

SHORT THESIS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY (PhD)

**Single-shot ablation techniques for atrial fibrillation ablation:
evaluation of clinical success and safety**

by Alexandra Kiss, MD

Supervisor: Zoltán Csanádi, MD, PhD



UNIVERSITY OF DEBRECEN
DOCTORAL SCHOOL OF KÁLMÁN LAKI

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Head of the **Examination Committee:** Prof. Dr. György Balla, MD, MHAS
Members of the Examination Committee: Prof. Dr. Ádám Böhm, MD, DSc
Dr. István Lőrincz, MD, PhD

The Examination takes place at the Department of Pediatrics, Faculty of Medicine, University of Debrecen, October 3, 2014. 11 am

Head of the **Defense Committee:** Prof. Dr. György Balla MD, MHAS
Reviewers: Prof. Dr. Róbert Gábor Kiss, MD, PhD
Dr. Tünde Csépany, MD, PhD

Members of the Defense Committee: Prof. Dr. Ádám Böhm, MD, DSc
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The PhD Defense takes place at the Lecture Hall of Bldg. A, Department of Internal Medicine, University of Debrecen, October 3, 2014. 1 pm

1. Introduction

1.1. Magnitude of atrial fibrillation, treatment strategies

Atrial fibrillation (AF) is the most frequent of all sustained arrhythmias and carries an increased risk of stroke, hospitalization and mortality. Its prevalence is increasing with the increasing age of the population. Common conditions associated with aging such as diabetes, obesity, sleep apnea, hypertension, heart failure, coronary artery disease, valvular heart disease, cardiac surgery, and smoking may predispose individuals to AF development and progression. Based on observations of long-term morbidity and mortality associated with AF, aggressive treatment of the rhythm should improve outcomes. According to the current guidelines and clinical practice in the first-line treatment of AF administration of antiarrhythmic drugs (AADs) is recommended both in paroxysmal and in persistent AF. However, AADs are able to maintain sinus rhythm (SR) only in 50 % of cases during one-year follow-up. Amiodarone, which has been the most efficient AAD in treatment of AF cannot be administered in one-third of patients due to long-term side effects.

Catheter ablation has emerged as a clinically useful alternative to drug therapy in case of failure of at least one AAD or even as a first-line therapy in selected cases. Left atrial muscular fibers extending into the pulmonary veins (PVs) are important source of ectopic beats, initiating frequent paroxysms of atrial fibrillation. These foci respond to treatment with radiofrequency (RF) ablation. Therefore pulmonary vein isolation (PVI) is the mainstay of any catheter-based treatment for patients with paroxysmal AF.

Conventionally, PVI consists of placement of multiple RF lesions encircling all pulmonary veins. To create these several cm-long lesions by achieving transmuralty at each point and ensuring a continuous line with no gap is technically challenging, requires long procedure and fluoroscopy time and the additional use of a 3-D electroanatomical mapping system. Novel catheter designs and energy forms have been introduced as alternatives to point-by-point RF ablation aiming at a safer and simpler procedure with a faster learning process and comparable success rates. Important principal of these technologies is that once they were properly placed at the ostium of the PVs usually no or only minimal manipulation is required for successful PVI. These “single-shoot” tools include the multipolar duty-cycled circular RF catheter (PVAC=Pulmonary Vein Ablation Catheter), the Cryoballoon (CB) catheter and the novel Multipoler Irrigated Radiofrequency Ablation Catheter (nMARQ).

1.2. Electrical reconnection accountable for arrhythmia recurrence

Regardless of the AF ablation technology used, repeat procedures are needed in 15-65% of the patients for a long-term arrhythmia control. In the majority of these cases, AF recurrence is due to electrical reconnection in some of the previously isolated PVs. Closure of the electrical gaps by placing focal RF lesions along the previous ablation line created during circumferential RF ablation has become a standard technique for repeat ablations. However, the standard method for a redo after an initial CB ablation, which is considered more sensitive to the anatomical characteristics of the left atrium, is less well established.

1.3. Risk of cerebral ischaemia during catheter ablation of atrial fibrillation

The transcatheter treatment of AF is a complex intervention which requires the introduction of often bulky hardware into the left atrium (LA), energy applications over a large area of the LA endocardium and prolonged indwelling in the systemic circulation. Further, these procedures are performed in patients, who are at inherently increased risk of a thromboembolic complication, including stroke. It is not surprising therefore, that cerebrovascular accidents have been among the most feared complications, evoking significant concern, since the inception of AF ablation.

The first worldwide survey on catheter ablation for AF, reflecting the early up to the year 2005 concluded that clinical stroke occurred in 0.28% and transient ischaemic attack (TIA) in 0.66% of the patients. The update of that survey, relating to AF ablations performed between 2003 and 2006, indicated similar rates of cerebrovascular complications (0.23% for stroke, 0.71% for TIA) despite an apparently more difficult patient population with a more enlarged LA and more persistent AF.

