electrograms, that the mechanism of human AF termination by flecainide is critically preceded by organization of the arrhythmia, as reflected by an increase in OI. While CL prolongation and corresponding reduction in DF occurred with flecainide administration, they did not predict return to SR. This suggests that an increase in organization of AF is a more sensitive predictor of cardioversion to SR than the change in DF or CL.

3.4.1 Mechanism of AF Termination with Flecainide

Class Ic anti-arrhythmic drugs block fast sodium channels on cardiac cell membranes by a rate dependent process[90, 237, 238]. While flecainide has been widely used in clinical practice to treat AF, its mechanism of action in terminating AF is still not fully understood.

According to the multiple wavelet hypothesis proposed by Moe and Abildskov[68], AF is the result of multiple wavelets of re-entrant circuits propagating throughout the atria. The arrhythmia is thought to be more likely to sustain itself with increasing number of wavelets. An experimental model of AF by Allessie et al[86] demonstrated that a critical number of wavelets (3-6) are required for the perpetuation of AF.

The relationship between re-entry and wavelength was first described by Mines[75] and also subsequently by Lewis[239]. Wavelength can be described as the minimal pathlength that supports re-entry. This was later formulated mathematically by Wiener and Rosenblueth[81], who showed that wavelength (WL) [defined as a product

of refractory period (RP) and conduction velocity (CV): WL = RP X CV] critically determined the likelihood of re-entry. The “leading circle” concept of re-entry [87] as proposed by Allessie et al further suggested that the dimension of a functional re-entrant circuit is dependent on its wavelength (i.e. a smaller wavelength is associated with a smaller circuit), which in turn is determined by the electrophysiological properties of the tissue.

Based on the above concepts, it can be understood that an increase in wavelength (associated with larger circuit size) reduces the number of re-entrant circuits the atria can contain, hence reducing the likelihood of AF being sustained. The use of wavelength as an index to predict susceptibility to atrial arrhythmia has been studied previously by Rensma et al[88] and found to be predictive in 75% of cases, making it a useful way to assess anti-arrhythmic potential of drugs. Asano et al [240] used wavelength index as a surrogate marker of wavelength in the evaluation of AF and found that an increase in this index and hence wavelength, preceded AF termination, either spontaneously or with disopyramide.

This concept has led to the question of how class Ic anti-arrhythmic drugs, which decrease conduction velocity without any apparent effect on refractory period, can result in the termination of AF. Wang et al[238] examined the response of human atrial tissue to flecainide and quinidine in comparison with that of guinea pig, rabbit and dog. They concluded that flecainide induced increases in atrial action potential duration and refractoriness enhanced by the rapid rates typical of AF, with human tissue showing greatest frequency-dependent drug effects. In a separate study, Wang et al [89] demonstrated in a dog vagal AF model, that flecainide terminated AF by causing a tachycardia dependent increase in atrial effective refractory period (to a

greater extent than reduction in conduction velocity) which increased the wavelength. This same study also showed that flecainide progressively increased the size and reduced the number of re-entry circuits, as well as slowing atrial activation until AF terminated. The same effect of rate dependent wavelength prolongation was also observed when propafenone, procainamide and sotalol were evaluated in the termination of experimental AF in dogs[90].

However, the theory that flecainide and class Ic anti-arrhythmic drugs in general can influence wavelength changes has been put in question by other studies. Wijffels et al[235] tested a number of anti-arrhythmic drugs in chronic AF in the goat and found that termination of AF was not attributed to wavelength prolongation. Class I agents used in the study (flecainide and cibenzoline) actually shortened wavelength of AF. Widening of the temporal excitable gap was the only finding correlated to pharmacological cardioversion of AF in this study. An earlier study on flecainide in humans[241], also failed to show any significant effect on atrial refractoriness. Similarly, Katristis et al[242] found no change in atrial effective refractory period with flecainide infusion and pacing at different cycle lengths in human subjects.

Further work using a mathematical model of AF to assess AF termination was carried out by Kneller et al[243]. It was found that pure sodium channel blockade terminated AF without prolonging wavelength, and likely mechanisms include the enlargement of the core size of primary rotors, increasing meander and extinction at boundaries and reduction in the number of wavelets.

3.4.2 Frequency Domain Analysis of AF

Using spectral analysis of canine AF, Everett et al showed that high AF organization increased the efficacy of burst pace termination[221] and also electrical cardioversion[220] of AF.

Bollmann et al[244] studied the use of oral flecainide in patients with persistent AF by carrying out frequency spectrum analysis on high resolution surface ECG recordings. A lower fibrillatory frequency post-flecainide and smaller left atrial size were found to predict restoration of SR. Using a similar technique, Husser et al[245] carried out time-frequency analysis of surface electrocardiogram of AF to compare effects of flecainide and amiodarone with baseline. A reduction in both fibrillatory rate (converted from dominant frequency) and also exponential decay of the frequency spectrum were seen after drug administration, with effects more apparent in the flecainide group. The authors suggested that a smaller exponential decay, seen after both flecainide and amiodarone, is observed in organized rhythms and could be reflective of organization of AF.

3.4.3 AF Cycle Length analysis

AF CL is generally thought to reflect local atrial refractoriness[246]. However, assessing the exact mechanism of AF CL prolongation in the setting of flecainide administration is difficult as it is believed to have a use-dependent effect on atrial refractoriness and conduction velocity[89, 237, 238], both of which can result in an increase in AF CL. Biffi et al[247] studied induced AF in 10 patients with paroxysmal AF. The mean of 100 consecutive AF intervals (measured from endocardial electrograms) was increased by both flecainide and propafenone prior to AF termination in all the study patients.

All 10 patients returned to SR within 10 minutes of drug administration. The authors also remarked that the extent of AF interval prolongation was greater with drugs than that observed in self-terminating AF in a previous study[248] also carried out by the same group.

3.5 Implication of findings

The mechanism of AF termination with sodium channel blockade remains poorly understood. Using spectral analysis, we have demonstrated, in human subjects, that AF CL prolonged and DF reduced uniformly after flecainide administration; however, this did not predict return to SR. Reversion to SR with flecainide was independently associated with a higher OI post-flecainide and also a greater relative rise in OI induced by flecainide, regardless of change in CL or DF. This suggests that the process of AF termination depends critically on an increase in organization of the arrhythmia, possibly even more crucially so than slowing of atrial activation. This implies that increase in the organization of AF is likely to be a more sensitive predictor of return to SR than the change in DF or CL. While other studies have shown AF CL prolongation with class Ic anti-arrhythmics, none have used spectral analysis to quantify the organization of the rhythm and its relationship with return to SR in human AF.

A higher organization index is a marker of greater regularity. The increase in organization index seen in our study can be hypothesized to reflect a reduction in the number of re-entrant circuits or drivers, prior to AF termination. When coupled with the increase in AF CL and reduction in DF (and hence slowing of atrial activation rate), it results in conditions favourable for return to SR. An isolated increase in AF CL or reduction in DF alone, with no increase in organization probably represents

continued presence of multiple re-entrant circuits or drivers which reduces the likelihood of return to SR.

3.6 Limitations

Our study reflects the behaviour of the ablated left atrium. However, it is probably not dissimilar to the spectral characteristics of the un-ablated left atrium since ablation alone was not sufficient to achieve termination of AF in these patients, implying the continued presence of unaddressed drivers or substrate maintaining AF.

Using a catheter placed in the coronary sinus has the advantage of stability in terms of positioning, but we acknowledge that the electrograms recorded from this catheter are not directly from within the left atrium, and do not fully assess the global spatiotemporal characteristics of AF. However, in the context of assessing cycle length and spectral characteristics of human AF, the utilization of coronary sinus electrograms alone for analysis is well described in published literature[229, 236, 249, 250], and while it has its limitations, it seems to be an acceptable technique.

Though not achieving statistical significance, there appears to be a trend suggesting that CL and DF changes may be greater in those who returned to SR after flecainide than those who did not. Due to the small sample size, the study is not powered to conclusively exclude any link in CL and DF changes with the likelihood of returning to SR, although it still supports the notion that increase in OI is a more sensitive indicator.

3.7 Conclusion

Our findings provide an insight into the controversial mechanism of human AF termination with class Ic agents such as flecainide. The spectral characteristics observed in our study appear to support findings reported by researchers using canine AF [89] and mathematical AF models.[243] Similarly, our findings also reflect observations from termination of AF with catheter ablation,[229] burst pacing[221] and electrical cardioversion,[220] adding further weight to the evidence that increased spectral organization plays a crucial role in AF termination, regardless of method used to achieve this. The OI of AF appears to be a useful tool to assess for likelihood of AF termination and may be useful as a target for development of AF therapy. Larger studies into spectral organization of AF are needed to further define its significance in human AF.

Chapter 4
Spectral Characteristics and outcomes after Catheter Ablation for Persistent Atrial Fibrillation

4.1 Introduction

Catheter ablation for persistent atrial fibrillation (AF) remains challenging and clear end points to guide the extent of ablation are lacking, especially in the event of persistence of AF after carrying out extensive ablation. Moreover, although termination of AF by ablation is considered to be desirable, studies examining this have produced conflicting results, and it remains unclear if AF termination during ablaton is associated with better long term outcomes. [251-253]

Catheter ablation of AF has been shown to reduce dominant frequency and increase organization of intracardiac atrial electrograms.[181, 182, 229] Changes in these spectral characteristics of AF and their relationship to follow-up outcome is not well understood.

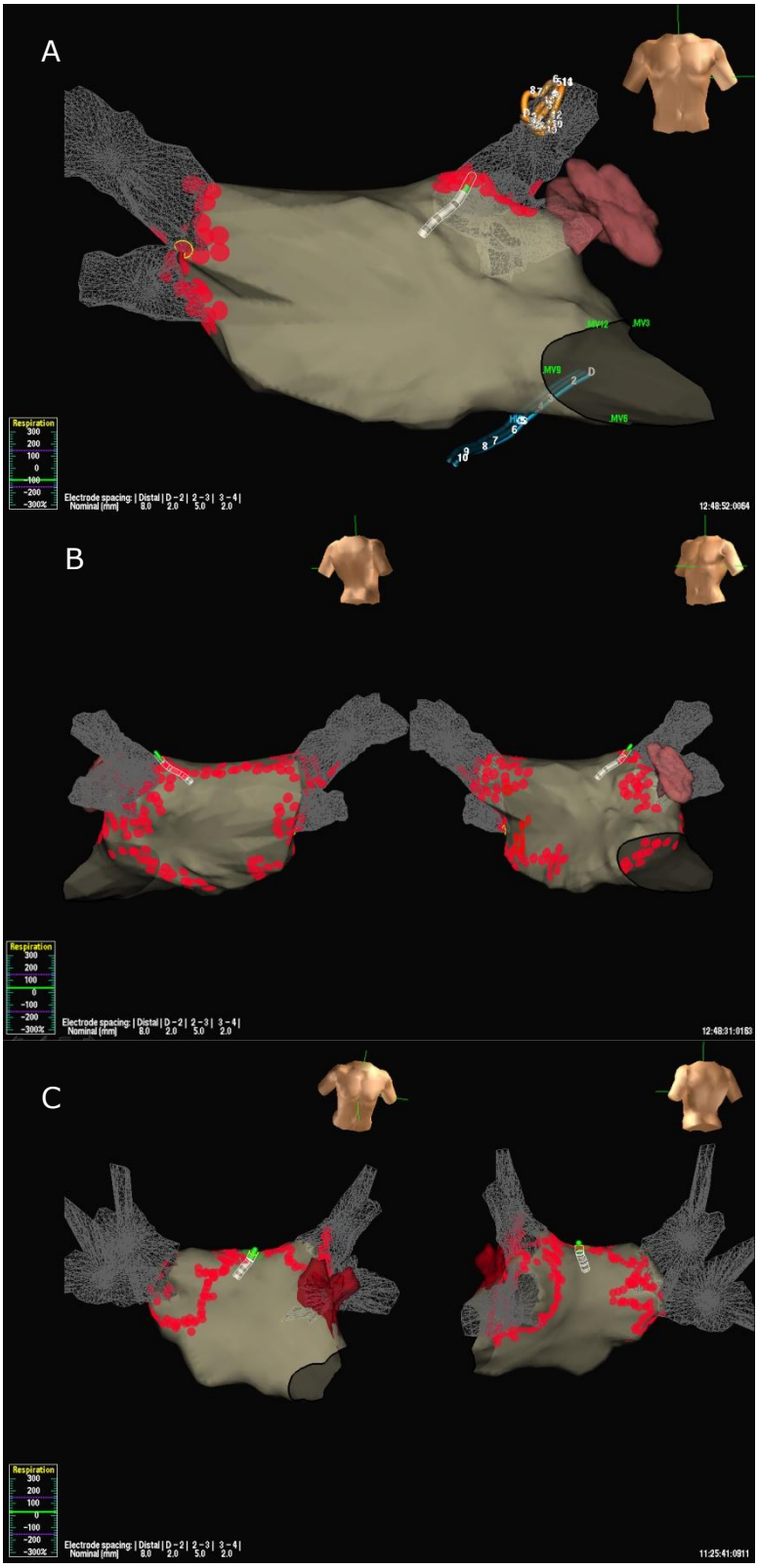
The aim of our study was to examine the frequency spectrum of AF at the start and also at the end of catheter ablation to assess the impact of any changes on follow-up outcomes.

4.2 Methods

Patients undergoing catheter ablation for persistent AF were included in the study. All anti-arrhythmic medication were stopped for at least 5 half-lives prior to the procedure.

AF ablation was carried out using bilateral femoral venous access. Under fluoroscopic guidance, a deflectable decapolar catheter and a quadripolar catheter were positioned in the coronary sinus and His position respectively. A single transseptal puncture technique was utilised in all cases to gain access to the left atrium with the use of non-steerable transseptal sheath (LAMP 90, St Jude Medical). Following this, a deflectable, variable loop circular pulmonary vein mapping catheter (Inquiry Optima, St Jude Medical) and a 4mm deflectable, irrigated tip ablation catheter (Thermocool, Biosense Webster) were advanced into the left atrium with subsequent creation of left atrial geometry using a 3-dimensional navigation system (Ensite NavX™, St Jude Medical). Ablation strategy consisted of either 1) segmental ostial pulmonary vein isolation (PVI) alone, 2) segmental ostial PVI with linear ablation (roof and mitral valve isthmus), followed by complex fractionated electrogram (CFE) ablation or 3) wide area circumferential ablation (WACA) followed by linear ablation (roof and mitral valve isthmus) (Figure 4.1). No isolation of the coronary sinus or ablation within the coronary sinus was carried out in any of the patients. Further mapping and ablation was carried for any atrial tachycardia/flutter noted during the procedure. For those who remained in AF or resistant atrial tachycardia/flutter at the end of the ablation procedure, DC cardioversion was carried out to restore sinus rhythm. The patients were recruited sequentially over a period of time when the ablation procedure for persistent AF was evolving, hence the reason for including the 3 different strategies above.

Figure 4.1: Ablation strategies used in the study



Examples of lesions created during A) Segmental ostial pulmonary vein isolation, B) Segmental ostial pulmonary vein isolation + linear ablation and CFE ablation. C) Wide area circumferential ablation + linear ablation

Intra-cardiac electrograms were recorded on a 30-250Hz filter with sampling frequency of 1 kHz (Labsystem Pro, Bard Electrophysiology, Lowell, MA, USA). Spectral analysis was carried out on bipolar AF electrograms recorded from the decapolar catheter in the coronary sinus. All electrograms with adequate signal to noise ratio (signal:noise amplitude ratio of > 2) on the decapolar catheter were analysed (over a period of 10s) at the start of the procedure and also at the end of the procedure after the intended ablation strategy as detailed above had been carried out satisfactorily. In those who converted to sinus rhythm or atrial flutter during ablation, electrograms just before rhythm change were analysed and compared to that at the start of the procedure.

For the purpose of the study, Fast Fourier Transform (FFT) was carried out on AF electrograms with a spectral resolution of 0.24Hz (4096 points), after processing with a Hamming window. A 1024 point sliding window was used to give the mean DF and OI of the signal. The DF and OI of all suitable coronary sinus electrograms across all 5 bipoles were averaged for each patient and used for analysis.

Follow-up assessment of arrhythmia recurrence was carried out by clinical review of symptoms and ECG monitoring at 4 and 12 months, including documentation of any medical advice sought for arrhythmia related symptoms outside of the scheduled follow-up intervals. 24 hour ambulatory ECG monitoring at 6 months post-procedure was also carried out. Any ECG documentation of sustained AF (lasting > 30s) during the 12 month follow-up period is considered to be indicative of AF recurrence if it occurred beyond a 3 month blanking period post-procedure. In the absence of any contraindications, patients were prescribed beta-blockers post-ablation if there were other indications such as co-existing hypertension or ischaemic heart disease. More

specific anti-arrhythmic drugs (including flecainide, propafenone and amiodarone) were prescribed only if there was evidence of early (< 3 months) or post-blanking period (> 3 months) recurrence of AF, or if patients were still aware of symptoms of palpitations (but with no ECG evidence of AF).

All continuous variables are expressed as mean +/- standard error of mean. Normally distributed data were analysed using paired and unpaired Student's t-test as appropriate. Non-parametric data were analysed by Mann Whitney or Wilcoxon signed rank test. One way ANOVA was used to compare more than 2 means for parametric data and Kruskal-Wallis test was used for non-parametric data. Categorical data were analyzed using Chi squared or Fisher's exact test. Logistic regression was carried out to identify independent predictors of freedom from AF on follow-up and receiver operator characteristic analysis was used to evaluate the relevant parameter. A p value of < 0.05 is considered to be statistically significant.

4.3 Results

A total of 30 consecutive patients having catheter ablation for persistent AF were included in the study. Ablation strategy included segmental ostial PVI alone (9), in combination with linear ablation and CFE ablation (13) and WACA with linear ablation (8). AF termination during ablation occurred in 6 (20%) patients. Out of these 6 patients, 2 (7%) patients reverted directly to sinus rhythm and 4 (13%) converted to atrial flutter during ablation.

14 (47%) patients had recurrence of AF beyond a 3 month blanking period and 16 (53%) were free from AF after mean follow-up of 12 ± 1 months. No significant difference in anti-arrhythmic drug usage was noted between the 2 groups. Out of the

16 patients free of AF, 4 developed atrial flutter/atrial tachycardia during the follow-up period.

Across the entire group of patients, DF reduced from 6.5 ± 0.2 Hz to 5.6 ± 0.2 Hz ($p < 0.0001$), while OI increased from 0.26 ± 0.01 to 0.30 ± 0.02 ($p = 0.03$) post ablation. Comparing patients free from AF with those with AF recurrence, absolute change in OI was 0.09 ± 0.02 vs -0.02 ± 0.02 ($p < 0.0001$), and relative change in OI was 40 ± 7 vs -5 ± 7% ($p = 0.0001$) respectively. No significant differences in the change in DF were noted on comparing both groups. There was a trend suggesting that more extensive ablation (i.e. PVI + linear + CFE, WACA + linear ablation, longer ablation time) and termination of AF with ablation were associated with freedom from AF. The summary of the patient characteristics between the 2 groups can be found in Table 4.1. Examples of electrograms used for analysis can be found in Figure 4.2.

Table 4.1 Characteristics of patients with and without recurrence of AF

	Recurrence of AF (n=14)	No recurrence of AF (n=16)	P value
Age	56 ± 2	54 ± 2	P = 0.48
Sex	14 Male (100%)	14 Male (88%)	P= 0.49
Hypertension	6 (43%)	8 (50%)	P = 0.73
Ejection Fraction (%)	48 ± 2	51 ± 2	P = 0.39
Left atrial size (mm)	47 ± 2	44 ± 2	P= 0.18
Ablation Strategy:			
- PVI Alone (n=9)	7 (50%)	2 (13%)	P = 0.07
- PVI + lines + CFE (n=13)	5 (36%)	8 (50%)	
- WACA + lines (n=8)	2 (14%)	6 (38%)	
Total ablation times (s)	2763 ± 281	3305 ± 278	P = 0.18
AF termination during ablation	1 (7%)	5 (31%)	P = 0.18
On Beta-Blockers*	8 (57%)	7 (44%)	P = 0.72
On Anti-Arrhythmic drugs* (other than Beta-Blockers)	7 (50%)	6 (38%)	P = 0.71
Follow-up (mths)	11 ± 1	12 ± 1	P = 0.82

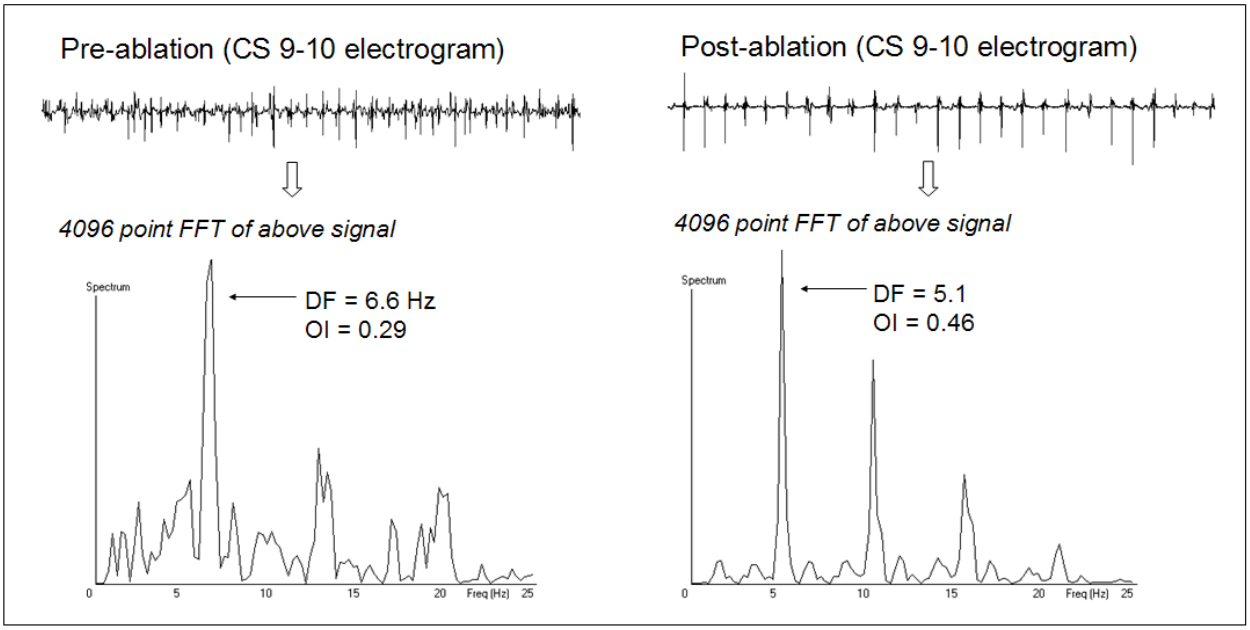
DF pre-ablation (Hz)	6.5 ± 0.2	6.6 ± 0.2	P = 0.76
DF post-ablation (Hz)	5.6 ± 0.2	5.6 ± 0.2	P = 0.87
Absolute change (Hz)	-0.9 ±0.1	-0.9 ± 0.2	P = 0.86
% Change in DF	-14 ± 2	-14 ± 3	P = 0.96
OI pre-ablation	0.28 ± 0.02	0.24 ± 0.02	P = 0.24
OI post-ablation	0.25 ± 0.02	0.34 ± 0.03	P = 0.03
Absolute change	-0.02 ± 0.02	0.09 ± 0.02	P < 0.0001
% Change in OI	-5 ± 7	40 ± 7	P = 0.0001