A meta-analysis based on the data of 6936 patients who underwent AF ablation by the end of 2006 found that stroke and TIA occurred in 0.3% and 0.2% respectively. Stroke incidences as high as 5% and as low as 0% have also been reported as single-center findings. Although the complication rates associated with any procedure, including AF ablation, generally decrease as the experience gained, this was not demonstrated in a high-volume center: while the overall complication rate fell in a 10-year period from 11.1% to 1.6%, the incidence of stroke and TIA remained unchanged. Thromboembolic events typically occur within 24 hours of the ablation procedure, with the high-risk period extending for 2 weeks thereafter. Importantly, stroke is a significant cause of periprocedural death during AF ablation: an international survey on AF ablation in 162 centers reported details of 32 deaths in

32,569 patients. The fatal outcome was attributed to stroke in 5 (16%) of these 32 cases. On the other hand, patients who survive a stroke related AF ablation often have a favorable long-term prognosis. During a mean 38-month follow-up of 26 patients who suffered AF ablation-related stroke in a high-volume center (2 patients died), complete long-term functional and neurocognitive recovery was documented in most patients, irrespective of the severity of the periprocedural stroke.

While major periprocedural complications, including clinical stroke or transient ischemic attack (TIA) has a relatively low risk, significant concern has been raised regarding the long-term safety of PVI as clinically silent cerebral ischemia (SCI) detected by diffusion-weighted (DW) MRI has been reported in 5-40% of the patients, depending on the ablation technology used. Further, the limited data available suggest that a cognitive decline potentially related to SCI can be demonstrated several months after these ablations in some patients. In some studies, a markedly higher incidence of SCI was reported after PVI performed with phased radiofrequency (RF) and the PV ablation catheter (PVAC), as compared with the cryoballoon (CB) or focal irrigated RF ablation. More recent reports with phased RF ablation, however, demonstrated a significant decrease in the incidence of SCI, these favourable results were attributed to the use of more rigorous periprocedural anticoagulation protocols and some specific modifications in the procedural technique. These procedural changes were based on the results of animal studies and included the avoidance of overlap between 2 electrodes during RF applications and careful sheath management to prevent air embolization. Software modifications refining the power handling of the GENius RF generator have also been implemented.

The recording of microembolic signals (MESs) in the middle cerebral arteries (MCAs) by transcranial Doppler (TCD) has been used to assess microembolization during different cardiovascular procedures. Although DW MRI is regarded as the gold standard for the demonstration of cerebral lesions post-ablation, recording of the MESs provides a unique opportunity through which to monitor the intensity of microembolus generation during different phases of the procedure. In line with the initial DW MRI results, when we used the earlier version of the GENius generator we detected a significantly higher number of MESs during ablation with the PVAC as compared with the CB. Further, we demonstrated that the majority of the microemboli were generated during the energy delivery (ED) with phased RF ablations, while the rate of microembolization was relatively uniform throughout the procedure with the CB. Whether the procedural and technical modifications which decreased

lesion formation on DW MRI with the phased RF technology would also reduce the microembolization detected by TCD is unknown.

We recently also demonstrated a significant correlation between different biophysical parameters of RF delivery and the number of microemboli. It is known, that there are significant anatomical differences between the left and the right-sided PVs: the left atrial appendage (LAA) and the ridge between the LAA and the left-sided PVs pose additional difficulties in positioning the ablation catheter with sufficiently good contact to ensure a durable lesion. Whether these anatomical differences have an impact of microembolization is not known. Further, PVI can be performed in either sinus rhythm (SR) or in AF, which may influence microembolization. It is also unknown, whether the ongoing rhythm affects microembolization rate.

Aims of our study:

- Evaluation of the long-term success rates achieved with the PVAC ablation performed as a redo procedure for arrhythmia recurrence after an initial CB ablation.
- Analyzation of our TCD data obtained with phased RF ablations in relation to the procedural changes implemented in recent years and to compared the MES data with those obtained by using the CB and the novel nMARQTM multipolar irrigated RF ablation system.
- Investigation, whether the site of ablation (as concerns the PVs on the left versus those on the right side) and the ongoing rhythm during energy deliveries have a significant effect on the MES count during phased RF ablation.

2. Patients and methods

The study was approved by the local ethics committee. All patients provided their signed written informed consent prior to inclusion.

2.1. Inclusion and exclusion criteria for pulmonary vein isolation

Regardless the aim of our studies patients, for PVI for symptomatic paroxysmal or persistent AF not adequately controlled by at least one antiarrhythmic drug were eligible for inclusion in the study. The exclusion criteria included long-standing persistent AF, hyper- and hypothyroidism, valvular heart disease, heart failure of NYHA class III or IV, a left ventricular ejection fraction $\leq 40\%$, a LA diameter exceeding 50 mm, a LA thrombus, documented carotid stenosis, previous ischemic stroke or TIA, prior cardiac surgery or ablation in the LA, unstable angina or myocardial infarction within the last 3 months, severe chronic obstructive pulmonary disease, known bleeding disorders, contraindication to oral anticoagulation and pregnancy.