*At most recent follow-up review

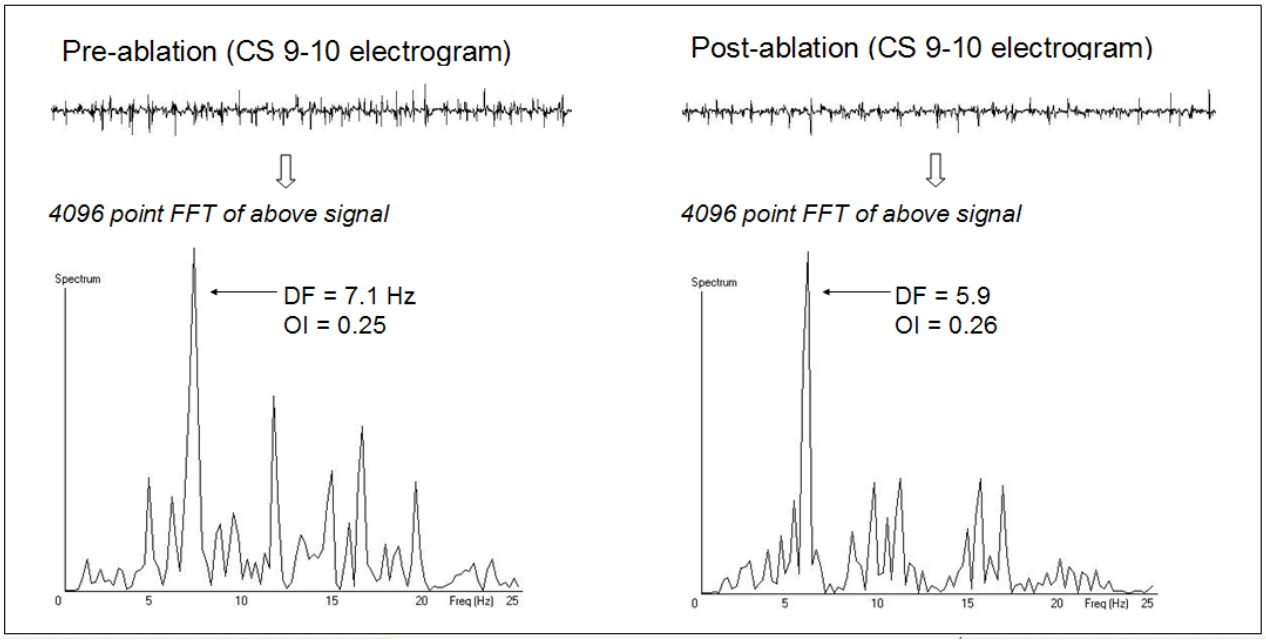
PVI = pulmonary vein isolation (segmental ostial), CFE = complex fractionated electrograms, WACA = wide area circumferential ablation, DF = dominant frequency, OI = organization index

Figure 4.2: Example of intracardiac electrograms and FFT

Patient A



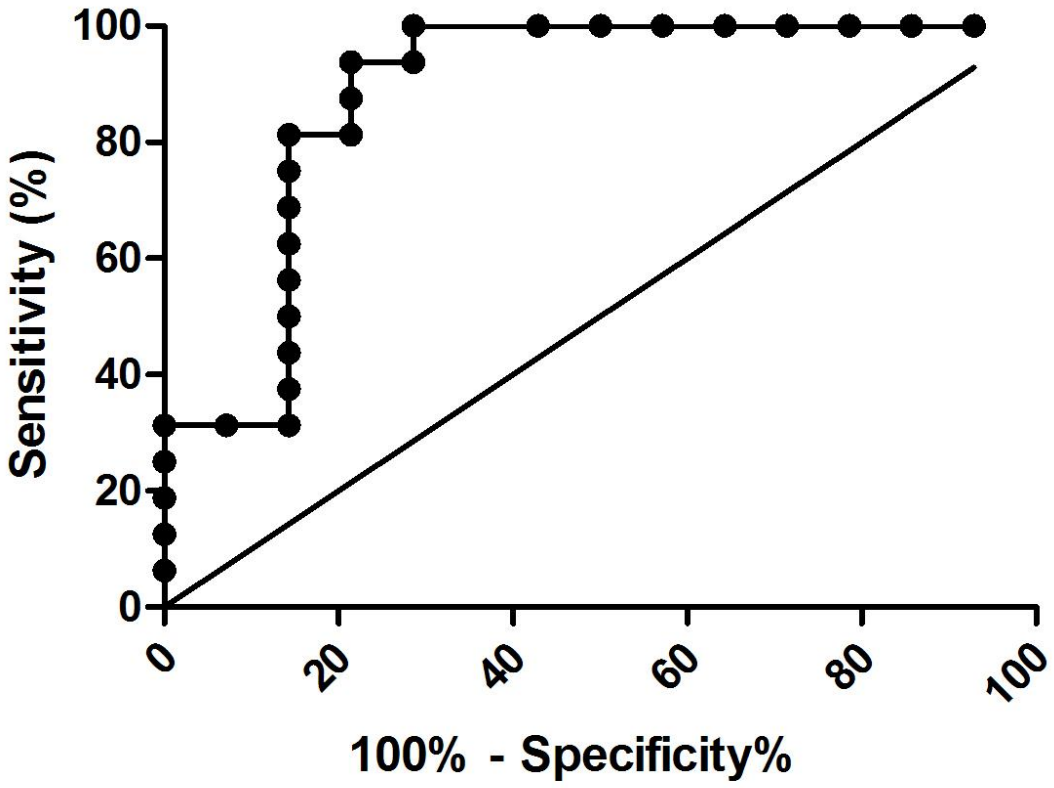
Patient B



Example of intra-procedure CS electrgrams of 2 patients pre and post PVI + linear + CFE ablation. Both were DC cardioverted to sinus rhythm at the end of the procedure. In Patient A (top panel), who stayed in sinus rhythm at 12 months, a reduction in DF and increase in OI were seen. In Patient B (lower panel), who had recurrence of AF at 4 months, a reduction in DF with only minor increase in OI were seen.

Logistic regression identified absolute increase in OI (p = 0.006) and relative increase in OI (p = 0.005) to be independently associated with freedom from AF. OI post ablation showed only borderline association with freedom from AF (p = 0.05). Receiver operator characteristic analysis indicate that a relative increase in OI of > 16% has a 81% sensitivity and 86% specificity for identifying those who will remain free of AF (see Figure 4.3).

Figure 4.3: Receiver operator characteristic curve of percent change in OI.



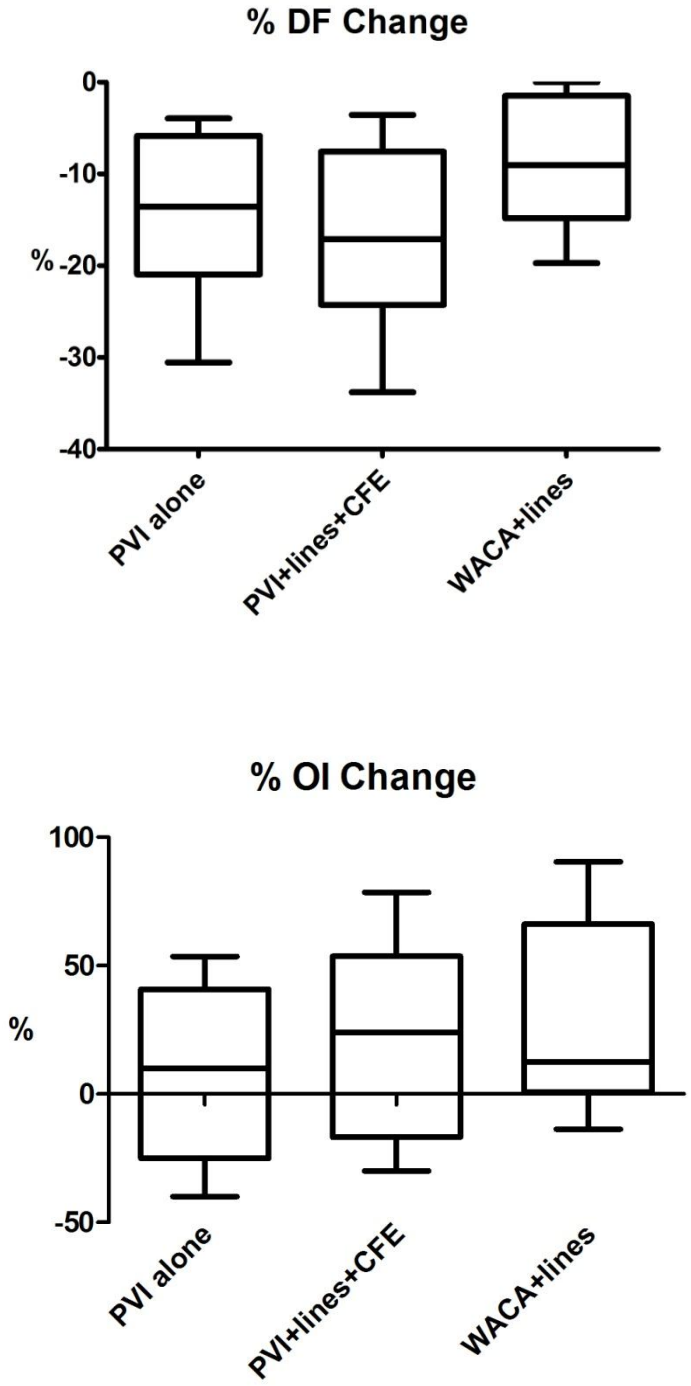
An increase in OI of >16% is associated with 81% sensitivity and 86% specificity of predicting freedom from AF after ablation. Area under curve = 0.88 (p = 0.0003).

No statistically significant differences were noted in the patient characteristics or changes in DF and OI when comparing PVI alone, PVI + linear + CFE ablation and WACA + linear ablation. (See Table 4.2 and Figure 4.4)

Table 4.2: Comparison between PVI alone, PVI+lines+CFE and WACA+lines

	PVI alone (n=9)	PVI+lines+CFE (n=13)	WACA+lines (n=8)	P Value
Age (years)	59 ± 2	54 ± 2	51 ± 3	0.17
HBP (no.)	4 (44%)	6 (46%)	4 (50%)	0.97
LA size (mm)	47 ± 3	44 ± 2	45 ± 2	0.64
EF (%)	50 ± 2	51 ± 2	47 ± 3	0.56
DF pre-ablation (Hz)	6.5 ± 0.2	6.6 ± 0.3	6.4 ± 0.3	0.76
DF-post ablation (Hz)	5.6 ± 0.2	5.5 ± 0.3	5.8 ± 0.3	0.42
% Change in DF	-14 ± 3	-16 ± 3	- 9 ± 2	0.29
OI pre ablation	0.27 ± 0.03	0.27 ± 0.02	0.23 ± 0.02	0.56
OI post-ablation	0.28 ± 0.03	0.32 ± 0.04	0.28 ± 0.02	0.77
% Change in OI	9 ± 11	21 ± 10	27 ± 13	0.74
Follow-up (mths)	14 ± 2	10 ± 2	11 ± 1	0.19

Figure 4.4: Comparison of %change in DF and OI



Box plots comparing percent change in DF (top panel) and OI (bottom panel) between the 3 ablation strategies. No significant differences were noted (p = 0.21 and 0.58 respectively.)

4.4 Discussion

Using spectral analysis, our study has shown that catheter ablation using the above ablation strategies result in reduction in DF in coronary sinus electrograms. The majority of patients remained in AF at the end of catheter ablation and the only predictor for freedom from AF on medium term follow-up was an increase in OI as a result of the ablation procedure. The OI may therefore play a role as a guide in determining the extent of ablation needed.

4.4.1 Dominant Frequency and Outcomes

Several studies have previously looked into the effect of ablation on spectral characteristics of AF and its significance. A study by Lemola et al[182] demonstrated a significant reduction in DF measured from coronary sinus electrograms as well as from different sites within the left atrium after catheter ablation of persistent AF. The same study also found that a greater reduction in DF after ablation using an electrogram-guided approach, was seen in patients who remained free of AF on follow-up. In a separate study, it was found that a critical decrease in DF by ≥ 11% was associated with maintenance of sinus rhythm after ablation for persistent AF.[183] This observation was not reflected in our study, where we found mean decrease in DF to be 14% in both AF recurrence and no AF recurrence groups. It is however, difficult to draw direct conclusions from this as the authors did not assess OI in their cohort of patients and employed a different ablation strategy.

Using real-time mapping and ablation of maximal DF sites in conjunction with circumferential pulmonary vein islolation, Atienza et al[181] found that ablation reduced DF in the left atrium, coronary sinus and right atrium. A proportion of the maximal DF

sites were not ablated due to technical and also safety reasons and on analysis of follow-up results, ablation of maximal DF sites was found to be an independent predictor of freedom from AF

.

4.4.2 Complexity/Organisation of Electrograms and Outcomes

Researchers have also tried to characterize and classify the complexity of AF electrograms to establish its relevance to AF ablation. Nademanee et al[171] showed highly favourable outcomes using an ablation strategy of targeting complex fractionated AF electrograms. Ablation at atrial sites containing a greater percentage of continuous electrical activity or a temporal activation gradient have also been found to be associated with slowing or termination of persistent AF.[254]

Yoshida et al[250] carried out time domain analysis (using complexity and fractionation index) as well as frequency domain analysis (using DF) to study AF electrograms in the coronary sinus before and after ablation. In patients with persistent AF, no significant link was seen in the time domain parameters when comparing patients who had AF recurrence with those who remained free of AF on follow-up. However, the authors found that percentage decrease in DF was greater in those who remained free of AF.

The significance of OI in the maintenance of AF and its response to ablation was first evaluated by Takahashi et al.[229] In this study, it was found that a higher OI as measured from coronary sinus electrograms was associated with greater likelihood of termination of AF with ablation, and that a progressive increase in OI is seen with isolation of each pulmonary vein, prior to AF termination. The authors concluded that

the OI may be representative of the number of driving sources responsible for maintaining AF. In another study, Bencsik et al[176] also showed a significant reduction in DF and an increase in organisation (expressed as regularity index) at the end of ablation of persistent AF using a stepwise approach.

Using spectral analysis of canine AF, it has also been shown that a greater OI increased the efficacy of burst pace termination of AF[221] and also electrical cardioversion[220] of AF. In addition, we have also previously shown that anti-arrhythmic medication such as flecainide restores sinus rhythm acutely in human subjects by reducing DF of AF electrograms, but only when an increase in OI is also seen[1], suggesting that a reduction in the number of re-entrant circuits or drivers of AF is key to termination of AF. Our current study adds further support to this hypothesis, with the findings of an increase in AF organisation, represented by an increase in OI post-ablation, being associated with a lower likelihood of AF recurrence.

4.4.3 Other electrographic features and termination of AF as a predictor of outcome

Reliable predictors of outcome following AF ablation remain a topic of ongoing research. A longer surface ECG[255], atrial appendage or coronary sinus AF cycle length[236, 253], smaller left atrial size[158, 253, 256], and shorter duration of AF[253] have been proposed to be associated with termination of AF during ablation. It has also been suggested that successful termination of persistent AF during ablation is associated with a lower risk of subsequent AF recurrence.[183, 253, 257] This however, was not demonstrated to be the case in a recently published study[251] involving over 300 patients, which found that AF termination during ablation could predict mode of arrhythmia recurrence, but did not seem to be associated with long term

maintenance of sinus rhythm. Moreover, rates of persistent AF termination with ablation are highly variable in published literature, ranging from 13% up to 91%,[194, 250, 258, 259] depending on technique used, and with the definition quite often including conversion to atrial flutter/atrial tachycardia together with direct conversion to sinus rhythm. In our study, only a small proportion (20%) of the patients had termination of AF during ablation (2 to sinus rhythm and 4 to atrial flutter). There was a trend suggesting termination to be associated with freedom from AF on follow-up, although this did not reach statistical significance.

4.5 Relevance of Findings

Clear end points to guide the extent of ablation needed in patients with persistent AF are lacking. While termination of AF during ablation (particularly to sinus rhythm) is conventionally seen as a desirable result, this only happens in a select proportion of patients with persistent AF during the first ablation procedure, and there continues to be conflicting data about its correlation with freedom from AF on follow-up. Our study supports published literature showing that catheter ablation reduces the DF of AF within the coronary sinus. Although we found no relationship between the extent of DF reduction and outcomes, we have demonstrated that an increase in OI during ablation can be a predictor of freedom from AF in the medium term. In patients who remain in AF despite extensive catheter ablation for persistent AF, the OI can therefore be a useful intra-procedural tool to help determine the extent of ablation that is required. Further studies involving larger number of patients are needed to fully evaluate the utility of OI and test its reliability as a predictor of outcome following catheter ablation of persistent AF.

4.6 Limitations

Analysis of electrograms recorded from the coronary sinus catheter allows for reliable comparison of pre and post-ablation data as it has the advantage of being in a stable position throughout the procedure. However, the limitation of this is that the data applies only to changes in the coronary sinus and are not fully representative of the global spatial characteristics of AF in the rest of the left atrium. Nevertheless, the technique of analysing coronary sinus electrograms is widely described in published literature, which allows for direct comparison between studies.

Monitoring of AF recurrence was limited only to scheduled clinical review over a 1 year period with spot ECG assessment and a 24 hour ambulatory ECG monitor. It is therefore not possible to exclude episodes of AF recurrence which were not detected with the above methods. Any late recurrence of AF beyond or study period was not assessed.

The study was not powered to compare different ablation strategies as the main aim was to assess if spectral characteristics of AF could predict outcome of ablation. Only pulmonary vein isolation was carried out in some of the patients as they were recruited during our early experience of ablating persistent AF.

4.7 Conclusion

Increase in OI of AF electrograms following catheter ablation of persistent AF appears to be associated with medium term freedom from AF. OI could be a useful guide to determine the extent of ablation required.

Chapter 5
Catheter ablation for persistent AF: Changes in Autonomic Tone and Frequency Spectrum mirror that of vagal blockade

5.1 Introduction

The autonomic nervous system is strongly implicated in the eletrophysiology of atrial fibrillation (AF), and both sympathetic and vagal influences are thought to play significant roles in the initiation of AF.[103, 107, 108, 118]

Studies on both animal as well as human hearts have demonstrated that autonomic tissue in the atria can be identified by means of high frequency stimulation,[118, 122, 260] and denervation of these by ablation in addition to standard isolation of pulmonary veins can potentially lead to improved outcomes following AF ablation. [122, 123]

Most studies employed autonomic testing while in sinus rhythm and hence the majority of patients studied are those with Paroxysmal AF. Assessment of cardiac autonomic tone while in persistent AF is not clearly defined and hence limited information is available with regard to autonomic influences in persistent AF. It is also unclear whether ablation using standard techniques not deliberately targeting autonomic tissue results in any modulation of the autonomic tone and its influence in persistent AF.

The aim of this study was to assess for changes in autonomic measures and spectral characteristics of persistent AF in response to atropine and radiofrequency catheter ablation.

5.2 Methods

Patients who were in persistent AF at the start of AF ablation procedures were studied. All anti-arrhythmic medication were stopped for at least 5 half-lives prior to the procedure. The procedure was carried out using bilateral femoral venous access. Under fluoroscopic guidance, a deflectable decapolar catheter and a quadripolar catheter were positioned in the coronary sinus and His position respectively. Surface ECG and intracardiac electrograms were recorded continuously on a 0.01 – 25 Hz and 30 - 250Hz acquisition filter respectively, with a sampling frequency of 1 kHz (Labsystem Pro, Bard Electrophysiology, Lowell, MA, USA). All coronary sinus electrograms with adequate signal to noise ratio (amplitude ratio of > 2) on all 5 bipoles of the decapolar catheter were analysed and mean values were taken. RR intervals were measured by a built-in automated detection function, detecting and measuring intervals between R wave peaks of QRS complexes from the 12-lead surface ECG (lead V6 position). These were manually checked and corrected if there were any errors.

The study patients are divided into 2 different groups, with Group 1 consisting of patients who had electrogram analysis carried out before and after atropine administration, and Group 2 consisting of patients who had the same analysis carried out before and after catheter ablation of AF.

5.2.1 Group 1 patients

Recording of the study was commenced once the ECG leads and intracardiac catheters were in position. The first 5 minutes of the recording was used for observation and to allow for a steady state to be achieved. Intravenous atropine (1-2

mg) was then given to achieve at least 15% rise in ventricular rate and recorded at the end of 5 minutes of steady and sustained heart rate increase, so as to ensure adequate evidence of vagal blockade. Once this data was collected, the ablation procedure could proceed as planned by the operator. Using FFT off-line, the mean DF and OI of coronary sinus electrograms from the decapolar catheter were obtained, over 30s at steady state, just before and 5 minutes after atropine administration. RR intervals from surface ECG were also measured at the same time points (to obtain mean RR intervals, SD of RR intervals and 5th percentile of RR intervals) but with duration of analysis extending to 1 full minute. The 5th percentile of the collected RR intervals is thought to approximate AV nodal functional refractory period[261, 262] and could therefore also be used as an indicator of autonomic tone.

5.2.2 Spectral analysis

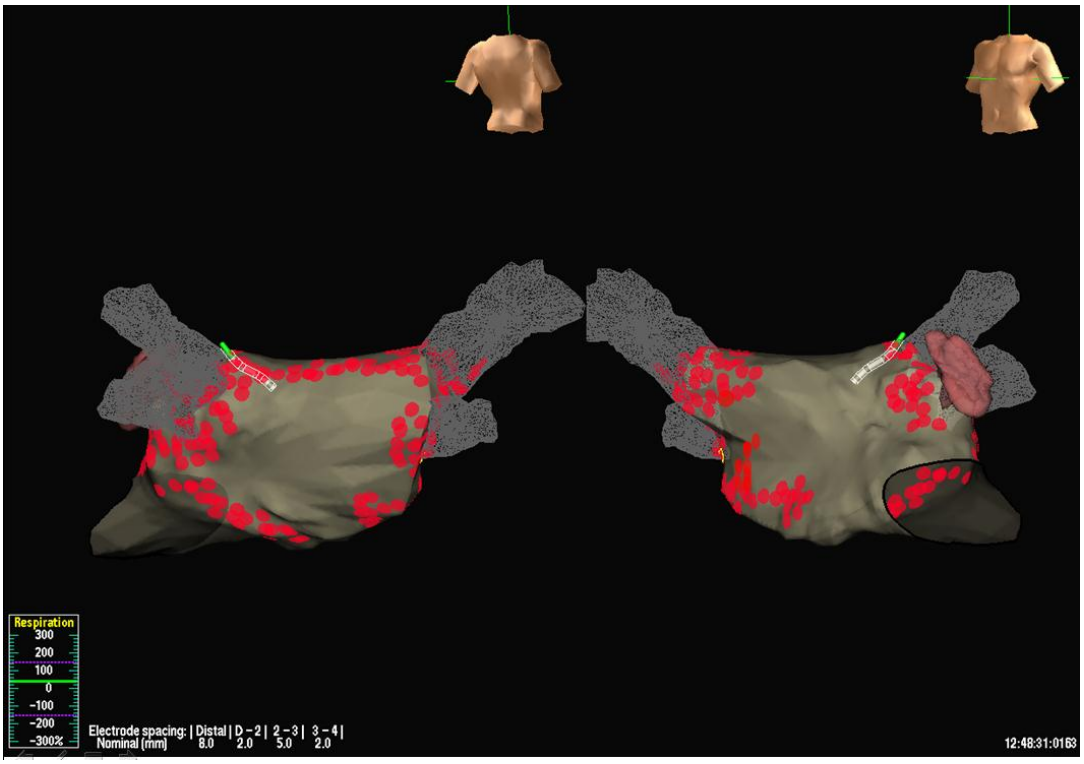
For the purpose of the study, FFT was carried out on AF electrograms with a spectral resolution of 0.24Hz (4096 points), after processing with a Hamming window. A 1024 point sliding window was used to give the mean DF and mean OI of the signal. The DF and OI of all suitable coronary sinus electrograms across all 5 bipoles were averaged for each patient and used for analysis.

5.2.3 Group 2 patients

In a separate group of patients, the same coronary sinus electrogram and RR interval analysis were carried out at the start and at the end of the AF ablation procedure without administration of any atropine. In the event of AF termination during ablation, the electrograms were analysed just before rhythm change. In brief, the ablation procedure was carried out using a single transseptal puncture technique with the use

of a non-steerable transseptal sheath (LAMP 90, St Jude Medical). A deflectable, variable loop circular pulmonary vein mapping catheter (Inquiry Optima, St Jude Medical) and a 4mm deflectable, irrigated tip radiofrequency ablation catheter (Thermocool, Biosense Webster) were used to create left atrial geometry with the assistance of a 3-dimensional navigation system (Ensite NavX™, St Jude Medical). Ablation strategy consisted of stepwise setmental ostial pulmonary vein isolation in combination with linear ablation and complex fractionated electrogram ablation, without specifically targeting anatomical sites of ganglionated plexi (Figure 5.1). A positive vagal response to ablation is defined as prolongation of the RR interval in AF by more than 50%[260] within 5 seconds of switching on radiofrequency energy. DC cardioversion was carried out if AF was still present after all intended ablation had been carried out.

Figure 5.1: Ablation strategy consisting of stepwise segmental ostial pulmonary vein isolation, linear ablation (roof and mitral valve isthmus), and complex fractionated electrogram ablation



All continuous variables are expressed as mean +/- standard error of mean. Normally distributed data were analysed using paired and unpaired Student's t-test as appropriate. Non-parametric data were analysed by Mann Whitney or Wilcoxon signed rank test. Categorical data were analyzed using Chi squared or Fisher's exact test. A p value of < 0.05 is considered to be statistically significant.

5.3 Results

A total of 45 consecutive patients undergoing catheter ablation for persistent AF for the first time were included in the study. The first 15 patients were allocated to Group 1 and the remaining 30 patients were allocated to Group 2.

In Group 1 (n = 15) patients, atropine reduced DF from 6.9 ± 0.2 to 6.5 ± 0.1 Hz (p = 0.002) and increased OI from 0.28 ± 0.01 to 0.31 ± 0.02 (p = 0.02). Mean RR interval decreased from 545 ± 20 to 414 ± 25 ms (p < 0.0001), SD of RR intervals decreased from 122 ± 7 to 74 ± 7 ms (p = 0.0002) and 5^{th} percentile of RR intervals decreased from 390 ± 19 to 325 ± 18 ms (p = 0.001).

In Group 2 (n=30) patients, DF reduced from 6.6 ± 0.2 to 5.7 ± 0.2 Hz (p < 0.0001) and OI increased from 0.26 ± 0.01 to 0.30 ± 0.02 (p = 0.01). Mean RR interval decreased from 594 ± 20 to 533 ± 18 ms (p < 0.0001), SD of RR intervals decreased from 134 ± 8 to 108 ± 7 ms (p = 0.003) and 5th percentile of RR intervals decreased from 423 ± 12 to 399 ± 11 ms (p = 0.0007) respectively. 7 (23%) patients converted from AF to atrial tachycardia/atrial flutter during ablation. Only 2 (7%) patients exhibited a positive vagal response during ablation.

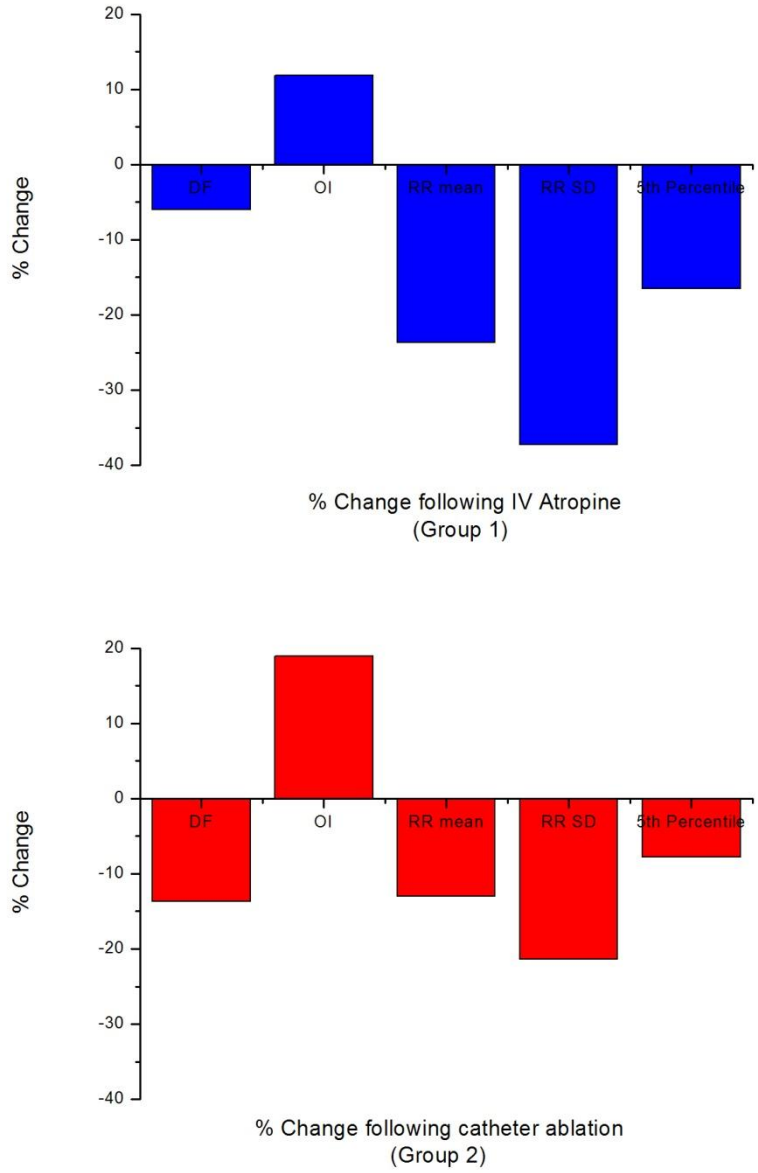
No significant differences in baseline characteristics between the 2 groups were noted. However, a greater reduction in DF was seen with catheter ablation (Group 2 patients) than with atropine (Group 1 patients). Changes in mean RR intervals, SD of RR intervals and 5^{th} percentile of RR intervals were greater with atropine than with catheter ablation. More detailed comparison between the 2 groups can be found in Table 5.1. Similar directional changes in DF, OI, mean RR intervals, SD of RR intervals and 5^{th} percentile of RR intervals were seen in both groups (Figure 5.2). Examples of the spectral changes seen in electrograms with atropine or with ablation are shown in Figure 5.4 and Figure 5.3 respectively.

Table 5.1: Comparison between characteristics of Group 1 (Atropine) and Group 2 (Ablation) patients

	Group 1 (n=15)	Group 2 (n=30)	P value
Age	55 ± 2	54 ± 2	P = 0.67
Sex	14 Male (93%)	27 Male (90%)	P = 1.0
Hypertension	8 (53%)	17 (57%)	P = 1.0
Ejection Fraction (%)	49 ± 2	50 ± 1	P = 0.70
Left atrial size (mm)	47 ± 2	45 ± 1	P = 0.48
DF			
- At baseline (Hz)	6.9 ± 0.2	6.6 ± 0.2	P = 0.13
- After atropine/ablation (Hz)	6.5 ± 0.1	5.7 ±0.2	P = 0.001
- Percent DF change (%)	-6 ± 2	-14 ± 2	P = 0.005
OI			
- At baseline	0.28 ± 0.01	0.26 ± 0.01	P = 0.33
- After atropine/ablation (Hz)	0.31 ± 0.02	0.30 ± 0.02	P = 0.75
- Percent OI change (%)	12 ± 5	19 ± 6	P = 0.47
Mean RR interval			
- At baseline (ms)	545 ± 25	594 ± 20	P = 0.18
- After atropine/ablation (ms)	414 ± 25	533 ± 18	P = 0.0009
- Percent Change (%)	-24 ± 3	-13 ± 2	P = 0.01

SD of RR intervals			
- At baseline (ms)	122 ± 7	134 ± 8	P = 0.36
- After atropine/ablation (ms)	74 ± 7	108 ± 7	P = 0.008
- Percent Change (%)	-37 ± 6	-21 ± 4	P = 0.05
5^{th} percentile of RR intervals			
- At baseline (ms)	390 ± 19	423 ± 12	P = 0.15
- After atropine/ablation (ms)	325 ± 18	399 ± 11	P = 0.001
- Percent Change (%)	-16 ± 3	-8 ± 2	P = 0.01

Figure 5.2: Comparison of changes between Group 1 and 2 patients



Percentage change in Dominant Freqency (DF), Organization Index (OI), RR mean, RR standard deviation (RR SD), and 5th percentile of RR intervals following atropine (top panel) and ablation (lower panel) are shown. Similar directional changes are noted in both groups.

Figure 5.3: Example of change in DF and OI in coronary sinus electrogram pre and post atropine injection

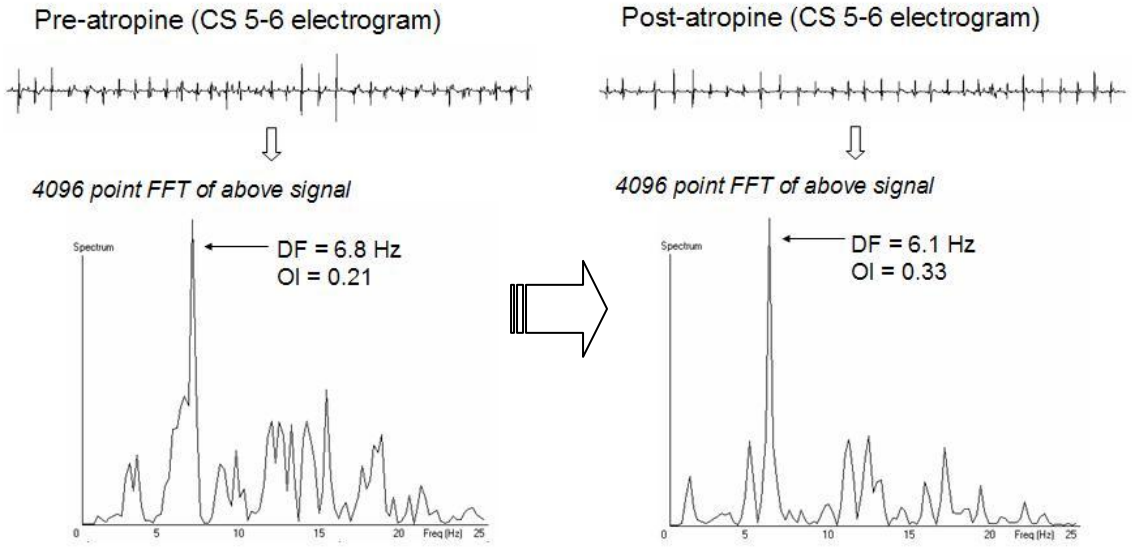
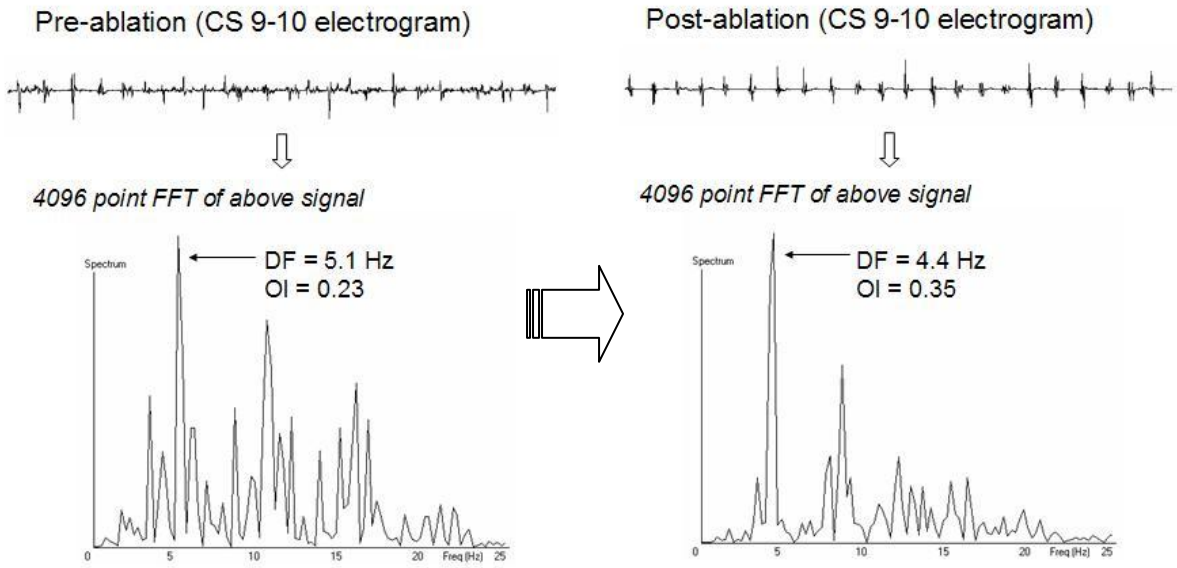


Figure 5.4: Example of change in DF and OI in coronary sinus electrogram pre and post ablation



5.4 Discussion

The main findings in this study are:

1) Catheter ablation using a stepwise approach described above reduced DF and increased OI of coronary sinus electrograms, while also affecting autonomic tone measures consistent with a reduction in vagal tone acutely
2) Vagal blockade with atropine also reduced DF and increased OI of coronary sinus electrograms, in addition to its expected autonomic effects
3) The pattern of changes in spectral characteristics of AF and autonomic tone brought about by ablation were similar to that caused by pharmacological vagal blockade with atropine.
4) Ablation appears to a have greater effect on the DF of coronary sinus electrograms while atropine has a greater effect on autonomic tone measures

The above findings provide further evidence highlighting the influence of the autonomic nervous system on the behaviour of human AF. The acute effects of ablation appear to be similar to pharmacological blockade with atropine, suggesting that conventional anatomical and electrogram-guided ablation not specifically aimed at autonomic tissue possibly also result in modulation of the autonomic tone.

In addition, our results also demonstrate that autonomic measures such as the SD of RR intervals and 5^{th} percentile of RR intervals changed as anticipated with atropine, suggesting that these measures could be used to estimate autonomic tone even in the presence of AF.

5.4.1 Autonomic influences on AF

The autonomic nervous system is thought to play a key role in the initiation and maintenance of cardiac arrhythmia. Anatomical studies have demonstrated the presence of clusters of autonomic tissue i.e ganglionated plexi (GP) located at specific sites around the human heart, which include locations near the pulmonary veins, predominantly on the posterior surfaces of the atria.[104, 105]

Coumel et al first described the observation of paroxysmal AF secondary to vagal over-activity, and then went on to propose the existence of vagal and sympathetic forms of AF; the former being prevalent in normal hearts, and the latter occurring in the presence of heart disease.[102, 103]

Schauerte et al[118] demonstrated that high frequency stimulation at pulmonary vein and superior vena cava sites produced rapid ectopic beats leading to induction of AF. These effects were blunted by beta-blockade and abolished by atropine, implying involvement of both sympathetic and parasympathetic pathways. In another study by the same researchers,[115] radiofrequency ablation of local parasympathetic tissue successfully abolished vagally mediated AF in dogs.

Acetylcholine is the main neurotransmitter released by the parasympathetic nervous system. Vagal stimulation or exogenous acetylcholine produces a negative chronotropic effect on the SA node and a negative dromotropic effect on the AV node.[112, 113] In addition, animal studies have shown that vagal stimulation shortens atrial effective refractory period (ERP)[114, 115] and also plays a role in promoting spatial heterogeneity[116], thereby creating factors favouring initiation and maintenance of AF. Atropine is a muscarinic receptor antagonist which blocks the effects of acetylcholine

and hence also that of vagal stimulation. It should therefore oppose the above changes in atrial electrophysiology and make conditions less hospitable for existence of AF. From our study results, it can be hypothesized that DF reduction is a consequence of an increase in atrial ERP and the increase in OI is due to a reduction in the number of re-entrant circuits, brought about by both an increase in atrial ERP and reduction in spatial heterogeneity of refractoriness as a result of vagal blockade with atropine. Reduction in dominant frequency in electrograms recorded from the left atrium and coronary sinus after catheter ablation has previously been demonstrated in several published reports[181-183]. An increase in organization of AF electrograms, as represented by an increase in the OI, can also be seen following ablation[176, 229] and has been shown to be an important factor in the termination of AF.[1, 229]

5.4.2 HRV and assessment of autonomic tone in AF

Heart rate variability (HRV) has been studied extensively as a tool to assess autonomic tone and has been shown to predict prognosis in heart failure[263] and post-myocardial infarction.[264] Assessment for HRV in published literature is almost always carried out in sinus rhythm, using both time and frequency domain parameters,[265] which essentially reflect autonomic influences on the sinoatrial node. The irregular and apparently chaotic atrial and ventricular activation in AF is conventionally considered to be unsuitable for HRV analysis. However, the beat-to-beat variability and frequency of RR intervals in AF is likely to be a product of atrial conduction and activation as well as the conduction properties of the atrioventricular node, both of which are susceptible to autonomic influence. It would therefore be reasonable to think that of some of the HRV indices in AF would at least in part, be reflective of autonomic tone.

Although the assessment of HRV in AF has only been examined in a small number of studies, most have found positive associations between HRV indices and patient outcomes similar to that seen in sinus rhythm.[266, 267] A study using an abbreviated duration of HRV analysis found analogous changes in HRV measures between AF and sinus rhythm patients after pharmacological autonomic blockade, concluding that HRV in AF is related to vagal tone.[268]

In our study, we made use of a simple, abbreviated method of HRV analysis by only examining SD of RR intervals, and also included the 5th percentile of RR intervals as a measure of autonomic tone. The use of the latter is based on the concept that the minimal RR interval during AF approximates the functional refractory period (FRP) of the AV node. Supportive evidence for this was provided by Billette et al[261] who found the 5th percentile of RR intervals to be equal to the FRP of the AV node while studying dog hearts. Using human subjects, Toivonen et al[269] described good correlation between shortest RR intervals and AV nodal FRP. Khand et al[262] also recently described a novel method of assessing autonomic function in AF, based on assessment of hourly 5th percentile of RR intervals over a 24 hour period in a cohort of heart failure patients.

5.4.3 Effects of ablation on parasympathetic tone

Studies looking into autonomic tone using HRV indices have found transient parasympathetic attenuation following catheter ablation for paroxysmal AF, lasting only up to 3 months.[123, 270] In an animal study,[271] vagal denervation by ablation acutely reduced AF inducibility with vagal stimulation, but this effect was no longer present on repeat testing 4 weeks later. Thus, it would appear that while radiofrequency ablation can modify cardiac autonomic tone, it does not seem to have

a lasting effect, casting doubt on whether targeting autonomic tissue will have long term efficacy. Early reports using a strategy of GP ablation together with pulmonary vein antral ablation have shown single procedure success to be around 86% in patients with paroxysmal AF after mean follow-up of 22 months.[184] Other authors have used a purely anatomical GP ablation approach and shown maintenance of sinus rhythm in 71% of patients with paroxysmal AF using implantable recorder for monitoring,[185] although the same technique was not as effective for patients with persistent AF (38% single procedure success rate).[186]

5.5 Relevance of findings

From existing published evidence, it is apparent that the autonomic nervous system plays a very significant role in the pathophysiology of AF at the electrophysiological level. While both sympathetic and parasympathetic arms are thought to participate in this, there probably is a predominance of parasympathetic influence. Our results demonstrate, for the first time, that vagal blockade with atropine results in frequency domain changes in AF electrograms similar to that seen with ablation, although occurring to a lesser extent. At the same time, using a combination of existing and alternative markers of vagal tone, we have also shown a degree of parasympathetic attenuation in persistent AF after catheter ablation which was not deliberately targeted at autonomic tissue. This could be explained by the observation that atrial sites associated with GP often also contain complex fractionated electrograms,[260] and are usually located near pulmonary vein antra, which are typical sites for delivering lesions. Hence the ablation strategy used in our study could have quite conceivably resulted in a degree of parasympathetic denervation without actively seeking to do so.

Our results indicate that catheter ablation using an anatomical and electrogram guided approach results in acute directional changes in autonomic tone and spectral characteristics, similar to that of vagal blockade with atropine, in the setting of persistent AF. It further strengthens the concept that the autonomic nervous system is intimately involved in the mechanism of persistent AF, and that some of the effects seen with ablation could be attributed to parasympathetic attenuation. In addition, the 5^{th} percentile of RR intervals as used in our study, appears to be a useful surrogate of autonomic tone and could be used as a quick guide to determine the extent of autonomic modulation in real-time, during ablation of persistent AF. Current evidence suggest that autonomic changes induced by ablation seem to be only a transient phenomenon. Larger scale studies examining the long-term effects of GP targeted ablation are needed.

5.6 Limitations

Analysis of electrograms recorded from the coronary sinus catheter allows for reliable comparison of pre and post-intervention data as it has the advantage of being in a stable position throughout the procedure. However, the limitation of this is that the data applies only to changes in the coronary sinus and are not fully representative of the global spatial characteristics of AF in the rest of the left atrium.

We did not directly carry out any assessment of the sympathetic nervous system and no sympathetic blockade was carried out either. Hence, it is not possible to exclude sympathetic influences and determine if the effects seen with vagal blockade with atropine were purely due to vagal withdrawal alone.

5.7 Conclusion

Ablation of persistent AF not directly aimed at autonomic tissue results in the same pattern of changes in DF, OI and autonomic tone measures as that observed with pharmacological vagal blockade.

Chapter 6
Regional fractionation and dominant frequency in persistent Atrial Fibrillation: Effects of left atrial ablation and evidence of spatial relationship

6.1 Introduction

Catheter ablation for persistent atrial fibrillation (AF) remains a challenging procedure and success rates vary significantly between different centres[272]. Repeat procedures to treat further arrhythmia are frequently required in a significant proportion of patients.

Addressing AF substrate in addition to triggers is key to achieving maintenance of sinus rhythm with catheter ablation of persistent AF. Potential substrate targets that have been described include autonomic nervous tissue such as ganglionated plexi[122, 123] and sites with complex fractionated electrograms[171] (CFE) and high dominant frequency[178] (DF). It is not known whether the targeting of a specific substrate is superior to another nor is it clear if these substrates may be related in any way.

To gain better understanding of the characteristics of AF substrate, we carried out a study to examine regional DF (frequency domain substrate) and CFE (time domain substrate) characteristics before and after catheter ablation, so as to study the effect of ablation and also to assess if there is any spatial relationship between the 2 parameters.

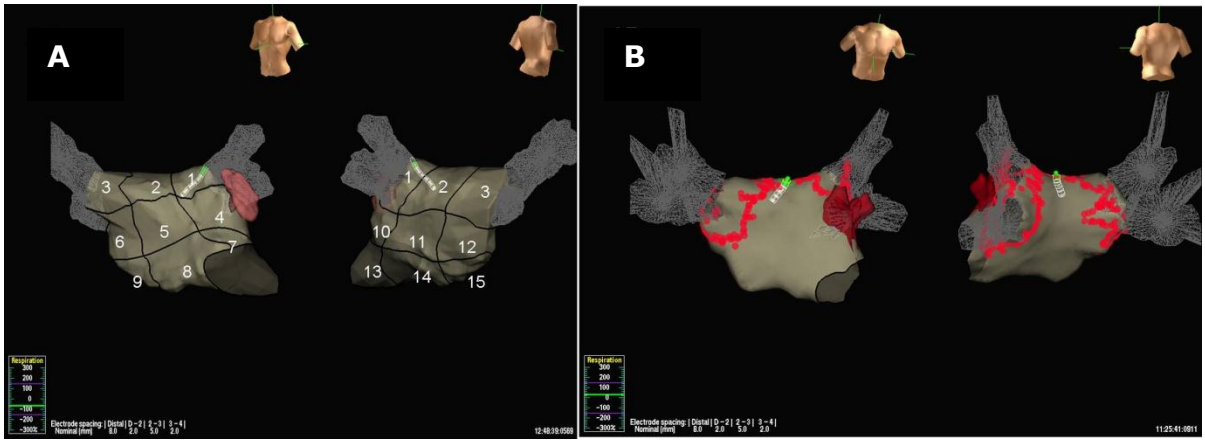
6.2 Methods

A consecutive series of patients undergoing catheter ablation of persistent AF for the first time were studied. Only patients in persistent AF at the start of the procedure were included. All anti-arrhythmic drugs apart from amiodarone were stopped for at least 5 half lives before the procedure. Once bilateral femoral venous access were achieved, a deflectable decapolar catheter and a quadripolar catheter were positioned in the coronary sinus and His position respectively under fluoroscopic guidance. A single transseptal puncture technique was utilised in all cases to gain

access to the left atrium with the use of non-steerable transseptal sheath (LAMP 90, St Jude Medical). Following this, a deflectable, variable loop circular pulmonary vein mapping catheter (Inquiry Optima, St Jude Medical) and a 4mm deflectable, irrigated tip ablation catheter (Thermocool, Biosense Webster) were advanced into the left atrium with subsequent creation of left atrial geometry using a 3-dimensional navigation system (Ensite NavX™, St Jude Medical).

Using the irrigated tip bipolar ablation catheter, point-by-point mapping of the left atrium was carried out according to a 15 segment grid superimposed on the 3-dimensional left atrial geometry (which includes the pulmonary vein antrum/ostia, left atrial roof, septal, lateral, posterior and anterior walls) before and after left atrial ablation. (Figure 6.1, panel A) In all cases, left atrial ablation involved wide area circumferential ablation around ipsilateral pairs of pulmonary veins followed by linear roof ablation (Figure 6.1, panel B). At least 2 samples of electrograms were collected at each segment and the trace with the best quality used for analysis. Once all signals required for the study had been obtained, further ablation strategy was left to the operator's preference.

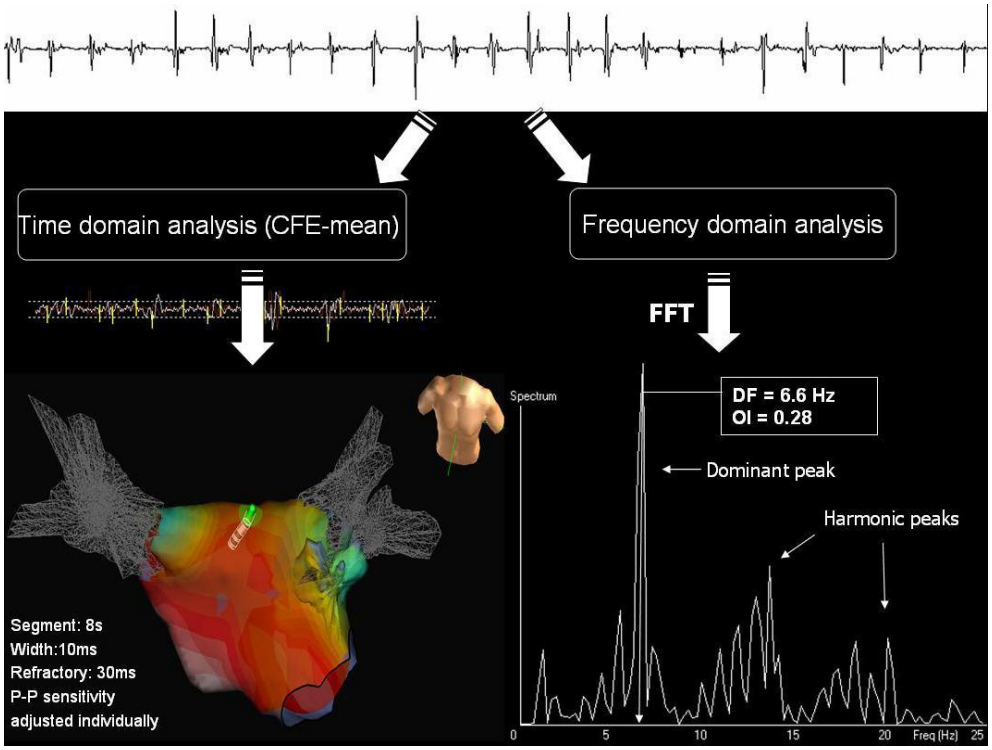
Figure 6.1: Left atrial electrogram mapping and ablation strategy



The 3D geometry of the left atrium is divided into 15 segments for atrial electrogram analysis (A). Typical set of ablation lesions delivered for the purpose of the study, consisting of wide circumferential pulmonary vein and linear roof ablation (B).

Analysis was done on electrograms over 8s of continuous recording at each grid segment: 1) in the time domain as degree of fractionation (expressed as “CFE-mean”) using automated detection software built into the navigation system to create a colour-coded CFE-mean map, and 2) in the frequency domain with Fast Fourier Transform to obtain regional Dominant Frequency (DF) and Organization Index (OI). Previous studies have suggested recording electrograms over at least 5s to increase the accuracy of fractionation and DF assessment[233, 234]. For the purpose of this study, we assessed degree of fractionation as a continuous variable by CFE-mean analysis to include all data between the range of 40 – 300 ms so as to include as wide a range of AF electrograms as possible. The CFE-mean is an average of the intervals between deflections on atrial electrograms which are detected based on a set of predefined criteria. Settings used for the generation of CFE-mean map on the navigation system were as follows: segment of 8s, width 10ms, refractory of 30ms and P-P sensitivity adjusted to adapt to the electrograms for each individual patient (between 0.05 to 0.12 mV). (Figure 6.2)

Figure 6.2: Analysis of data in time and frequency domains



Electrograms were analysed using both time (CFE-mean) and frequency domain analysis [by Fast Fourier Transform (FFT) to obtain dominant frequency (DF) and organization index (OI)]. Settings used to produce the CFE-mean colour map on the left atrial geometry are displayed.

All intra-cardiac electrograms were recorded with a 30-250Hz band pass filter with sampling frequency of 1 kHz and exported for spectral analysis. Fast Fourier Transform (FFT) was carried out with a spectral resolution of 0.24Hz (4096 points), after processing with a Hamming window. A 1024 point sliding window was used to give the mean DF and OI of the signal. (Figure 6.2)

All continuous variables are expressed as mean +/- standard error of the mean. Normally distributed data were analysed using paired and unpaired Student's t-test as appropriate. Non-parametric data were analysed by Mann Whitney or Wilcoxon signed rank test. Categorical data were analyzed using Chi squared or Fisher's exact test. Correlation testing was carried out using Spearman's coefficient.

6.3 Results

A total of 600 wall segments of left atrial electrograms were collected from 23 patients recruited into the study, and 418 of these were used for analysis after discarding signals with poor signal to noise ratio (signal:noise amplitude < 2). Eight patients were on amiodarone at the time of the procedure. Post-ablation data were unavailable in 6 patients who converted to atrial flutter during ablation limited to the study protocol. Baseline characteristics of the study population are summarised in Table 6.1.

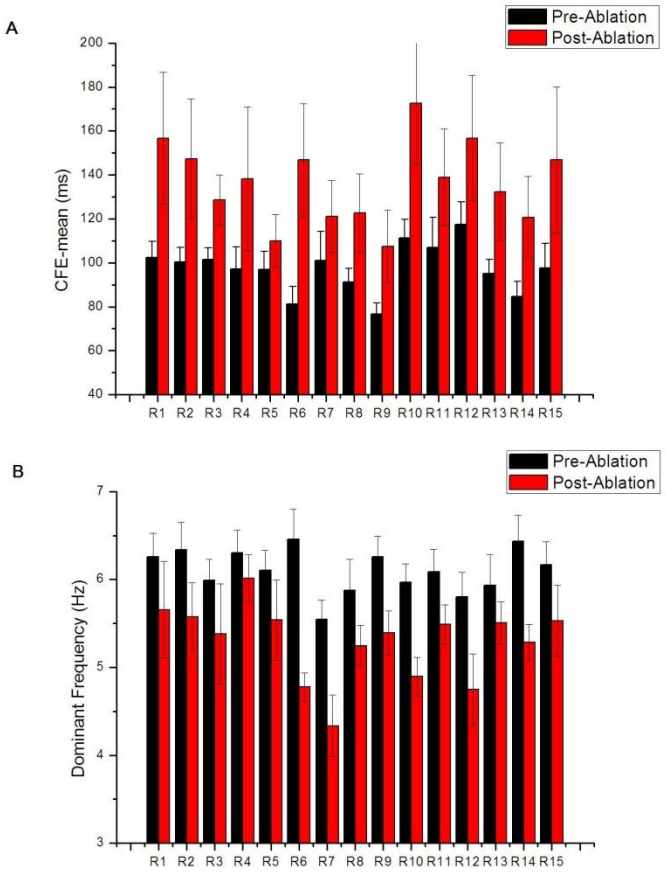
Table 6.1: Patient characteristics of the study population.

Patient Characteristics (n = 23)	
Age (yrs)	57 ± 2
Male	19 (83%)
Duration of AF (months)	56 ± 7
Hypertension	12 (52%)
Diabetes Mellitus	3 (13%)
EF (%)	48 ± 1
LA size (mm)	47 ± 1

Global CFE-mean increased from 100 ± 5 to 147 ± 11 ms (mean ± SEM, p = 0.0003) and DF decreased from 6.1 ± 0.2 to 5.3 ± 0.2 Hz (p = 0.0003) after ablation. Mean OI was unchanged (0.27 ± 0.01 before and 0.26 ± 0.02 after ablation, p = 0.70).

Comparing across all 15 segments of the atrium, CFE-mean increased and DF reduced in all regions post-ablation. The 3 most fractionated sites pre-ablation were localised to the septal and infero-posterior regions of the left atrium (region 6, 9 and 14) which also appeared to contain the higher DF signals. (See Figure 6.3)

Figure 6.3: : Regional CFE-mean and DF



Regional CFE-mean (A) and DF (B) pre and post-ablation across all 15 segments of the left atrium [Region 1 (R1) to Region 15 (R15) as displayed in Figure 6.1]

At sites close to ablation lines (defined as those collected from points located within 5mm of ablation lines) CFE-mean increased from 93 ± 5 to 173 ± 10 ms ($p < 0.0001$), DF decreased from 6.2 ± 0.2 to 5.3 ± 0.2 Hz ($p < 0.0001$), and OI change was 0.26 ± 0.01 to 0.25 ± 0.02 ($p = 0.73$).

At sites distant from ablation lines (defined as those collected from points located more than 5mm from ablation lines) CFE-mean increased from 90 ± 3 to 122 ± 7 ms ($p < 0.0001$), DF decreased from 6.2 ± 0.1 to 5.4 ± 0.1 Hz ($p < 0.0001$) and OI change was 0.27 ± 0.01 to 0.26 ± 0.01 ($p=0.85$). (Table 6.2)

Table 6.2: Change in CFE-mean, DF and OI between points close to (≤ 5mm) and distant (> 5mm) from ablation lines

		Pre-ablation	Post-ablation	P Value
Points close to ablation lines[§]	**CFE-mean (ms)**	93 ± 5*	173 ± 10	<0.0001
	DF (Hz)	6.2 ± 0.2[†]	5.3 ± 0.2	<0.0001
	OI	0.26 ± 0.01[‡]	0.25 ± 0.02	0.73
Points distant from ablation lines[‖]	**CFE-mean (ms)**	90 ± 3*	122 ± 7	<0.0001
	DF (Hz)	6.2 ± 0.1[†]	5.4 ± 0.1	<0.0001
	OI	0.27 ± 0.01[‡]	0.26 ± 0.01	0.85

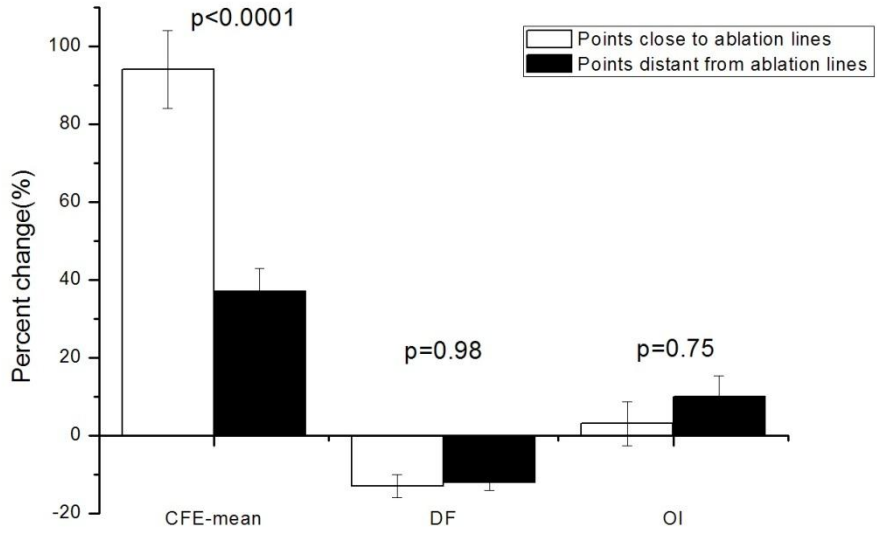
*p = 0.64 [†]p = 0.96 [‡]p = 0.90

[§]Points close to ablation lines (n = 53, typically at atrial segments 1, 2, 3, 6, 10 and 12)

[‖]Points distant from ablation lines (n = 115, typically at atrial segments 4, 5, 7, 8, 9, 11, 13, 14, 15)

Comparing sites close to ablation lines with those distant from ablation lines, percentage change in CFE-mean was 94 ± 10 vs 37 ± 6 % ($p < 0.0001$), DF change was -13 ± 3 vs -12 ± 2 % ($p = 0.98$), OI change was 3 ± 6 vs 10 ± 5 % ($p = 0.75$) respectively. (Figure 6.4)

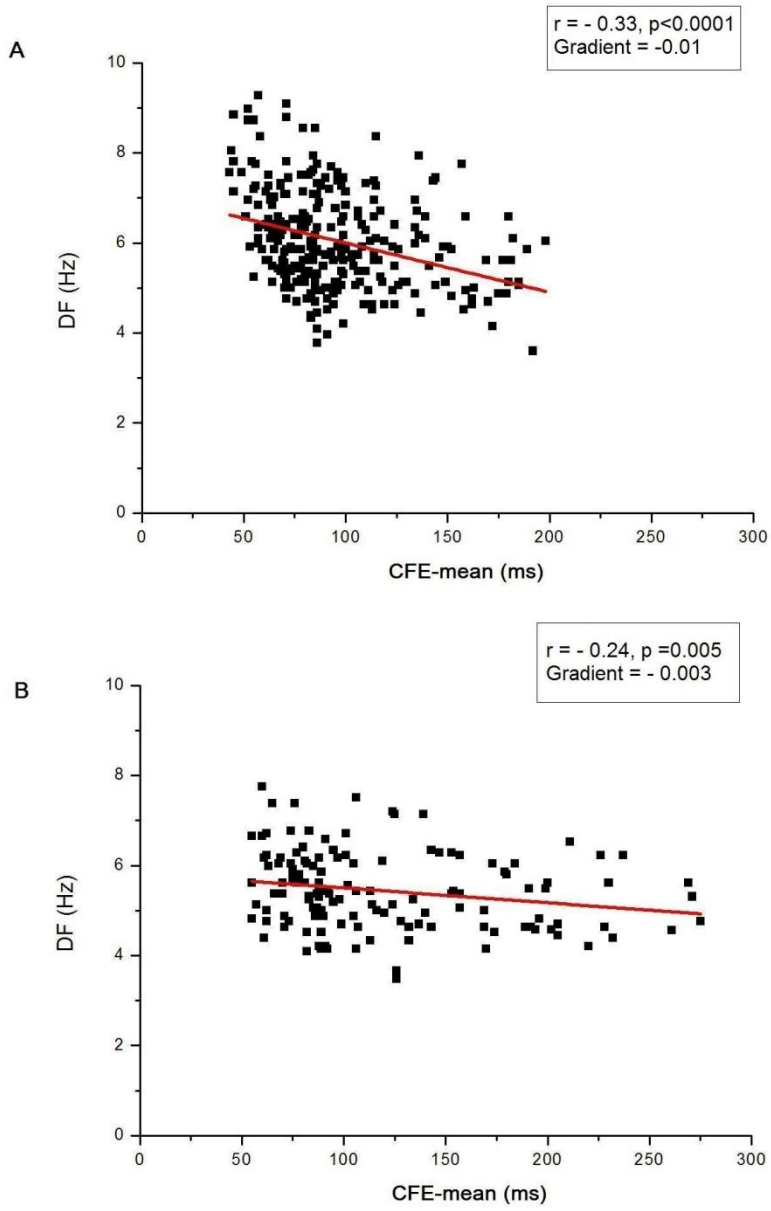
Figure 6.4: Percent change in CFE-mean, DF and OI



Comparison between sites close to (≤ 5mm) and distant (> 5mm) from ablation lines.

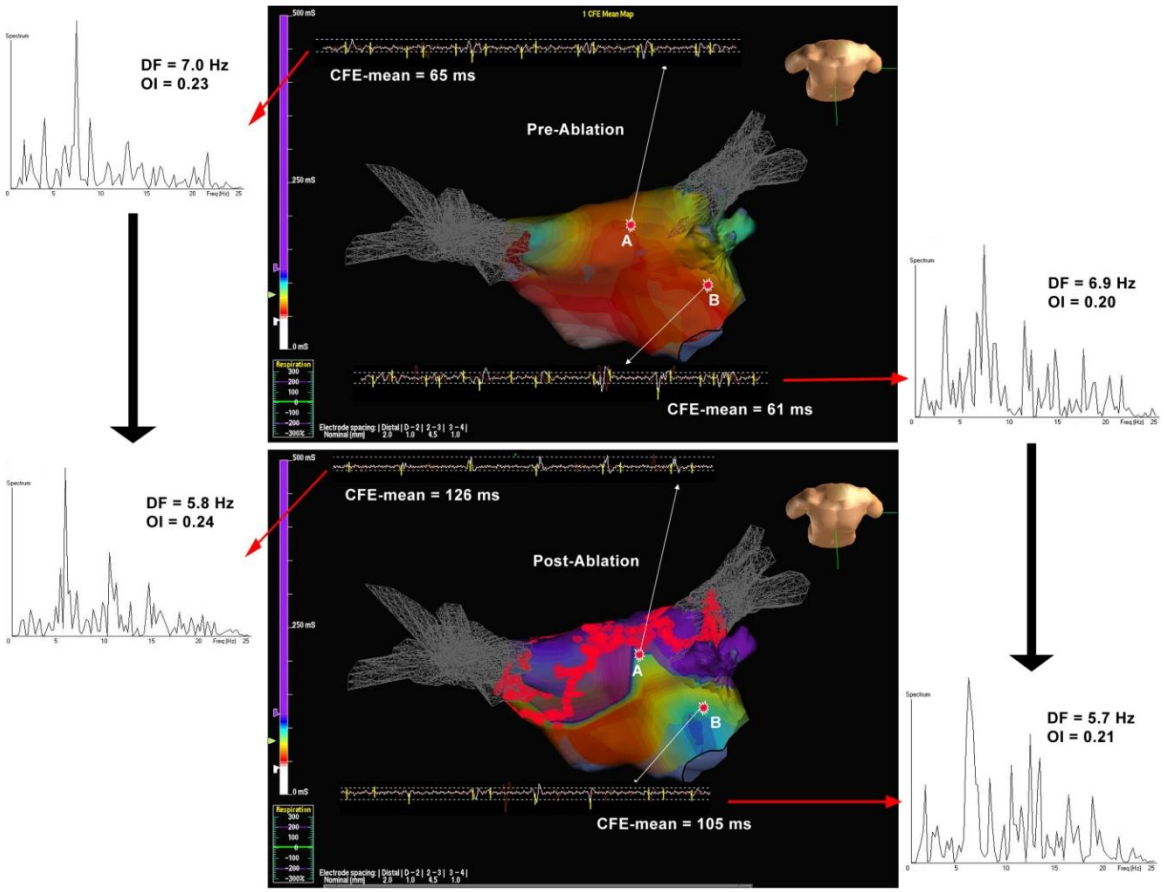
There was moderate correlation between CFE-mean and DF points prior to ablation ($r = -0.33$, $p < 0.0001$, see Figure 6.5, panel A). The strength of this relationship was reduced following left atrial ablation ($r = -0.24$, $p = 0.005$, see Figure 6.5, panel B). Examples of regional CFE-mean and DF change in one of the study patients are shown in Figure 6.6.

Figure 6.5: CFE-mean vs DF plots



Pre and post-ablation plots are shown at top panel (A) and lower panel (B) respectively. There was a moderate degree of correlation which was further weakened post-ablation.

Figure 6.6: Example of the effect of ablation seen in a study patient



Pre (top) and post-ablation (bottom) CFE-mean maps are shown. The yellow markers on the electrograms indicate automatic detection of deflections meeting pre-specified CFE-mean criteria as defined in Figure 2. Electrograms shown are 1s representations of each regional recording lasting 8s, with 4096 point FFT shown adjacent to it. An increase in CFE-mean and reduction in DF is observed at sites close to (point A) and distant from (point B) ablation lines.

6.4 Discussion

We have shown that left atrial ablation using the ablation strategy described above during persistent AF, reduces global left atrial DF and also decreases the degree of fractionation (seen as an increase in CFE-mean). CFE-mean increase was greater at sites close to ablation lines compared to sites distant from ablation lines, although corresponding DF decreased to a similar extent. Regions containing fractionated electrograms seem to harbour high frequency activation, suggesting that CFE and DF sites may be spatially related in some way. However, this relationship appears to be modest, and ablation using an anatomical approach appears to affect these 2 parameters to different extents depending on proximity to the ablation lines, resulting in a weakening of their relationship following ablation. This could imply that though possibly related spatially, CFE and DF are not identical entities and could reflect either entirely different processes, or different components of the same substrate.

6.4.1 Complex fractionated electrograms

Konings et al previously described observation of different characteristics of unipolar electrograms during AF[170] and it was postulated that the different electrogram appearances represented specific patterns of conduction. Fragmented electrograms for example were shown to exist at areas with slow conduction or pivot points, which could represent sites of wavelet reentry facilitating the maintenance of AF. Several studies have also verified using human subjects, that the distribution and location of CFE sites are usually stable and reproducible[273, 274]. Nademanee et al[171] took this concept further by targeting radiofrequency ablation at sites containing CFE within the atria, demonstrating high rates of AF termination with 91% of patients free of arrhythmia and symptoms at 1 year follow-up. However, subsequent studies by other

authors using ablation targeted at CFE[173-175] have shown somewhat conflicting results, which in turn has cast doubt on our understanding of the significance of CFE.

6.4.2 Dominant Frequency Analysis

For many years, thoughts on the mechanism of AF was dominated by the multiple wavelet hypothesis put forward by Moe and Abildskov[68]. In recent years however, there has been more interest in the concept that AF may be more organised and focal, a theory which was previously proposed by Scherf et al[83]. Using experiments carried out on sheep hearts, Skanes et al demonstrated evidence of wave-front spatiotemporal periodicity during AF, with correlation between frequency of the periodic activity and global atrial DF[95]. This, in conjunction with other supportive studies, have led to the proposal that relatively stable rotors giving rise to vortices of electrical waves could be responsible for maintaining AF[98, 99]. Sanders et al[178] carried out spectral analysis and frequency mapping of the atria in human AF and found that ablation at sites with high DF was associated with increased likelihood of terminating AF, hence highlighting the importance of high DF sites.

6.5 Relevance of findings

In the current study, using contact mapping at multiple sites within the left atrium, we have demonstrated the characteristics of regional and global CFE and DF pre- and post-ablation using an anatomically guided technique of wide area circumferential ablation around the pulmonary veins together with left atrial roof ablation. Degree of fractionation (inversely related to CFE-mean) and DF reduced following ablation targeted at isolation of the pulmonary veins, which is in agreement with recently

published work by Lin et al[275]. This trend is consistent at a local as well as a global atrial level, even at sites distant from ablation lines, suggesting dynamic interaction between pulmonary veins and atrium in persistent AF. Our findings also suggest possible spatial relationship between CFE-mean and DF of atrial electrograms, although the correlation is not strong. This is in keeping with results from animal studies carried out by Kalifa et al[276], where the greatest fractionation in sheep AF was seen at the peripheries or outer boundaries of maximal DF sites. This is an observation which was also reported by Stiles et al[277], who carried out high density mapping on a subset of 10 patients with persistent AF. Using activation mapping, the authors found that CFE sites were usually located adjacent to sites with high DF. CFE and DF correlation was also observed in their study but this was restricted to within an individual but not point-by-point basis. Using a larger group of patients in persistent AF, our study has gone further to show that regional CFE-mean and DF appear to be affected to different degrees by ablation, adding support to the notion that the relationship between fractionation and DF is probably not a direct one.

Previous studies have suggested increased likelihood of the presence of CFE at atrial sites innervated by ganglionated plexi[260, 278]. These autonomic tissues are typically located in the antral regions around pulmonary veins and are usually within the path of ablation lesions delivered using standard circumferential pulmonary vein isolation techniques. This could explain the greater change in CFE-mean compared to change in DF seen in our study, as the ablation strategy used would have encompassed locations of ganglionated plexi.

Based on the above discussion, we can hence hypothesize that while sites of fractionation are linked to sites of higher DF, they probably do not exist in the exact same location, and appear to be affected to different degrees by ablation.

6.6 Limitations

Our study involved a relatively small sample of patients and we used sequential contact mapping of the atrium. Higher density simultaneous atrial mapping could provide more detailed information but this would be technically more challenging. Mapping of the right atrium was also not carried out in our study. It has however, been shown by other studies that the left atrium plays a greater role in driving AF as it usually contains sites with higher frequency of activation in persistent AF.[95, 222, 226, 277, 279]

6.7 Conclusion

The role of substrate modification in ablation of persistent AF needs to be more clearly defined and our study has provided further insight into the characteristics of electrogram fractionation and DF in persistent AF before and after limited left atrial ablation. Further studies comparing the long-term outcomes of CFE and DF targeted ablation or a combined ablation strategy of both substrates are needed to help clarify their clinical significance and relevance.

(The study described in this chapter has subsequently been published in a peer-reviewed journal which included additional information obtained from further analysis of the data. Please refer to the Appendix for further details.)

Chapter 7 Synopsis

7.1 Overview of results

The treatment of AF remains a challenging task for all clinicians. The number of adults affected by this common arrhythmia is expected to rise significantly in the coming decades with an aging population, and there is a need for greater understanding of the condition to treat it effectively. Despite intense research into the mechanism of AF, there does not appear to be a clear consensus. Although several conflicting theories exits, it is quite possible that more than one form of AF mechanism exists, and that dynamic interplay between trigger and substrate in the presence of electrical and structural remodelling leads to a final common form of AF over time.

Catheter ablation has offered an additional dimension to the management of AF, but hard endpoints such as morbidity and mortality data are lacking. In particular, ablation of the more long-standing and persistent forms of AF have not yielded universally satisfactory outcomes, highlighting inadequate understanding of the underlying mechanisms.

Using both time and frequency domain analysis, the series of studies that are presented in this thesis have provided further insight into the characteristics of AF. Novel data on the effect of drugs such as flecainide and atropine and impact of ablation on DF and OI as well as measures of autonomic tone have all been examined and presented.

We demonstrated that pharmacological interventions such as that seen with flecainide and atropine, reduced the DF of atrial electrograms while increasing the degree of organization as represented by the OI. We showed that an increase in the

OI appears to be associated with lower AF recurrence rates following ablation, and that it also predicts return to sinus rhythm with flecainide. This agrees well with previous studies demonstrating the use of AF organization to predict likelihood of return to sinus rhythm with DC cardioversion, burst pacing, and catheter ablation. It could thus be hypothesized that an increase in OI is reflective of a reduction in the number of reentrant circuits or rotors maintaining AF. Assessment of AF organization could therefore be a potentially useful way to determine the extent of AF ablation required.

Current evidence indicate that ablation of ganglionated plexi can result in maintenance of sinus rhythm in patients with persistent AF, possibly by modifying the autonomic tone. However, the effect of pharmacological vagal blockade with drugs, such as atropine, on persistent AF is not well studied and has not been previously compared with ablation. The findings presented in this thesis showed that catheter ablation aimed at isolating pulmonary veins and addressing left atrial substrates in persistent AF appear to also result in attenuation of the parasympathetic tone, suggesting that autonomic modulation can occur even with ablation not deliberately targeting ganglionated plexi. Changes in the frequency domain measures of DF and OI are seen following pharmacological vagal blockade (by administration of atropine) and mirrors that achieved with catheter ablation. This supports the notion that the autonomic nervous system may play an active role in the persistence of AF.

We have also shown that potential targets of ablation such as fractionated sites and DF sites are not strongly linked spatially and that catheter ablation appear to affect these 2 substrates to different extents, suggesting that they could well be separate

entities. This is in agreement with data from animal studies and complements the existing literature by providing additional post-ablation data.

The work carried out in this thesis therefore adds to the existing knowledge of AF and provides further interesting insight into the behaviour of AF in the frequency domain, in the presence of pharmacological influences as well as radiofrequency ablation.

7.2 Future directions

Most of the electrograms analysed in this thesis are recorded from stationary catheters in the coronary sinus. Further examination of the spatial characteristics of human AF electrograms in the entire left atrium is needed to fully appreciate the complex behaviour of AF. High density mapping using non-conctact balloon mapping methods will achieve this by allowing for a large number of sites to be analysed at the same point in time. Further assessment of the spatiotemporal effects of ablation and drugs using this technique is recommended.

Head-to-head comparison of ablation of substrates, such as sites containing fractionated electrograms or maximal DF have not been carried out to date. Considering that these may in fact be spatially separate entities, it would be worthwhile conducting randomized studies to compare the efficacy of either approach. Larger scale studies examining AF organization in real-time during ablation will also help to define and establish its role as a tool to guiding the extent of ablation.

References

1. *Tuan J, Osman F, Jeilan M, Kundu S, Mantravadi R, Stafford PJ, Ng GA. Increase in organization index predicts atrial fibrillation termination with flecainide post-ablation: spectral analysis of intracardiac electrograms. Europace 2010;12(4):488-93.*

2. *Gley E. [Exercitatio...de motu cordis et sanguinis by Harvey; its significance; its development.]. Prog Med (Paris) 1957;85(11):243-6.*

3. *McMichael J. History of atrial fibrillation 1628-1819 Harvey - de Senac - Laennec. Br Heart J 1982;48(3):193-7.*

4. *Toledo-Pereyra LH. Exercitatio Anatomica De Motus Cordis et Sanguinis in Animalibus surgical revolution. J Invest Surg 2008;21(6):302-10.*

5. *Mackenzie J. The Interpretation of the Pulsations in the Jugluar Veins. Am J Med Sci 1907;134(1):12-34.*

6. *Lewis T. Report Cxix. Auricular Fibrillation: a Common Clinical Condition. Br Med J 1909;2(2552):1528.*

7. *Wolf PA, Benjamin EJ, Belanger AJ, Kannel WB, Levy D, D'Agostino RB. Secular trends in the prevalence of atrial fibrillation: The Framingham Study. Am Heart J 1996;131(4):790-5.*

8. *Fuster V, Ryden LE, Cannom DS, Crijns HJ, Curtis AB, Ellenbogen KA, Halperin JL, Le Heuzey JY, Kay GN, Lowe JE, Olsson SB, Prystowsky EN, Tamargo JL, Wann S. ACC/AHA/ESC 2006 guidelines for the management of patients with atrial fibrillation-executive summary: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines and the European Society of Cardiology Committee for Practice Guidelines (Writing Committee to Revise the 2001 Guidelines for the Management of Patients with Atrial Fibrillation). Eur Heart J 2006;27(16):1979-2030.*

9. *Camm AJ, Kirchhof P, Lip GY, Schotten U, Savelieva I, Ernst S, Van Gelder IC, Al-Attar N, Hindricks G, Prendergast B, Heidbuchel H, Alfieri O, Angelini A, Atar D, Colonna P, De Caterina R, De Sutter J, Goette A, Gorenek B, Heldal M, Hohloser SH, Kolh P, Le Heuzey JY, Ponikowski P, Rutten FH, Vahanian A, Auricchio A, Bax J, Ceconi C, Dean V, Filippatos G, Funck-Brentano C, Hobbs R, Kearney P, McDonagh T, Popescu BA, Reiner Z, Sechtem U, Sirnes PA, Tendera M, Vardas PE, Widimsky P, Vardas PE, Agladze V, Aliot E, Balabanski T, Blomstrom-Lundqvist C, Capucci A, Crijns H, Dahlof B, Folliguet T, Glikson M, Goethals M, Gulba DC, Ho SY, Klautz RJ, Kose S, McMurray J, Perrone Filardi P, Raatikainen P, Salvador MJ, Schalij MJ, Shpektor A, Sousa J, Stepinska J, Uuetoa H, Zamorano JL, Zupan I. Guidelines for the management of atrial fibrillation: The Task Force for the Management of Atrial Fibrillation of the European Society of Cardiology (ESC). Eur Heart J 2010.*

10. *Go AS, Hylek EM, Phillips KA, Chang Y, Henault LE, Selby JV, Singer DE. Prevalence of diagnosed atrial fibrillation in adults: national implications for rhythm management and stroke prevention: the AnTicoagulation and Risk Factors in Atrial Fibrillation (ATRIA) Study. Jama 2001;285(18):2370-5.*

11. *Miyasaka Y, Barnes ME, Gersh BJ, Cha SS, Bailey KR, Abhayaratna WP, Seward JB, Tsang TS. Secular trends in incidence of atrial fibrillation in Olmsted County, Minnesota, 1980 to 2000, and implications on the projections for future prevalence. Circulation 2006;114(2):119-25.*

12. *Heeringa J, van der Kuip DA, Hofman A, Kors JA, van Herpen G, Stricker BH, Stijnen T, Lip GY, Witteman JC. Prevalence, incidence and lifetime risk of atrial fibrillation: the Rotterdam study. Eur Heart J 2006;27(8):949-53.*

13. *Lloyd-Jones DM, Wang TJ, Leip EP, Larson MG, Levy D, Vasan RS, D'Agostino RB, Massaro JM, Beiser A, Wolf PA, Benjamin EJ. Lifetime risk for development of atrial fibrillation: the Framingham Heart Study. Circulation 2004;110(9):1042-6.*

14. *Friberg J, Buch P, Scharling H, Gadsbphioll N, Jensen GB. Rising rates of hospital admissions for atrial fibrillation. Epidemiology 2003;14(6):666-72.*

15. *Wattigney WA, Mensah GA, Croft JB. Increasing trends in hospitalization for atrial fibrillation in the United States, 1985 through 1999: implications for primary prevention. Circulation 2003;108(6):711-6.*

16. *Le Heuzey JY, Paziaud O, Piot O, Said MA, Copie X, Lavergne T, Guize L. Cost of care distribution in atrial fibrillation patients: the COCAF study. Am Heart J 2004;147(1):121-6.*

17. *Wolf PA, Abbott RD, Kannel WB. Atrial fibrillation as an independent risk factor for stroke: the Framingham Study. Stroke 1991;22(8):983-8.*

18. *Cairns JA, Connolly SJ. Nonrheumatic atrial fibrillation. Risk of stroke and role of antithrombotic therapy. Circulation 1991;84(2):469-81.*

19. *Simons LA, McCallum J, Friedlander Y, Simons J. Risk factors for ischemic stroke: Dubbo Study of the elderly. Stroke 1998;29(7):1341-6.*

20. *Hylek EM, Go AS, Chang Y, Jensvold NG, Henault LE, Selby JV, Singer DE. Effect of intensity of oral anticoagulation on stroke severity and mortality in atrial fibrillation. N Engl J Med 2003;349(11):1019-26.*

21. *van Walraven C, Hart RG, Singer DE, Laupacis A, Connolly S, Petersen P, Koudstaal PJ, Chang Y, Hellemons B. Oral anticoagulants vs aspirin in nonvalvular atrial fibrillation: an individual patient meta-analysis. Jama 2002;288(19):2441-8.*

22. *Stroke Prevention In Atrial Fibrillation I. The stroke prevention in atrial fibrillation III study: rationale, design, and patient features. J Stroke Cerebrovasc Dis 1997;6(5):341-53.*

23. *Patients with nonvalvular atrial fibrillation at low risk of stroke during treatment with aspirin: Stroke Prevention in Atrial Fibrillation III Study. The SPAF III Writing Committee for the Stroke Prevention in Atrial Fibrillation Investigators. Jama 1998;279(16):1273-7.*

24. *Gage BF, Waterman AD, Shannon W, Boechler M, Rich MW, Radford MJ. Validation of clinical classification schemes for predicting stroke: results from the National Registry of Atrial Fibrillation. Jama 2001;285(22):2864-70.*

25. *Ott A, Breteler MM, de Bruyne MC, van Harskamp F, Grobbee DE, Hofman A. Atrial fibrillation and dementia in a population-based study. The Rotterdam Study. Stroke 1997;28(2):316-21.*

26. *Kilander L, Andren B, Nyman H, Lind L, Boberg M, Lithell H. Atrial fibrillation is an independent determinant of low cognitive function: a cross-sectional study in elderly men. Stroke 1998;29(9):1816-20.*

27. *Knecht S, Oelschlager C, Duning T, Lohmann H, Albers J, Stehling C, Heindel W, Breithardt G, Berger K, Ringelstein EB, Kirchhof P, Wersching H. Atrial fibrillation in stroke-free patients is associated with memory impairment and hippocampal atrophy. Eur Heart J 2008.*

28. *Packer DL, Bardy GH, Worley SJ, Smith MS, Cobb FR, Coleman RE, Gallagher JJ, German LD. Tachycardia-induced cardiomyopathy: a reversible form of left ventricular dysfunction. Am J Cardiol 1986;57(8):563-70.*

29. *Redfield MM, Kay GN, Jenkins LS, Mianulli M, Jensen DN, Ellenbogen KA. Tachycardia-related cardiomyopathy: a common cause of ventricular dysfunction in patients with atrial fibrillation referred for atrioventricular ablation. Mayo Clin Proc 2000;75(8):790-5.*

30. *Thrall G, Lane D, Carroll D, Lip GY. Quality of life in patients with atrial fibrillation: a systematic review. Am J Med 2006;119(5):448 e1-19.*

31. *Krahn AD, Manfreda J, Tate RB, Mathewson FA, Cuddy TE. The natural history of atrial fibrillation: incidence, risk factors, and prognosis in the Manitoba Follow-Up Study. Am J Med 1995;98(5):476-84.*

32. *Benjamin EJ, Wolf PA, D'Agostino RB, Silbershatz H, Kannel WB, Levy D. Impact of atrial fibrillation on the risk of death: the Framingham Heart Study. Circulation 1998;98(10):946-52.*

33. *Stewart S, Hart CL, Hole DJ, McMurray JJ. A population-based study of the long-term risks associated with atrial fibrillation: 20-year follow-up of the Renfrew/Paisley study. Am J Med 2002;113(5):359-64.*

34. *Vidaillet H, Granada JF, Chyou PH, Maassen K, Ortiz M, Pulido JN, Sharma P, Smith PN, Hayes J. A population-based study of mortality among patients with atrial fibrillation or flutter. Am J Med 2002;113(5):365-70.*

35. *Poole-Wilson PA, Swedberg K, Cleland JG, Di Lenarda A, Hanrath P, Komajda M, Lubsen J, Lutiger B, Metra M, Remme WJ, Torp-Pedersen C, Scherhag A, Skene A. Comparison of carvedilol and metoprolol on clinical outcomes in patients with chronic heart failure in the Carvedilol Or Metoprolol European Trial (COMET): randomised controlled trial. Lancet 2003;362(9377):7-13.*

36. *Maggioni AP, Latini R, Carson PE, Singh SN, Barlera S, Glazer R, Masson S, Cere E, Tognoni G, Cohn JN. Valsartan reduces the incidence of atrial fibrillation in patients with heart failure: results from the Valsartan Heart Failure Trial (Val-HeFT). Am Heart J 2005;149(3):548-57.*

37. *Wang TJ, Larson MG, Levy D, Vasan RS, Leip EP, Wolf PA, D'Agostino RB, Murabito JM, Kannel WB, Benjamin EJ. Temporal relations of atrial fibrillation and congestive heart failure and their joint influence on mortality: the Framingham Heart Study. Circulation 2003;107(23):2920-5.*

38. *Nieuwlaat R, Capucci A, Camm AJ, Olsson SB, Andresen D, Davies DW, Cobbe S, Breithardt G, Le Heuzey JY, Prins MH, Levy S, Crijns HJ. Atrial fibrillation management: a prospective survey in ESC member countries: the Euro Heart Survey on Atrial Fibrillation. Eur Heart J 2005;26(22):2422-34.*

39. *Benjamin EJ, Levy D, Vaziri SM, D'Agostino RB, Belanger AJ, Wolf PA. Independent risk factors for atrial fibrillation in a population-based cohort. The Framingham Heart Study. Jama 1994;271(11):840-4.*

40. *Borzecki AM, Bridgers DK, Liebschutz JM, Kader B, Kazis LE, Berlowitz DR. Racial differences in the prevalence of atrial fibrillation among males. J Natl Med Assoc 2008;100(2):237-45.*

41. *Fox CS, Parise H, D'Agostino RB, Sr., Lloyd-Jones DM, Vasan RS, Wang TJ, Levy D, Wolf PA, Benjamin EJ. Parental atrial fibrillation as a risk factor for atrial fibrillation in offspring. Jama 2004;291(23):2851-5.*

42. *Brugada R, Tapscott T, Czernuszewicz GZ, Marian AJ, Iglesias A, Mont L, Brugada J, Girona J, Domingo A, Bachinski LL, Roberts R. Identification of a genetic locus for familial atrial fibrillation. N Engl J Med 1997;336(13):905-11.*

43. *Ellinor PT, Shin JT, Moore RK, Yoerger DM, MacRae CA. Locus for atrial fibrillation maps to chromosome 6q14-16. Circulation 2003;107(23):2880-3.*

44. *Madu EC, Baugh DS, Gbadebo TD, Dhala A, Cardoso S. Effect of ethnicity and hypertension on atrial conduction: evaluation with high-resolution P-wave signal averaging. Clin Cardiol 2001;24(9):597-602.*

45. *Ravelli F, Allessie M. Effects of atrial dilatation on refractory period and vulnerability to atrial fibrillation in the isolated Langendorff-perfused rabbit heart. Circulation 1997;96(5):1686-95.*

46. *Psaty BM, Manolio TA, Kuller LH, Kronmal RA, Cushman M, Fried LP, White R, Furberg CD, Rautaharju PM. Incidence of and risk factors for atrial fibrillation in older adults. Circulation 1997;96(7):2455-61.*

47. *Kannel WB, Wolf PA, Benjamin EJ, Levy D. Prevalence, incidence, prognosis, and predisposing conditions for atrial fibrillation: population-based estimates. Am J Cardiol 1998;82(8A):2N-9N.*

48. *Li D, Fareh S, Leung TK, Nattel S. Promotion of atrial fibrillation by heart failure in dogs: atrial remodeling of a different sort. Circulation 1999;100(1):87-95.*

49. *Mukamal KJ, Tolstrup JS, Friberg J, Jensen G, Gronbaek M. Alcohol consumption and risk of atrial fibrillation in men and women: the Copenhagen City Heart Study. Circulation 2005;112(12):1736-42.*

50. *Wang TJ, Parise H, Levy D, D'Agostino RB, Sr., Wolf PA, Vasan RS, Benjamin EJ. Obesity and the risk of new-onset atrial fibrillation. Jama 2004;292(20):2471-7.*

51. *Gami AS, Hodge DO, Herges RM, Olson EJ, Nykodym J, Kara T, Somers VK. Obstructive sleep apnea, obesity, and the risk of incident atrial fibrillation. J Am Coll Cardiol 2007;49(5):565-71.*

52. *Buch P, Friberg J, Scharling H, Lange P, Prescott E. Reduced lung function and risk of atrial fibrillation in the Copenhagen City Heart Study. Eur Respir J 2003;21(6):1012-6.*

53. *Cappola AR, Fried LP, Arnold AM, Danese MD, Kuller LH, Burke GL, Tracy RP, Ladenson PW. Thyroid status, cardiovascular risk, and mortality in older adults. Jama 2006;295(9):1033-41.*

54. *Dublin S, Glazer NL, Smith NL, Psaty BM, Lumley T, Wiggins KL, Page RL, Heckbert SR. Diabetes mellitus, glycemic control, and risk of atrial fibrillation. J Gen Intern Med 2010;25(8):853-8.*

55. *Calkins H, Brugada J, Packer DL, Cappato R, Chen SA, Crijns HJ, Damiano RJ, Jr., Davies DW, Haines DE, Haissaguerre M, Iesaka Y, Jackman W, Jais P, Kottkamp H, Kuck KH, Lindsay BD, Marchlinski FE, McCarthy PM, Mont JL, Morady F, Nademanee K, Natale A, Pappone C, Prystowsky E, Raviele A, Ruskin JN, Shemin RJ. HRS/EHRA/ECAS expert consensus statement on catheter and surgical ablation of atrial fibrillation: recommendations for personnel, policy, procedures and follow-up. A report of the Heart Rhythm Society (HRS) Task Force on Catheter and Surgical Ablation of Atrial Fibrillation developed in partnership with the European Heart Rhythm Association (EHRA) and the European Cardiac Arrhythmia Society (ECAS); in collaboration with the American College of Cardiology (ACC), American Heart Association (AHA), and the Society of Thoracic Surgeons (STS). Endorsed and approved by the governing bodies of the American College of Cardiology, the American Heart Association, the European Cardiac Arrhythmia Society, the European Heart Rhythm Association, the Society of Thoracic Surgeons, and the Heart Rhythm Society. Europace 2007;9(6):335-79.*

56. *Wijffels MC, Kirchhof CJ, Dorland R, Allessie MA. Atrial fibrillation begets atrial fibrillation. A study in awake chronically instrumented goats. Circulation 1995;92(7):1954-68.*

57. *Daoud EG, Bogun F, Goyal R, Harvey M, Man KC, Strickberger SA, Morady F. Effect of atrial fibrillation on atrial refractoriness in humans. Circulation 1996;94(7):1600-6.*

58. *Yue L, Feng J, Gaspo R, Li GR, Wang Z, Nattel S. Ionic remodeling underlying action potential changes in a canine model of atrial fibrillation. Circ Res 1997;81(4):512-25.*

59. *Gaspo R, Bosch RF, Bou-Abboud E, Nattel S. Tachycardia-induced changes in Na+ current in a chronic dog model of atrial fibrillation. Circ Res 1997;81(6):1045-52.*

60. *Gaspo R, Bosch RF, Talajic M, Nattel S. Functional mechanisms underlying tachycardia-induced sustained atrial fibrillation in a chronic dog model. Circulation 1997;96(11):4027-35.*

61. *Yue L, Melnyk P, Gaspo R, Wang Z, Nattel S. Molecular mechanisms underlying ionic remodeling in a dog model of atrial fibrillation. Circ Res 1999;84(7):776-84.*

62. *Van Wagoner DR, Pond AL, Lamorgese M, Rossie SS, McCarthy PM, Nerbonne JM. Atrial L-type Ca2+ currents and human atrial fibrillation. Circ Res 1999;85(5):428-36.*

63. *Bosch RF, Zeng X, Grammer JB, Popovic K, Mewis C, Kuhlkamp V. Ionic mechanisms of electrical remodeling in human atrial fibrillation. Cardiovasc Res 1999;44(1):121-31.*

64. *Garrey WE. The nature of fibrillary contraction of the heart - its relation to tissue mass and form. Am J Physiol 1914(33):397-414.*

65. *Vaziri SM, Larson MG, Benjamin EJ, Levy D. Echocardiographic predictors of nonrheumatic atrial fibrillation. The Framingham Heart Study. Circulation 1994;89(2):724-30.*

66. *Parkash R, Green MS, Kerr CR, Connolly SJ, Klein GJ, Sheldon R, Talajic M, Dorian P, Humphries KH. The association of left atrial size and occurrence of atrial fibrillation: a prospective cohort study from the Canadian Registry of Atrial Fibrillation. Am Heart J 2004;148(4):649-54.*

67. *Sanfilippo AJ, Abascal VM, Sheehan M, Oertel LB, Harrigan P, Hughes RA, Weyman AE. Atrial enlargement as a consequence of atrial fibrillation. A prospective echocardiographic study. Circulation 1990;82(3):792-7.*

68. *Moe GK, Abildskov JA. Atrial fibrillation as a self-sustaining arrhythmia independent of focal discharge. Am Heart J 1959;58(1):59-70.*

69. *Ausma J, Wijffels M, Thone F, Wouters L, Allessie M, Borgers M. Structural changes of atrial myocardium due to sustained atrial fibrillation in the goat. Circulation 1997;96(9):3157-63.*

70. *Frustaci A, Chimenti C, Bellocci F, Morgante E, Russo MA, Maseri A. Histological substrate of atrial biopsies in patients with lone atrial fibrillation. Circulation 1997;96(4):1180-4.*

71. *Kostin S, Klein G, Szalay Z, Hein S, Bauer EP, Schaper J. Structural correlate of atrial fibrillation in human patients. Cardiovasc Res 2002;54(2):361-79.*

72. *Burstein B, Nattel S. Atrial fibrosis: mechanisms and clinical relevance in atrial fibrillation. J Am Coll Cardiol 2008;51(8):802-9.*

73. *Wetzel U, Boldt A, Lauschke J, Weigl J, Schirdewahn P, Dorszewski A, Doll N, Hindricks G, Dhein S, Kottkamp H. Expression of connexins 40 and 43 in human left atrium in atrial fibrillation of different aetiologies. Heart 2005;91(2):166-70.*

74. *Mayer AG. Rhythmical pulsation in scyphomedusae. Carnegie Instituition of Washington 1906.*

75. *Mines GR. On dynamic equilibrium in the heart. J Physiol 1913;46(4-5):349-83.*

76. *Mines GR. On circulating excitations in heart muscle and their possible relation to tachycardia and fibrillation. Trans R Soc Can 1914;4:43-52.*

77. *Winterberg H. Ueber Herzflimmern und seine Beeinflussung durch Kampher. Z Exp Pathol Ther 1906(3):182-208.*

78. *Rothberger CJ, Winterberg H. ber Vorhofflimmern und Vorhofflattern. Pflugers Arch Gesamte Physiol Menschen Tiere 1915(160):42-90.*

79. *Lewis T, Schleiter HG. The relation of regular tachycardias of auricular origin to auricular fibrillation. Heart 1912;3:173- 193.*

80. *Lewis T, Drury AN, Iliescu CC. A demonstration of circus movement in clinical fibrillation of the auricles. Heart 1921;8:361- 369.*

81. *Wiener N, Rosenblueth A. The mathematical formulation of the problem of conduction of impulses in a network of connected excitable elements, specifically in cardiac muscle. Arch Inst Cardiol Mex 1946(16):205-265.*

82. *Scherf D. Studies on auricular tachycardia caused by aconitine administration. Proc Soc Exp Biol Med 1947;64(2):233-9.*

83. *Scherf D, Romano FJ, Terranova R. Experimental studies on auricular flutter and auricular fibrillation. Am Heart J 1948;36(2):241-51.*

84. *Moe GK. On Multiple Wavelet Hypothesis of Atrial Fibrillation. Archives Internationales De Pharmacodynamie Et De Therapie 1962;140(1-2):183-&.*

85. *Moe GK, Rheinboldt WC, Abildskov JA. A Computer Model of Atrial Fibrillation. Am Heart J 1964;67:200-20.*

86. *Allessie MA, Lammers WJ, Bonke IM, Hollen J. Experimental evaluation of Moe's multiple wavelet hypothesis of atrial fibrillation. New York: Grune and Stratton; 1985.*

87. *Allessie MA, Bonke FI, Schopman FJ. Circus movement in rabbit atrial muscle as a mechanism of tachycardia. III. The "leading circle" concept: a new model of circus movement in cardiac tissue without the involvement of an anatomical obstacle. Circ Res 1977;41(1):9-18.*

88. *Rensma PL, Allessie MA, Lammers WJ, Bonke FI, Schalij MJ. Length of excitation wave and susceptibility to reentrant atrial arrhythmias in normal conscious dogs. Circ Res 1988;62(2):395-410.*

89. *Wang Z, Page P, Nattel S. Mechanism of flecainide's antiarrhythmic action in experimental atrial fibrillation. Circ Res 1992;71(2):271-87.*

90. *Wang J, Bourne GW, Wang Z, Villemaire C, Talajic M, Nattel S. Comparative mechanisms of antiarrhythmic drug action in experimental atrial fibrillation. Importance of use-dependent effects on refractoriness. Circulation 1993;88(3):1030-44.*

91. *Panfilov AV, Pertsov AM. [Mechanism of spiral waves initiation in active media, connected with critical curvature phenomenon]. Biofizika 1982;27(5):886-9.*

92. *Winfree AT. Varieties of spiral wave behavior: An experimentalist's approach to the theory of excitable media. Chaos 1991;1(3):303-334.*

93. *Davidenko JM. Spiral wave activity: a possible common mechanism for polymorphic and monomorphic ventricular tachycardias. J Cardiovasc Electrophysiol 1993;4(6):730-46.*

94. *Pertsov AM, Davidenko JM, Salomonsz R, Baxter WT, Jalife J. Spiral waves of excitation underlie reentrant activity in isolated cardiac muscle. Circ Res 1993;72(3):631-50.*

95. *Skanes AC, Mandapati R, Berenfeld O, Davidenko JM, Jalife J. Spatiotemporal periodicity during atrial fibrillation in the isolated sheep heart. Circulation 1998;98(12):1236-48.*

96. *Mandapati R, Skanes A, Chen J, Berenfeld O, Jalife J. Stable microreentrant sources as a mechanism of atrial fibrillation in the isolated sheep heart. Circulation 2000;101(2):194-9.*

97. *Schuessler RB, Grayson TM, Bromberg BI, Cox JL, Boineau JP. Cholinergically mediated tachyarrhythmias induced by a single extrastimulus in the isolated canine right atrium. Circ Res 1992;71(5):1254-67.*

98. *Jalife J, Berenfeld O, Skanes A, Mandapati R. Mechanisms of atrial fibrillation: mother rotors or multiple daughter wavelets, or both? J Cardiovasc Electrophysiol 1998;9(8 Suppl):S2-12.*

99. *Jalife J, Berenfeld O, Mansour M. Mother rotors and fibrillatory conduction: a mechanism of atrial fibrillation. Cardiovasc Res 2002;54(2):204-16.*

100. *Jais P, Haissaguerre M, Shah DC, Chouairi S, Gencel L, Hocini M, Clementy J. A focal source of atrial fibrillation treated by discrete radiofrequency ablation. Circulation 1997;95(3):572-6.*

101. *Haissaguerre M, Jais P, Shah DC, Takahashi A, Hocini M, Quiniou G, Garrigue S, Le Mouroux A, Le Metayer P, Clementy J. Spontaneous initiation of atrial fibrillation by ectopic beats originating in the pulmonary veins. N Engl J Med 1998;339(10):659-66.*

102. *Coumel P, Attuel P, Lavallee J, Flammang D, Leclercq JF, Slama R. [The atrial arrhythmia syndrome of vagal origin]. Arch Mal Coeur Vaiss 1978;71(6):645-56.*

103. *Coumel P. Paroxysmal atrial fibrillation: a disorder of autonomic tone? Eur Heart J 1994;15 Suppl A:9-16.*

104. *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(2):289-98.*

105. *Vaitkevicius R, Saburkina I, Rysevaite K, Vaitkeviciene I, Pauziene N, Zaliunas R, Schauerte P, Jalife J, Pauza DH. Nerve supply of the human pulmonary veins: an anatomical study. Heart Rhythm 2009;6(2):221-8.*

106. *Tan AY, Chen PS, Chen LS, Fishbein MC. Autonomic nerves in pulmonary veins. Heart Rhythm 2007;4(3 Suppl):S57-60.*

107. *Sharifov OF, Fedorov VV, Beloshapko GG, Glukhov AV, Yushmanova AV, Rosenshtraukh LV. Roles of adrenergic and cholinergic stimulation in spontaneous atrial fibrillation in dogs. J Am Coll Cardiol 2004;43(3):483-90.*

108. *Patterson E, Po SS, Scherlag BJ, Lazzara R. Triggered firing in pulmonary veins initiated by in vitro autonomic nerve stimulation. Heart Rhythm 2005;2(6):624-31.*

109. *Arora R, Ulphani JS, Villuendas R, Ng J, Harvey L, Thordson S, Inderyas F, Lu Y, Gordon D, Denes P, Greene R, Crawford S, Decker R, Morris A, Goldberger J, Kadish AH. Neural substrate for atrial fibrillation: implications for targeted parasympathetic blockade in the posterior left atrium. Am J Physiol Heart Circ Physiol 2008;294(1):H134-44.*

110. *Lewis T, Drury AN, Bulger HA. Flutter and Fibrillation; the Effects of Vagal Stimulation. Heart 1921;8:141.*

111. *Armour JA, Hageman GR, Randall WC. Arrhythmias induced by local cardiac nerve stimulation. Am J Physiol 1972;223(5):1068-75.*

112. *Higgins CB, Vatner SF, Braunwald E. Parasympathetic control of the heart. Pharmacol Rev 1973;25(1):119-55.*

113. *Lazzara R, Scherlag BJ, Robinson MJ, Samet P. Selective in situ parasympathetic control of the canine sinoatrial and atrioventricular nodes. Circ Res 1973;32(3):393-401.*

114. *Zipes DP, Mihalick MJ, Robbins GT. Effects of selective vagal and stellate ganglion stimulation of atrial refractoriness. Cardiovasc Res 1974;8(5):647-55.*

115. *Schauerte P, Scherlag BJ, Pitha J, Scherlag MA, Reynolds D, Lazzara R, Jackman WM. Catheter ablation of cardiac autonomic nerves for prevention of vagal atrial fibrillation. Circulation 2000;102(22):2774-80.*

116. *Liu L, Nattel S. Differing sympathetic and vagal effects on atrial fibrillation in dogs: role of refractoriness heterogeneity. Am J Physiol 1997;273(2 Pt 2):H805-16.*

117. *Inoue H, Zipes DP. Changes in atrial and ventricular refractoriness and in atrioventricular nodal conduction produced by combinations of vagal and sympathetic stimulation that result in a constant spontaneous sinus cycle length. Circ Res 1987;60(6):942-51.*

118. *Schauerte P, Scherlag BJ, Patterson E, Scherlag MA, Matsudaria K, Nakagawa H, Lazzara R, Jackman WM. Focal atrial fibrillation: experimental evidence for a pathophysiologic role of the autonomic nervous system. J Cardiovasc Electrophysiol 2001;12(5):592-9.*

119. *Wit AL, Cranefield PF. Triggered and automatic activity in the canine coronary sinus. Circ Res 1977;41(4):434-45.*

120. *Elvan A, Pride HP, Eble JN, Zipes DP. Radiofrequency catheter ablation of the atria reduces inducibility and duration of atrial fibrillation in dogs. Circulation 1995;91(8):2235-44.*

121. *Cox JL, Boineau JP, Schuessler RB, Ferguson TB, Jr., Cain ME, Lindsay BD, Corr PB, Kater KM, Lappas DG. Successful surgical treatment of atrial fibrillation. Review and clinical update. Jama 1991;266(14):1976-80.*

122. *Scherlag BJ, Nakagawa H, Jackman WM, Yamanashi WS, Patterson E, Po S, Lazzara R. Electrical stimulation to identify neural elements on the heart: their role in atrial fibrillation. J Interv Card Electrophysiol 2005;13 Suppl 1:37-42.*

123. *Pappone C, Santinelli V, Manguso F, Vicedomini G, Gugliotta F, Augello G, Mazzone P, Tortoriello V, Landoni G, Zangrillo A, Lang C, Tomita T, Mesas C, Mastella E, Alfieri O. Pulmonary Vein Denervation Enhances Long-Term Benefit After Circumferential Ablation for Paroxysmal Atrial Fibrillation. Circulation 2004;109(3):327-334.*

124. *Burashnikov A, Antzelevitch C. Reinduction of atrial fibrillation immediately after termination of the arrhythmia is mediated by late phase 3 early afterdepolarization-induced triggered activity. Circulation 2003;107(18):2355-60.*

125. *Patterson E, Lazzara R, Szabo B, Liu H, Tang D, Li YH, Scherlag BJ, Po SS. Sodium-calcium exchange initiated by the Ca2+ transient: an arrhythmia trigger within pulmonary veins. J Am Coll Cardiol 2006;47(6):1196-206.*

126. *Lip GY, Boos CJ. Antithrombotic treatment in atrial fibrillation. Heart 2006;92(2):155-61.*

127. *Lip GY, Edwards SJ. Stroke prevention with aspirin, warfarin and ximelagatran in patients with non-valvular atrial fibrillation: a systematic review and meta-analysis. Thromb Res 2006;118(3):321-33.*

128. *Hart RG, Pearce LA, Aguilar MI. Meta-analysis: antithrombotic therapy to prevent stroke in patients who have nonvalvular atrial fibrillation. Ann Intern Med 2007;146(12):857-67.*

129. *Tieleman RG, Van Gelder IC, Crijns HJ, De Kam PJ, Van Den Berg MP, Haaksma J, Van Der Woude HJ, Allessie MA. Early recurrences of atrial fibrillation after electrical cardioversion: a result of fibrillation-induced electrical remodeling of the atria? J Am Coll Cardiol 1998;31(1):167-73.*

130. *Capucci A, Villani GQ, Aschieri D, Rosi A, Piepoli MF. Oral amiodarone increases the efficacy of direct-current cardioversion in restoration of sinus rhythm in patients with chronic atrial fibrillation. Eur Heart J 2000;21(1):66-73.*

131. *Wyse DG, Waldo AL, DiMarco JP, Domanski MJ, Rosenberg Y, Schron EB, Kellen JC, Greene HL, Mickel MC, Dalquist JE, Corley SD. A comparison of rate control and rhythm control in patients with atrial fibrillation. N Engl J Med 2002;347(23):1825-33.*

132. *Corley SD, Epstein AE, DiMarco JP, Domanski MJ, Geller N, Greene HL, Josephson RA, Kellen JC, Klein RC, Krahn AD, Mickel M, Mitchell LB, Nelson JD, Rosenberg Y, Schron E, Shemanski L, Waldo AL, Wyse DG. Relationships between sinus rhythm, treatment, and survival in the Atrial Fibrillation Follow-Up Investigation of Rhythm Management (AFFIRM) Study. Circulation 2004;109(12):1509-13.*

133. *Van Gelder IC, Hagens VE, Bosker HA, Kingma JH, Kamp O, Kingma T, Said SA, Darmanata JI, Timmermans AJ, Tijssen JG, Crijns HJ. A comparison of rate control and rhythm control in patients with recurrent persistent atrial fibrillation. N Engl J Med 2002;347(23):1834-40.*

134. *Roy D, Talajic M, Nattel S, Wyse DG, Dorian P, Lee KL, Bourassa MG, Arnold JM, Buxton AE, Camm AJ, Connolly SJ, Dubuc M, Ducharme A, Guerra PG, Hohnloser SH, Lambert J, Le Heuzey JY, O'Hara G, Pedersen OD, Rouleau JL, Singh BN, Stevenson LW, Stevenson WG, Thibault B, Waldo AL. Rhythm control versus rate control for atrial fibrillation and heart failure. N Engl J Med 2008;358(25):2667-77.*

135. *Van Gelder IC, Groenveld HF, Crijns HJ, Tuininga YS, Tijssen JG, Alings AM, Hillege HL, Bergsma-Kadijk JA, Cornel JH, Kamp O, Tukkie R, Bosker HA, Van Veldhuisen DJ, Van den Berg MP. Lenient versus strict rate control in patients with atrial fibrillation. N Engl J Med 2010;362(15):1363-73.*

136. *Jenkins LS, Brodsky M, Schron E, Chung M, Rocco T, Jr., Lader E, Constantine M, Sheppard R, Holmes D, Mateski D, Floden L, Prasun M, Greene HL, Shemanski L. Quality of life in atrial fibrillation: the Atrial Fibrillation Follow-up Investigation of Rhythm Management (AFFIRM) study. Am Heart J 2005;149(1):112-20.*

137. *Hagens VE, Ranchor AV, Van Sonderen E, Bosker HA, Kamp O, Tijssen JG, Kingma JH, Crijns HJ, Van Gelder IC. Effect of rate or rhythm control on quality of life in persistent atrial fibrillation. Results from the Rate Control Versus Electrical Cardioversion (RACE) Study. J Am Coll Cardiol 2004;43(2):241-7.*

138. *Hohnloser SH, Kuck KH, Lilienthal J. Rhythm or rate control in atrial fibrillation--Pharmacological Intervention in Atrial Fibrillation (PIAF): a randomised trial. Lancet 2000;356(9244):1789-94.*

139. *Carlsson J, Miketic S, Windeler J, Cuneo A, Haun S, Micus S, Walter S, Tebbe U. Randomized trial of rate-control versus rhythm-control in persistent atrial fibrillation: the Strategies of Treatment of Atrial Fibrillation (STAF) study. J Am Coll Cardiol 2003;41(10):1690-6.*

140. *Dorian P, Paquette M, Newman D, Green M, Connolly SJ, Talajic M, Roy D. Quality of life improves with treatment in the Canadian Trial of Atrial Fibrillation. Am Heart J 2002;143(6):984-90.*

141. *Singh SN, Tang XC, Singh BN, Dorian P, Reda DJ, Harris CL, Fletcher RD, Sharma SC, Atwood JE, Jacobson AK, Lewis HD, Jr., Lopez B, Raisch DW, Ezekowitz MD. Quality of life and exercise performance in patients in sinus rhythm versus persistent atrial fibrillation: a Veterans Affairs Cooperative Studies Program Substudy. J Am Coll Cardiol 2006;48(4):721-30.*

142. *Allessie M, Ausma J, Schotten U. Electrical, contractile and structural remodeling during atrial fibrillation. Cardiovasc Res 2002;54(2):230-46.*

143. *Pokharel S, van Geel PP, Sharma UC, Cleutjens JP, Bohnemeier H, Tian XL, Schunkert H, Crijns HJ, Paul M, Pinto YM. Increased myocardial collagen content in transgenic rats overexpressing cardiac angiotensin-converting enzyme is related to enhanced breakdown of N-acetyl-Ser-Asp-Lys-Pro and increased phosphorylation of Smad2/3. Circulation 2004;110(19):3129-35.*

144. *Weber KT, Brilla CG, Campbell SE, Guarda E, Zhou G, Sriram K. Myocardial fibrosis: role of angiotensin II and aldosterone. Basic Res Cardiol 1993;88 Suppl 1:107-24.*

145. *Wachtell K, Lehto M, Gerdts E, Olsen MH, Hornestam B, Dahlof B, Ibsen H, Julius S, Kjeldsen SE, Lindholm LH, Nieminen MS, Devereux RB. Angiotensin II receptor blockade reduces new-onset atrial fibrillation and subsequent stroke compared to atenolol: the Losartan Intervention For End Point Reduction in Hypertension (LIFE) study. J Am Coll Cardiol 2005;45(5):712-9.*

146. *Ducharme A, Swedberg K, Pfeffer MA, Cohen-Solal A, Granger CB, Maggioni AP, Michelson EL, McMurray JJ, Olsson L, Rouleau JL, Young JB, Yusuf S. Prevention of atrial fibrillation in patients with symptomatic chronic heart failure by candesartan in the Candesartan in Heart failure: assessment of Reduction in Mortality and morbidity (CHARM) program. Am Heart J 2006;151(5):985-91.*

147. *Cox JL, Boineau JP, Schuessler RB, Jaquiss RD, Lappas DG. Modification of the maze procedure for atrial flutter and atrial fibrillation. I. Rationale and surgical results. J Thorac Cardiovasc Surg 1995;110(2):473-84.*

148. *Damiano RJ, Jr., Gaynor SL, Bailey M, Prasad S, Cox JL, Boineau JP, Schuessler RP. The long-term outcome of patients with coronary disease and atrial fibrillation undergoing the Cox maze procedure. J Thorac Cardiovasc Surg 2003;126(6):2016-21.*

149. *Reston JT, Shuhaiber JH. Meta-analysis of clinical outcomes of maze-related surgical procedures for medically refractory atrial fibrillation. Eur J Cardiothorac Surg 2005;28(5):724-30.*

150. *Haissaguerre M, Marcus FI, Fischer B, Clementy J. Radiofrequency catheter ablation in unusual mechanisms of atrial fibrillation: report of three cases. J Cardiovasc Electrophysiol 1994;5(9):743-51.*

151. *Haissaguerre M, Jais P, Shah DC, Gencel L, Pradeau V, Garrigues S, Chouairi S, Hocini M, Le Metayer P, Roudaut R, Clementy J. Right and left atrial radiofrequency catheter therapy of paroxysmal atrial fibrillation. J Cardiovasc Electrophysiol 1996;7(12):1132-44.*

152. *Chen SA, Hsieh MH, Tai CT, Tsai CF, Prakash VS, Yu WC, Hsu TL, Ding YA, Chang MS. Initiation of atrial fibrillation by ectopic beats originating from the pulmonary veins: electrophysiological characteristics, pharmacological responses, and effects of radiofrequency ablation. Circulation 1999;100(18):1879-86.*

153. *Lin WS, Tai CT, Hsieh MH, Tsai CF, Lin YK, Tsao HM, Huang JL, Yu WC, Yang SP, Ding YA, Chang MS, Chen SA. Catheter ablation of paroxysmal atrial fibrillation initiated by non-pulmonary vein ectopy. Circulation 2003;107(25):3176-83.*

154. *Haissaguerre M, Jais P, Shah DC, Garrigue S, Takahashi A, Lavergne T, Hocini M, Peng JT, Roudaut R, Clementy J. Electrophysiological end point for catheter ablation of atrial fibrillation initiated from multiple pulmonary venous foci. Circulation 2000;101(12):1409-17.*

155. *Haissaguerre M, Shah DC, Jais P, Hocini M, Yamane T, Deisenhofer I, Chauvin M, Garrigue S, Clementy J. Electrophysiological breakthroughs from the left atrium to the pulmonary veins. Circulation 2000;102(20):2463-5.*

156. *Pappone C, Oreto G, Lamberti F, Vicedomini G, Loricchio ML, Shpun S, Rillo M, Calabro MP, Conversano A, Ben-Haim SA, Cappato R, Chierchia S. Catheter ablation of paroxysmal atrial fibrillation using a 3D mapping system. Circulation 1999;100(11):1203-8.*

157. *Pappone C, Rosanio S, Oreto G, Tocchi M, Gugliotta F, Vicedomini G, Salvati A, Dicandia C, Mazzone P, Santinelli V, Gulletta S, Chierchia S. Circumferential radiofrequency ablation of pulmonary vein ostia: A new anatomic approach for curing atrial fibrillation. Circulation 2000;102(21):2619-28.*

158. *Pappone C, Oreto G, Rosanio S, Vicedomini G, Tocchi M, Gugliotta F, Salvati A, Dicandia C, Calabro MP, Mazzone P, Ficarra E, Di Gioia C, Gulletta S, Nardi S, Santinelli V, Benussi S, Alfieri O. Atrial electroanatomic remodeling after circumferential radiofrequency pulmonary vein ablation: efficacy of an anatomic approach in a large cohort of patients with atrial fibrillation. Circulation 2001;104(21):2539-44.*

159. *Oral H, Scharf C, Chugh A, Hall B, Cheung P, Good E, Veerareddy S, Pelosi F, Jr., Morady F. Catheter ablation for paroxysmal atrial fibrillation: segmental pulmonary vein ostial ablation versus left atrial ablation. Circulation 2003;108(19):2355-60.*

160. *Karch MR, Zrenner B, Deisenhofer I, Schreieck J, Ndrepepa G, Dong J, Lamprecht K, Barthel P, Luciani E, Schomig A, Schmitt C. Freedom from atrial tachyarrhythmias after catheter ablation of atrial fibrillation: a randomized comparison between 2 current ablation strategies. Circulation 2005;111(22):2875-80.*

161. *Ouyang F, Bansch D, Ernst S, Schaumann A, Hachiya H, Chen M, Chun J, Falk P, Khanedani A, Antz M, Kuck KH. Complete isolation of left atrium surrounding the pulmonary veins: new insights from the double-Lasso technique in paroxysmal atrial fibrillation. Circulation 2004;110(15):2090-6.*

162. *Scharf C, Oral H, Chugh A, Hall B, Good E, Cheung P, Pelosi F, Jr., Morady F. Acute effects of left atrial radiofrequency ablation on atrial fibrillation. J Cardiovasc Electrophysiol 2004;15(5):515-21.*

163. *Gerstenfeld EP, Callans DJ, Sauer W, Jacobson J, Marchlinski FE. Reentrant and nonreentrant focal left atrial tachycardias occur after pulmonary vein isolation. Heart Rhythm 2005;2(11):1195-202.*

164. *Pappone C, Manguso F, Vicedomini G, Gugliotta F, Santinelli O, Ferro A, Gulletta S, Sala S, Sora N, Paglino G, Augello G, Agricola E, Zangrillo A, Alfieri O, Santinelli V. Prevention of iatrogenic atrial tachycardia after ablation of atrial fibrillation: a prospective randomized study comparing circumferential pulmonary vein ablation with a modified approach. Circulation 2004;110(19):3036-42.*

165. *Jais P, Hocini M, Hsu LF, Sanders P, Scavee C, Weerasooriya R, Macle L, Raybaud F, Garrigue S, Shah DC, Le Metayer P, Clementy J, Haissaguerre M. Technique and results of linear ablation at the mitral isthmus. Circulation 2004;110(19):2996-3002.*

166. *Hocini M, Sanders P, Jais P, Hsu LF, Takahashi Y, Rotter M, Clementy J, Haissaguerre M. Techniques for curative treatment of atrial fibrillation. J Cardiovasc Electrophysiol 2004;15(12):1467-71.*

167. *Willems S, Klemm H, Rostock T, Brandstrup B, Ventura R, Steven D, Risius T, Lutomsky B, Meinertz T. Substrate modification combined with pulmonary vein isolation improves outcome of catheter ablation in patients with persistent atrial fibrillation: a prospective randomized comparison. Eur Heart J 2006;27(23):2871-8.*

168. *Wazni O, Marrouche NF, Martin DO, Gillinov AM, Saliba W, Saad E, Klein A, Bhargava M, Bash D, Schweikert R, Erciyes D, Abdul-Karim A, Brachman J, Gunther J, Pisano E, Potenza D, Fanelli R, Natale A. Randomized study comparing combined pulmonary vein-left atrial junction disconnection and cavotricuspid isthmus ablation versus pulmonary vein-left atrial junction disconnection alone in patients presenting with typical atrial flutter and atrial fibrillation. Circulation 2003;108(20):2479-83.*

169. *Jais P, Hocini M, Sanders P, Hsu LF, Takahashi Y, Rotter M, Rostock T, Sacher F, Clementy J, Haissaguerre M. Long-term evaluation of atrial fibrillation ablation guided by noninducibility. Heart Rhythm 2006;3(2):140-5.*

170. *Konings KT, Smeets JL, Penn OC, Wellens HJ, Allessie MA. Configuration of unipolar atrial electrograms during electrically induced atrial fibrillation in humans. Circulation 1997;95(5):1231-41.*

171. *Nademanee K, McKenzie J, Kosar E, Schwab M, Sunsaneewitayakul B, Vasavakul T, Khunnawat C, Ngarmukos T. A new approach for catheter ablation of atrial fibrillation: mapping of the electrophysiologic substrate. J Am Coll Cardiol 2004;43(11):2044-53.*

172. *Nademanee K, Schwab MC, Kosar EM, Karwecki M, Moran MD, Visessook N, Michael AD, Ngarmukos T. Clinical outcomes of catheter substrate ablation for high-risk patients with atrial fibrillation. J Am Coll Cardiol 2008;51(8):843-9.*

173. *Oral H, Chugh A, Good E, Wimmer A, Dey S, Gadeela N, Sankaran S, Crawford T, Sarrazin JF, Kuhne M, Chalfoun N, Wells D, Frederick M, Fortino J, Benloucif-Moore S, Jongnarangsin K, Pelosi F, Jr., Bogun F, Morady F. Radiofrequency catheter ablation of chronic atrial fibrillation guided by complex electrograms. Circulation 2007;115(20):2606-12.*

174. *Oral H, Chugh A, Yoshida K, Sarrazin JF, Kuhne M, Crawford T, Chalfoun N, Wells D, Boonyapisit W, Veerareddy S, Billakanty S, Wong WS, Good E, Jongnarangsin K, Pelosi F, Jr., Bogun F, Morady F. A randomized assessment of the incremental role of ablation of complex fractionated atrial electrograms after antral pulmonary vein isolation for long-lasting persistent atrial fibrillation. J Am Coll Cardiol 2009;53(9):782-9.*

175. *Verma A, Novak P, Macle L, Whaley B, Beardsall M, Wulffhart Z, Khaykin Y. A prospective, multicenter evaluation of ablating complex fractionated electrograms (CFEs) during atrial fibrillation (AF) identified by an automated mapping algorithm: acute effects on AF and efficacy as an adjuvant strategy. Heart Rhythm 2008;5(2):198-205.*

176. *Bencsik G, Martinek M, Hassanein S, Aichinger J, Nesser HJ, Purerfellner H. Acute effects of complex fractionated atrial electrogram ablation on dominant frequency and regulatory index for the fibrillatory process. Europace 2009;11(8):1011-7.*

177. *Estner HL, Hessling G, Ndrepepa G, Wu J, Reents T, Fichtner S, Schmitt C, Bary CV, Kolb C, Karch M, Zrenner B, Deisenhofer I. Electrogram-guided substrate ablation with or without pulmonary vein isolation in patients with persistent atrial fibrillation. Europace 2008;10(11):1281-7.*

178. *Sanders P, Berenfeld O, Hocini M, Jais P, Vaidyanathan R, Hsu LF, Garrigue S, Takahashi Y, Rotter M, Sacher F, Scavee C, Ploutz-Snyder R, Jalife J, Haissaguerre M. Spectral analysis identifies sites of high-frequency activity maintaining atrial fibrillation in humans. Circulation 2005;112(6):789-97.*

179. *Lazar S, Dixit S, Marchlinski FE, Callans DJ, Gerstenfeld EP. Presence of left-to-right atrial frequency gradient in paroxysmal but not persistent atrial fibrillation in humans. Circulation 2004;110(20):3181-6.*

180. *Lazar S, Dixit S, Callans DJ, Lin D, Marchlinski FE, Gerstenfeld EP. Effect of pulmonary vein isolation on the left-to-right atrial dominant frequency gradient in human atrial fibrillation. Heart Rhythm 2006;3(8):889-95.*

181. *Atienza F, Almendral J, Jalife J, Zlochiver S, Ploutz-Snyder R, Torrecilla EG, Arenal A, Kalifa J, Fernandez-Aviles F, Berenfeld O. Real-time dominant frequency mapping and ablation of dominant frequency sites in atrial fibrillation with left-to-right frequency gradients predicts long-term maintenance of sinus rhythm. Heart Rhythm 2009;6(1):33-40.*

182. *Lemola K, Ting M, Gupta P, Anker JN, Chugh A, Good E, Reich S, Tschopp D, Igic P, Elmouchi D, Jongnarangsin K, Bogun F, Pelosi F, Jr., Morady F, Oral H. Effects of two different catheter ablation techniques on spectral characteristics of atrial fibrillation. J Am Coll Cardiol 2006;48(2):340-8.*

183. *Yoshida K, Chugh A, Good E, Crawford T, Myles J, Veerareddy S, Billakanty S, Wong WS, Ebinger M, Pelosi F, Jongnarangsin K, Bogun F, Morady F, Oral H. A critical decrease in dominant frequency and clinical outcome after catheter ablation of persistent atrial fibrillation. Heart Rhythm 2010;7(3):295-302.*

184. *Po SS, Nakagawa H, Jackman WM. Localization of left atrial ganglionated plexi in patients with atrial fibrillation. J Cardiovasc Electrophysiol 2009;20(10):1186-9.*

185. *Pokushalov E, Romanov A, Artyomenko S, Turov A, Shirokova N, Katritsis DG. Left Atrial Ablation at the Anatomic Areas of Ganglionated Plexi for Paroxysmal Atrial Fibrillation. Pacing Clin Electrophysiol 2010(epub).*

186. *Pokushalov E, Romanov A, Artyomenko S, Turov A, Shugayev P, Shirokova N, Katritsis DG. Ganglionated plexi ablation for longstanding persistent atrial fibrillation. Europace 2010;12(3):342-6.*

187. *Pokushalov E, Romanov A, Shugayev P, Artyomenko S, Shirokova N, Turov A, Katritsis DG. Selective ganglionated plexi ablation for paroxysmal atrial fibrillation. Heart Rhythm 2009;6(9):1257-64.*

188. *Cappato R, Calkins H, Chen SA, Davies W, Iesaka Y, Kalman J, Kim YH, Klein G, Packer D, Skanes A. Worldwide survey on the methods, efficacy, and safety of catheter ablation for human atrial fibrillation. Circulation 2005;111(9):1100-5.*

189. *Cappato R, Calkins H, Chen SA, Davies W, Iesaka Y, Kalman J, Kim YH, Klein G, Natale A, Packer D, Skanes A, Ambrogi F, Biganzoli E. Updated worldwide survey on the methods, efficacy, and safety of catheter ablation for human atrial fibrillation. Circ Arrhythm Electrophysiol 2010;3(1):32-8.*

190. *Krittayaphong R, Raungrattanaamporn O, Bhuripanyo K, Sriratanasathavorn C, Pooranawattanakul S, Punlee K, Kangkagate C. A randomized clinical trial of the efficacy of radiofrequency catheter ablation and amiodarone in the treatment of symptomatic atrial fibrillation. J Med Assoc Thai 2003;86 Suppl 1:S8-16.*

191. *Wazni OM, Marrouche NF, Martin DO, Verma A, Bhargava M, Saliba W, Bash D, Schweikert R, Brachmann J, Gunther J, Gutleben K, Pisano E, Potenza D, Fanelli R, Raviele A, Themistoclakis S, Rossillo A, Bonso A, Natale A. Radiofrequency ablation vs antiarrhythmic drugs as first-line treatment of symptomatic atrial fibrillation: a randomized trial. Jama 2005;293(21):2634-40.*

192. *Stabile G, Bertaglia E, Senatore G, De Simone A, Zoppo F, Donnici G, Turco P, Pascotto P, Fazzari M, Vitale DF. Catheter ablation treatment in patients with drug-refractory atrial fibrillation: a prospective, multi-centre, randomized, controlled study (Catheter Ablation For The Cure Of Atrial Fibrillation Study). Eur Heart J 2006;27(2):216-21.*

193. *Pappone C, Augello G, Sala S, Gugliotta F, Vicedomini G, Gulletta S, Paglino G, Mazzone P, Sora N, Greiss I, Santagostino A, LiVolsi L, Pappone N, Radinovic A, Manguso F, Santinelli V. A randomized trial of circumferential pulmonary vein ablation versus antiarrhythmic drug therapy in paroxysmal atrial fibrillation: the APAF Study. J Am Coll Cardiol 2006;48(11):2340-7.*

194. *Oral H, Pappone C, Chugh A, Good E, Bogun F, Pelosi F, Jr., Bates ER, Lehmann MH, Vicedomini G, Augello G, Agricola E, Sala S, Santinelli V, Morady F. Circumferential pulmonary-vein ablation for chronic atrial fibrillation. N Engl J Med 2006;354(9):934-41.*

195. *Jais P, Cauchemez B, Macle L, Daoud E, Khairy P, Subbiah R, Hocini M, Extramiana F, Sacher F, Bordachar P, Klein G, Weerasooriya R, Clementy J, Haissaguerre M. Catheter ablation versus antiarrhythmic drugs for atrial fibrillation: the A4 study. Circulation 2008;118(24):2498-505.*

196. *Wilber DJ, Pappone C, Neuzil P, De Paola A, Marchlinski F, Natale A, Macle L, Daoud EG, Calkins H, Hall B, Reddy V, Augello G, Reynolds MR, Vinekar C, Liu CY, Berry SM, Berry DA. Comparison of antiarrhythmic drug therapy and radiofrequency catheter ablation in patients with paroxysmal atrial fibrillation: a randomized controlled trial. Jama 2010;303(4):333-40.*

197. *Calkins H, Reynolds MR, Spector P, Sondhi M, Xu Y, Martin A, Williams CJ, Sledge I. Treatment of atrial fibrillation with antiarrhythmic drugs or radiofrequency ablation: two systematic literature reviews and meta-analyses. Circ Arrhythm Electrophysiol 2009;2(4):349-61.*

198. *Nair GM, Nery PB, Diwakaramenon S, Healey JS, Connolly SJ, Morillo CA. A systematic review of randomized trials comparing radiofrequency ablation with antiarrhythmic medications in patients with atrial fibrillation. J Cardiovasc Electrophysiol 2009;20(2):138-44.*

199. *Jayam VK, Dong J, Vasamreddy CR, Lickfett L, Kato R, Dickfeld T, Eldadah Z, Dalal D, Blumke DA, Berger R, Halperin HR, Calkins H. Atrial volume reduction following catheter ablation of atrial fibrillation and relation to reduction in pulmonary vein size: an evaluation using magnetic resonance angiography. J Interv Card Electrophysiol 2005;13(2):107-14.*

200. *Reant P, Lafitte S, Jais P, Serri K, Weerasooriya R, Hocini M, Pillois X, Clementy J, Haissaguerre M, Roudaut R. Reverse remodeling of the left cardiac chambers after catheter ablation after 1 year in a series of patients with isolated atrial fibrillation. Circulation 2005;112(19):2896-903.*

201. *Verma A, Kilicaslan F, Adams JR, Hao S, Beheiry S, Minor S, Ozduran V, Claude Elayi S, Martin DO, Schweikert RA, Saliba W, Thomas JD, Garcia M, Klein A, Natale A. Extensive ablation during pulmonary vein antrum isolation has no adverse impact on left atrial function: an echocardiography and cine computed tomography analysis. J Cardiovasc Electrophysiol 2006;17(7):741-6.*

202. *Lemola K, Desjardins B, Sneider M, Case I, Chugh A, Good E, Han J, Tamirisa K, Tsemo A, Reich S, Tschopp D, Igic P, Elmouchi D, Bogun F, Pelosi F, Jr., Kazerooni E, Morady F, Oral H. Effect of left atrial circumferential ablation for atrial fibrillation on left atrial transport function. Heart Rhythm 2005;2(9):923-8.*

203. *Hsu LF, Jais P, Sanders P, Garrigue S, Hocini M, Sacher F, Takahashi Y, Rotter M, Pasquie JL, Scavee C, Bordachar P, Clementy J, Haissaguerre M. Catheter ablation for atrial fibrillation in congestive heart failure. N Engl J Med 2004;351(23):2373-83.*

204. *Kuck KH, Ernst S, Cappato R, Braun E, Lang M, Ben-Haim SA, Hebe J, Ouyang F, Khanedani A, Antz M, Volkmer M. Nonfluoroscopic endocardial catheter mapping of atrial fibrillation. J Cardiovasc Electrophysiol 1998;9(8 Suppl):S57-62.*

205. *Pappone C, Vicedomini G, Manguso F, Gugliotta F, Mazzone P, Gulletta S, Sora N, Sala S, Marzi A, Augello G, Livolsi L, Santagostino A, Santinelli V. Robotic magnetic navigation for atrial fibrillation ablation. J Am Coll Cardiol 2006;47(7):1390-400.*

206. *Saliba W, Reddy VY, Wazni O, Cummings JE, Burkhardt JD, Haissaguerre M, Kautzner J, Peichl P, Neuzil P, Schibgilla V, Noelker G, Brachmann J, Di Biase L, Barrett C, Jais P, Natale A. Atrial fibrillation ablation using a robotic catheter remote control system: initial human experience and long-term follow-up results. J Am Coll Cardiol 2008;51(25):2407-11.*

207. *Natale A, Pisano E, Shewchik J, Bash D, Fanelli R, Potenza D, Santarelli P, Schweikert R, White R, Saliba W, Kanagaratnam L, Tchou P, Lesh M. First human experience with pulmonary vein isolation using a through-the-balloon circumferential ultrasound ablation system for recurrent atrial fibrillation. Circulation 2000;102(16):1879-82.*

208. *Nakagawa H, Antz M, Wong T, Schmidt B, Ernst S, Ouyang F, Vogtmann T, Wu R, Yokoyama K, Lockwood D, Po SS, Beckman KJ, Davies DW, Kuck KH, Jackman WM. Initial experience using a forward directed, high-intensity focused ultrasound balloon catheter for pulmonary vein antrum isolation in patients with atrial fibrillation. J Cardiovasc Electrophysiol 2007;18(2):136-44.*

209. *Neumann T, Vogt J, Schumacher B, Dorszewski A, Kuniss M, Neuser H, Kurzidim K, Berkowitsch A, Koller M, Heintze J, Scholz U, Wetzel U, Schneider MA, Horstkotte D, Hamm CW, Pitschner HF. Circumferential pulmonary vein isolation with the cryoballoon technique results from a prospective 3-center study. J Am Coll Cardiol 2008;52(4):273-8.*

210. *Reddy VY, Neuzil P, Themistoclakis S, Bonso A, Rossillo A, Raviele A, Saliba W, Schwiekert R, Ernst S, Kuck K-H, Ruskin JN, Natale A. Abstract 2439: Visually-Guided Pulmonary Vein Isolation Using a Balloon Ablation Catheter to Treat Patients with Paroxysmal Atrial Fibrillation: One-Year Clincial Outcome Following a Single Ablation Procedure. Circulation 2007;116(16_MeetingAbstracts):II_536-a-.*

211. *Phillips KP, Schweikert RA, Saliba WI, Themistoclakis S, Raviele A, Bonso A, Rossillo A, Burkhardt JD, Cummings J, Natale A. Anatomic location of pulmonary vein electrical disconnection with balloon-based catheter ablation. J Cardiovasc Electrophysiol 2008;19(1):14-8.*

212. *Boersma L, Mulder A, Jansen W, Wever E, Wijffels M. Voltage analysis after multi-electrode ablation with duty-cycled bipolar and unipolar radiofrequency energy: a case report. Europace 2009;11(11):1546-8.*

213. *Scharf C, Boersma L, Davies W, Kanagaratnam P, Peters NS, Paul V, Rowland E, Grace A, Fynn S, Dang L, Oral H, Morady F. Ablation of persistent atrial fibrillation using multielectrode catheters and duty-cycled radiofrequency energy. J Am Coll Cardiol 2009;54(15):1450-6.*

214. *Cleland JG, Coletta AP, Buga L, Ahmed D, Clark AL. Clinical trials update from the American College of Cardiology meeting 2010: DOSE, ASPIRE, CONNECT, STICH, STOP-AF, CABANA, RACE II, EVEREST II, ACCORD, and NAVIGATOR. Eur J Heart Fail 2010;12(6):623-9.*

215. *Botteron GW, Smith JM. Quantitative assessment of the spatial organization of atrial fibrillation in the intact human heart. Circulation 1996;93(3):513-8.*

216. *Gerstenfeld EP, Sahakian AV, Swiryn S. Evidence for transient linking of atrial excitation during atrial fibrillation in humans. Circulation 1992;86(2):375-82.*

217. *Slocum J, Sahakian A, Swiryn S. Computer discrimination of atrial fibrillation and regular atrial rhythms from intra-atrial electrograms. Pacing Clin Electrophysiol 1988;11(5):610-21.*

218. *Ropella KM, Sahakian AV, Baerman JM, Swiryn S. Effects of procainamide on intra-atrial electrograms during atrial fibrillation: implications for detection algorithms. Circulation 1988;77(5):1047-54.*

219. *Rosenbaum DS, Cohen RJ. Frequency based measures of atrial fibrillation in man. IEEE Eng Med Biol Sci 1990;12(2):582-583.*

220. *Everett THt, Moorman JR, Kok LC, Akar JG, Haines DE. Assessment of global atrial fibrillation organization to optimize timing of atrial defibrillation. Circulation 2001;103(23):2857-61.*

221. *Everett THt, Akar JG, Kok LC, Moorman JR, Haines DE. Use of global atrial fibrillation organization to optimize the success of burst pace termination. J Am Coll Cardiol 2002;40(10):1831-40.*

222. *Berenfeld O, Zaitsev AV, Mironov SF, Pertsov AM, Jalife J. Frequency-dependent breakdown of wave propagation into fibrillatory conduction across the pectinate muscle network in the isolated sheep right atrium. Circ Res 2002;90(11):1173-80.*

223. *Nyquist H. Certain topics in telegraph transmission theory. Trans AIEE 1928;47:617-644.*

224. *Botteron GW, Smith JM. A technique for measurement of the extent of spatial organization of atrial activation during atrial fibrillation in the intact human heart. IEEE Trans Biomed Eng 1995;42(6):579-86.*

225. *Everett THt, Kok LC, Vaughn RH, Moorman JR, Haines DE. Frequency domain algorithm for quantifying atrial fibrillation organization to increase defibrillation efficacy. IEEE Trans Biomed Eng 2001;48(9):969-78.*

226. *Sahadevan J, Ryu K, Peltz L, Khrestian CM, Stewart RW, Markowitz AH, Waldo AL. Epicardial mapping of chronic atrial fibrillation in patients: preliminary observations. Circulation 2004;110(21):3293-9.*

227. *Ng J, Kadish AH, Goldberger JJ. Effect of electrogram characteristics on the relationship of dominant frequency to atrial activation rate in atrial fibrillation. Heart Rhythm 2006;3(11):1295-305.*

228. *Berenfeld O, Mandapati R, Dixit S, Skanes AC, Chen J, Mansour M, Jalife J. Spatially distributed dominant excitation frequencies reveal hidden organization in atrial fibrillation in the Langendorff-perfused sheep heart. J Cardiovasc Electrophysiol 2000;11(8):869-79.*

229. *Takahashi Y, Sanders P, Jais P, Hocini M, Dubois R, Rotter M, Rostock T, Nalliah CJ, Sacher F, Clementy J, Haissaguerre M. Organization of frequency spectra of atrial fibrillation: relevance to radiofrequency catheter ablation. J Cardiovasc Electrophysiol 2006;17(4):382-8.*

230. *Elvan A, Linnenbank AC, van Bemmel MW, Misier AR, Delnoy PP, Beukema WP, de Bakker JM. Dominant frequency of atrial fibrillation correlates poorly with atrial fibrillation cycle length. Circ Arrhythm Electrophysiol 2009;2(6):634-44.*

231. *Dibs SR, Ng J, Arora R, Passman RS, Kadish AH, Goldberger JJ. Spatiotemporal characterization of atrial activation in persistent human atrial fibrillation: multisite electrogram analysis and surface electrocardiographic correlations--a pilot study. Heart Rhythm 2008;5(5):686-93.*

232. *Habel N, Znojkiewicz P, Thompson N, Muller JG, Mason B, Calame J, Calame S, Sharma S, Mirchandani G, Janks D, Bates J, Noori A, Karnbach A, Lustgarten DL, Sobel BE, Spector P. The temporal variability of dominant frequency and complex fractionated atrial electrograms constrains the validity of sequential mapping in human atrial fibrillation. Heart Rhythm 2010;7(5):586-93.*

233. *Lin YJ, Tai CT, Kao T, Chang SL, Wongcharoen W, Lo LW, Tuan TC, Udyavar AR, Chen YJ, Higa S, Ueng KC, Chen SA. Consistency of complex fractionated atrial electrograms during atrial fibrillation. Heart Rhythm 2008;5(3):406-12.*

234. *Stiles MK, Brooks AG, John B, Wilson L, Kuklik P, Dimitri H, Lau DH, Roberts-Thomson RL, Mackenzie L, Willoughby S, Young GD, Sanders P. The effect of electrogram duration on quantification of complex fractionated atrial electrograms and dominant frequency. J Cardiovasc Electrophysiol 2008;19(3):252-8.*

235. *Wijffels MC, Dorland R, Mast F, Allessie MA. Widening of the excitable gap during pharmacological cardioversion of atrial fibrillation in the goat: effects of cibenzoline, hydroquinidine, flecainide, and d-sotalol. Circulation 2000;102(2):260-7.*

236. *Haissaguerre M, Sanders P, Hocini M, Hsu LF, Shah DC, Scavee C, Takahashi Y, Rotter M, Pasquie JL, Garrigue S, Clementy J, Jais P. Changes in atrial fibrillation cycle length and inducibility during catheter ablation and their relation to outcome. Circulation 2004;109(24):3007-13.*

237. *Campbell TJ. Kinetics of onset of rate-dependent effects of Class I antiarrhythmic drugs are important in determining their effects on refractoriness in guinea-pig ventricle, and provide a theoretical basis for their subclassification. Cardiovasc Res 1983;17(6):344-52.*

238. *Wang ZG, Pelletier LC, Talajic M, Nattel S. Effects of flecainide and quinidine on human atrial action potentials. Role of rate-dependence and comparison with guinea pig, rabbit, and dog tissues. Circulation 1990;82(1):274-83.*

239. *Lewis T. In: The Mechanism and Graphic Registration of the Heart Beat. 3 ed. London: Shaw & Sons; 1925. p. 319-324.*

240. *Asano Y, Saito J-i, Matsumoto K, Kaneko K, Yamamoto T, Uchida M. On the mechanism of termination and perpetuation of atrial fibrillation. The American Journal of Cardiology 1992;69(12):1033-1038.*

241. *Hellestrand KJ, Bexton RS, Nathan AW, Spurrell RA, Camm AJ. Acute electrophysiological effects of flecainide acetate on cardiac conduction and refractoriness in man. Br Heart J 1982;48(2):140-8.*

242. *Katristis D, Rowland E, O'Nunain S, Shakespeare CF, Poloniecki J, Camm AJ. Effect of flecainide on atrial and ventricular refractoriness and conduction in patients with normal left ventricle: Implications for possible antiarrhythmic and proarrhythmic mechanisms. Eur Heart J 1995;16(12):1930-1935.*

243. *Kneller J, Kalifa J, Zou R, Zaitsev AV, Warren M, Berenfeld O, Vigmond EJ, Leon LJ, Nattel S, Jalife J. Mechanisms of atrial fibrillation termination by pure sodium channel blockade in an ionically-realistic mathematical model. Circ Res 2005;96(5):e35-47.*

244. *Bollmann A, Binias KH, Toepffer I, Molling J, Geller C, Klein HU. Importance of left atrial diameter and atrial fibrillatory frequency for conversion of persistent atrial fibrillation with oral flecainide. Am J Cardiol 2002;90(9):1011-4.*

245. *Husser D, Stridh M, Sornmo L, Geller C, Klein HU, Olsson SB, Bollmann A. Time-frequency analysis of the surface electrocardiogram for monitoring antiarrhythmic drug effects in atrial fibrillation. Am J Cardiol 2005;95(4):526-8.*

246. *Kim KB, Rodefeld MD, Schuessler RB, Cox JL, Boineau JP. Relationship between local atrial fibrillation interval and refractory period in the isolated canine atrium. Circulation 1996;94(11):2961-7.*

247. *Biffi M, Boriani G, Bronzetti G, Capucci A, Branzi A, Magnani B. Electrophysiological effects of flecainide and propafenone on atrial fibrillation cycle and relation with arrhythmia termination. Heart 1999;82(2):176-82.*

248. *Capucci A, Biffi M, Boriani G, Ravelli F, Nollo G, Sabbatani P, Orsi C, Magnani B. Dynamic electrophysiological behavior of human atria during paroxysmal atrial fibrillation. Circulation 1995;92(5):1193-202.*

249. *Yoshida K, Chugh A, Ulfarsson M, Good E, Kuhne M, Crawford T, Sarrazin JF, Chalfoun N, Wells D, Boonyapisit W, Veerareddy S, Billakanty S, Wong WS, Jongnarangsin K, Pelosi F, Jr., Bogun F, Morady F, Oral H. Relationship between the spectral characteristics of atrial fibrillation and atrial tachycardias that occur after catheter ablation of atrial fibrillation. Heart Rhythm 2009;6(1):11-7.*

250. *Yoshida K, Ulfarsson M, Tada H, Chugh A, Good E, Kuhne M, Crawford T, Sarrazin JF, Chalfoun N, Wells D, Jongnarangsin K, Pelosi F, Jr., Bogun F, Morady F, Oral H. Complex electrograms within the coronary sinus: time- and frequency-domain characteristics, effects of antral pulmonary vein isolation, and relationship to clinical outcome in patients with paroxysmal and persistent atrial fibrillation. J Cardiovasc Electrophysiol 2008;19(10):1017-23.*

251. *Elayi CS, Di Biase L, Barrett C, Ching CK, Aly MA, Lucciola M, Bai R, Horton R, Fahmy TS, Verma A, Khaykin Y, Shah J, Morales G, Hongo R, Hao S, Beheiry S, Arruda M, Schweikert RA, Cummings J, Burkhardt JD, Wang P, Al-Ahmad A, Cauchemez B, Gaita F, Natale A. Atrial fibrillation termination as a procedural endpoint during ablation in long-standing persistent atrial fibrillation. Heart Rhythm 2010;7(9):1216-23.*

252. *Estner HL, Hessling G, Ndrepepa G, Luik A, Schmitt C, Konietzko A, Ucer E, Wu J, Kolb C, Pflaumer A, Zrenner B, Deisenhofer I. Acute effects and long-term outcome of pulmonary vein isolation in combination with electrogram-guided substrate ablation for persistent atrial fibrillation. Am J Cardiol 2008;101(3):332-7.*

253. *O'Neill MD, Wright M, Knecht S, Jais P, Hocini M, Takahashi Y, Jonsson A, Sacher F, Matsuo S, Lim KT, Arantes L, Derval N, Lellouche N, Nault I, Bordachar P, Clementy J, Haissaguerre M. Long-term follow-up of persistent atrial fibrillation ablation using termination as a procedural endpoint. Eur Heart J 2009;30(9):1105-12.*

254. *Takahashi Y, O'Neill MD, Hocini M, Dubois R, Matsuo S, Knecht S, Mahapatra S, Lim KT, Jais P, Jonsson A, Sacher F, Sanders P, Rostock T, Bordachar P, Clementy J, Klein GJ, Haissaguerre M. Characterization of electrograms associated with termination of chronic atrial fibrillation by catheter ablation. J Am Coll Cardiol 2008;51(10):1003-10.*

255. *Matsuo S, Lellouche N, Wright M, Bevilacqua M, Knecht S, Nault I, Lim KT, Arantes L, O'Neill MD, Platonov PG, Carlson J, Sacher F, Hocini M, Jais P, Haissaguerre M. Clinical predictors of termination and clinical outcome of catheter ablation for persistent atrial fibrillation. J Am Coll Cardiol 2009;54(9):788-95.*

256. *Lo LW, Tai CT, Lin YJ, Chang SL, Udyavar AR, Hu YF, Ueng KC, Tsai WC, Tuan TC, Chang CJ, Kao T, Tsao HM, Wongcharoen W, Higa S, Chen SA. Predicting factors for atrial fibrillation acute termination during catheter ablation procedures: implications for catheter ablation strategy and long-term outcome. Heart Rhythm 2009;6(3):311-8.*

257. *Oral H, Chugh A, Good E, Crawford T, Sarrazin JF, Kuhne M, Chalfoun N, Wells D, Boonyapisit W, Gadeela N, Sankaran S, Kfahagi A, Jongnarangsin K, Pelosi F, Jr., Bogun F, Morady F. Randomized evaluation of right atrial ablation after left atrial ablation of complex fractionated atrial electrograms for long-lasting persistent atrial fibrillation. Circ Arrhythm Electrophysiol 2008;1(1):6-13.*

258. *Porter M, Spear W, Akar JG, Helms R, Brysiewicz N, Santucci P, Wilber DJ. Prospective study of atrial fibrillation termination during ablation guided by automated detection of fractionated electrograms. J Cardiovasc Electrophysiol 2008;19(6):613-20.*

259. *Haissaguerre M, Hocini M, Sanders P, Sacher F, Rotter M, Takahashi Y, Rostock T, Hsu LF, Bordachar P, Reuter S, Roudaut R, Clementy J, Jais P. Catheter ablation of long-lasting persistent atrial fibrillation: clinical outcome and mechanisms of subsequent arrhythmias. J Cardiovasc Electrophysiol 2005;16(11):1138-47.*

260. *Lemery R, Birnie D, Tang ASL, Green M, Gollob M. Feasibility study of endocardial mapping of ganglionated plexuses during catheter ablation of atrial fibrillation. Heart Rhythm 2006;3(4):387-396.*

261. *Billette J, Nadeau RA, Roberge F. Relation between the minimum RR interval during atrial fibrillation and the functional refractory period of the AV junction. Cardiovasc Res 1974;8(3):347-51.*

262. *Khand AU, Rankin AC, Cleland JG, Gemmell I, Clark E, Macfarlane PW. The assessment of autonomic function in chronic atrial fibrillation: description of a non-invasive technique based on circadian rhythm of atrioventricular nodal functional refractory periods. Europace 2006;8(11):927-34.*

263. *Bilchick KC, Fetics B, Djoukeng R, Fisher SG, Fletcher RD, Singh SN, Nevo E, Berger RD. Prognostic value of heart rate variability in chronic congestive heart failure (Veterans Affairs' Survival Trial of Antiarrhythmic Therapy in Congestive Heart Failure). Am J Cardiol 2002;90(1):24-8.*

264. *Zuanetti G, Neilson JM, Latini R, Santoro E, Maggioni AP, Ewing DJ. Prognostic significance of heart rate variability in post-myocardial infarction patients in the fibrinolytic era. The GISSI-2 results. Gruppo Italiano per lo Studio della Sopravvivenza nell' Infarto Miocardico. Circulation 1996;94(3):432-6.*

265. *Heart rate variability: standards of measurement, physiological interpretation and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. Circulation 1996;93(5):1043-65.*

266. *Stein KM, Borer JS, Hochreiter C, Devereux RB, Kligfield P. Variability of the ventricular response in atrial fibrillation and prognosis in chronic nonischemic mitral regurgitation. Am J Cardiol 1994;74(9):906-11.*

267. *Frey B, Heinz G, Binder T, Wutte M, Schneider B, Schmidinger H, Weber H, Pacher R. Diurnal variation of ventricular response to atrial fibrillation in patients with advanced heart failure. Am Heart J 1995;129(1):58-65.*

268. *van den Berg MP, Haaksma J, Brouwer J, Tieleman RG, Mulder G, Crijns HJ. Heart rate variability in patients with atrial fibrillation is related to vagal tone. Circulation 1997;96(4):1209-16.*

269. *Toivonen L, Kadish A, Kou W, Morady F. Determinants of the ventricular rate during atrial fibrillation. J Am Coll Cardiol 1990;16(5):1194-200.*

270. *Hsieh MH, Chiou CW, Wen ZC, Wu CH, Tai CT, Tsai CF, Ding YA, Chang MS, Chen SA. Alterations of heart rate variability after radiofrequency catheter ablation of focal atrial fibrillation originating from pulmonary veins. Circulation 1999;100(22):2237-43.*

271. *Oh S, Zhang Y, Bibevski S, Marrouche NF, Natale A, Mazgalev TN. Vagal denervation and atrial fibrillation inducibility: epicardial fat pad ablation does not have long-term effects. Heart Rhythm 2006;3(6):701-8.*

272. *Calkins H, Brugada J, Packer DL, Cappato R, Chen SA, Crijns HJ, Damiano RJ, Jr., Davies DW, Haines DE, Haissaguerre M, Iesaka Y, Jackman W, Jais P, Kottkamp H, Kuck KH, Lindsay BD, Marchlinski FE, McCarthy PM, Mont JL, Morady F, Nademanee K, Natale A, Pappone C, Prystowsky E, Raviele A, Ruskin JN, Shemin RJ. HRS/EHRA/ECAS expert Consensus Statement on catheter and surgical ablation of atrial fibrillation: recommendations for personnel, policy, procedures and follow-up. A report of the Heart Rhythm Society (HRS) Task Force on catheter and surgical ablation of atrial fibrillation. Heart Rhythm 2007;4(6):816-61.*

273. *Verma A, Wulffhart Z, Beardsall M, Whaley B, Hill C, Khaykin Y. Spatial and temporal stability of complex fractionated electrograms in patients with persistent atrial fibrillation over longer time periods: relationship to local electrogram cycle length. Heart Rhythm 2008;5(8):1127-33.*

274. *Roux JF, Gojraty S, Bala R, Liu CF, Hutchinson MD, Dixit S, Callans DJ, Marchlinski F, Gerstenfeld EP. Complex fractionated electrogram distribution and temporal stability in patients undergoing atrial fibrillation ablation. J Cardiovasc Electrophysiol 2008;19(8):815-20.*

275. *Lin YJ, Tai CT, Kao T, Chang SL, Lo LW, Tuan TC, Udyavar AR, Wongcharoen W, Hu YF, Tso HW, Tsai WC, Chang CJ, Ueng KC, Higa S, Chen SA. Spatiotemporal organization of the left atrial substrate after circumferential pulmonary vein isolation of atrial fibrillation. Circ Arrhythm Electrophysiol 2009;2(3):233-41.*

276. *Kalifa J, Tanaka K, Zaitsev AV, Warren M, Vaidyanathan R, Auerbach D, Pandit S, Vikstrom KL, Ploutz-Snyder R, Talkachou A, Atienza F, Guiraudon G, Jalife J, Berenfeld O. Mechanisms of wave fractionation at boundaries of high-frequency excitation in the posterior left atrium of the isolated sheep heart during atrial fibrillation. Circulation 2006;113(5):626-33.*

277. *Stiles MK, Brooks AG, Kuklik P, John B, Dimitri H, Lau DH, Wilson L, Dhar S, Roberts-Thomson RL, Mackenzie L, Young GD, Sanders P. High-density mapping of atrial fibrillation in humans: relationship between high-frequency activation and electrogram fractionation. J Cardiovasc Electrophysiol 2008;19(12):1245-53.*

278. *Katritsis D, Giazitzoglou E, Sougiannis D, Voridis E, Po SS. Complex fractionated atrial electrograms at anatomic sites of ganglionated plexi in atrial fibrillation. Europace 2009;11(3):308-15.*

279. *Wu TJ, Doshi RN, Huang HL, Blanche C, Kass RM, Trento A, Cheng W, Karagueuzian HS, Peter CT, Chen PS. Simultaneous biatrial computerized mapping during permanent atrial fibrillation in patients with organic heart disease. J Cardiovasc Electrophysiol 2002;13(6):571-7.*

Appendix

Published Papers

Tuan J, Osman F, Jeilan M, Kundu S, Mantravadi R, Stafford P, Ng G.A. Increase in Organization Index predicts Atrial Fibrillation termination with Flecainide post-ablation – spectral analysis of intracardiac electrograms. Europace 2010; 12(4):488-93 http://europace.oxfordjournals.org/content/12/4/488.abstract

Tuan J, Jeilan M, Kundu S, Nicolson W, Chung I, Stafford P, Ng G.A. Regional fractionation and dominant frequency in persistent Atrial Fibrillation: Effects of left atrial ablation and evidence of spatial relationship. Europace 2011; 13(11):1550-56 http://europace.oxfordjournals.org/content/13/11/1550.abstract

Abstracts

Tuan J, Kundu S, Jeilan M, Osman F, Mantravadi R, Stafford P, Ng G.A. Organization of Atrial Signals is Critical Prior to Cardioversion of Atrial Fibrillation with Intravenous Flecainide: Insight from Spectral Analysis of Intracardiac Electrograms. Circulation. 2008;118:S829

http://circ.ahajournals.org/cgi/content/meeting_abstract/118/18_MeetingAbstracts/S_829-b

Tuan J, Jeilan M, Kundu S, Osman F, Stafford P, Mantravadi R, Ng G.A. Spectal analysis and outcomes after catheter ablation for atrial fibrillation. Heart Rhythm 2008;5(5 suppl):S267

http://www.heartrhythmjournal.com/article/S1547-5271(11)00295-5/pdf

Tuan J, Jeilan M, Nicolson W, Yusuf S, Kundu S, Stafford PJ, Ng, G.A. Catheter ablation for persistent atrial fibrillation: changes in autonomic tone and frequency spectrum mirror that of vagal blockade. Europace 2009;11(Suppl 4):iv9

http://europace.oxfordjournals.org/content/11/suppl_4/iv9.full.pdf

Tuan J, Jeilan M, Nicolson W, Kundu S, Stafford PJ, Ng G.A. Regional fractionation and dominant frequency in persistent atrial fibrillation: effects of left atrial ablation and evidence of spatial relationship. Europace 2009;11(Suppl 4):iv3

http://europace.oxfordjournals.org/content/11/suppl_4/iv1.full.pdf