2.2. Further prerequisites in the different studies

2.2.1. Phased radiofrequency ablation for atrial fibrillation recurrence after cryoballoon ablation

Consecutive patients with paroxysmal or persistent AF undergoing a repeat PVI with phased RF ablation using the PVAC after an initial CB ablation were enrolled in the study. All patients had documented episodes of AF after CB despite a trial of IC or III AAD. Results of a follow-up at least 6 months after the redo procedure had to be available for all patients.

2.2.2. Transcranial measurement of cerebral microembolic signals during pulmonary vein isolation performed with single-shot techniques

Patients undergoing PVI with one of the 3 single-shot ablation techniques and Transcranial Doppler monitoring during the whole procedure were eligible for inclusion in the study.

In cases of phased RF ablation patients' data were analyzed in 3 treatment groups based on procedural modifications implemented through recent years, as follows:

1. PVAC Group I. Initial series of ablations were performed with the use of software version

14.3 for the GENius generator. Bipolar/unipolar RF application was started at a ratio of 4:1 for each PV and changed to a bipolar/unipolar ratio of 2:1 for a deeper lesion when a sufficient reduction in local electrogram amplitude could not be achieved after multiple RF deliveries. No attempts were made to avoid the potential interaction between the first and the last electrode (E1-E10) in the PVAC.

2. PVAC Group II. Ablations were performed with software version 14.4 for the GENius generator. Modifications in the procedural technique were implemented in this group of patients as follows. Potential interaction between the most distal (E1) and the most proximal (E10) electrodes was considered and simultaneous EDs on these poles were attempted only after fluoroscopic assessment of the interelectrode distance, which was considered adequate if the space between E1 and E10 was at least double the fixed 3-mm interelectrode distance as assessed from multiple projections. Furthermore, ablations were started in the 2:1 mode and changed to 1:1 in those rare instances when adequate amplitude change and PVI could not be achieved after multiple RF applications. In addition, the distal circular segment of the PVAC catheter was submerged and captured by the introduction device in a saline bath prior to insertion into the FlexCath sheath. This maneuver was used to prevent air entrapment around the array and the introduction of air into the LA through the transseptal sheath.

3. PVAC Group III. Ablations were performed using the most recent software version (15.0) for the GENius generator. RF delivery to E10 is not supported by this software thereby excluding the possibility of E1-E10 interaction. No procedural modifications as compared to those in PVAC Group II. were implemented during these ablations.

2.2.3. Effect of the ongoing rhythm and the site of energy delivery on cerebral microembolization during phased radiofrequency ablation

Patients undergoing PVI with the use of PVAC and the GENius™ 14.4 RF generator were enrolled to this study.

2.3. Patient preparation and pre-procedural evaluation

Patients were admitted to the hospital 1 or 2 days prior to the procedure. Those on oral anticoagulation continued to take the drug and the procedure was performed with an international normalized ratio in the therapeutic range. For all other patients, low molecular

weight heparin was started twice daily in a weight- adjusted dose and administered until 12 hours prior to the procedure. All patients scheduled for ablation were examined by transesophageal echocardiography within 24 hours to rule out an intracardiac thrombus. The LA and PV anatomies were assessed by means of multislice cardiac CT imaging before the first (CB) ablation.

All procedures were performed under conscious sedation with midazolam and fentanyl. Decapolar (BARD Electrophysiology Inc., Lowell, MA, USA) and quadripolar (Woxx 4 J, 6F, Biotronik, SE & Co. KG, Berlin, Germany) catheters were advanced from the femoral vein and positioned into the coronary sinus and the right ventricle. Surface electrocardiograms and bipolar intracardiac electrograms were registered with a Prucka, GE Medical digital recording system. A single transseptal puncture was performed under fluoroscopic, or in some cases under intracardiac echocardiographic (ICE) guidance, using a Brockenbrough needle (St. Jude, Inc., Zaventem, Belgium) and a *Swartz* sheath (St. Jude, Zaventem, Belgium). This sheath was exchanged for the deflectable 12 Fr FlexCath sheath (Medtronic CryoCath LP, Kirkland, Quebec, Canada), which was flushed continuously with heparinized saline, and which was used with any of the ablation catheters utilized in this study. Immediately after the transseptal puncture, a 150 IU/kg body weight intravenous (iv) heparin bolus was administered, followed by a continuous infusion to maintain a minimum ACT target level of 300 s during ablations. Additional 2000-5000 IU iv boluses of heparin were administered as needed to attain the minimum target ACT level.

2.4. Ablation procedures

2.4.1. Cryoballoon ablation

A 28-mm CB was used in all cases. The CB was introduced into the PV ostium over an inner lumen mapping catheter (Achieve, Medtronic Ablation Frontiers LCC, Carlsbad, CA, USA) which is capable of mapping PV potentials before, during and after cryo applications. The best possible occlusion of the PVs was facilitated by the steerable sheath and by positioning the guidewire in a PV branch to provide maximal support. Furthermore, all special maneuvers described previously, such as the “pull-down” or the “hockey stick” techniques, were used as needed.

PV occlusion was assessed by means of a hand-held injection of contrast medium (Optiray Covidien Deutschland GmbH, Neustadt/Donau, Germany) through the injection side-port of the Arctic Front catheter. A minimum of two 5-min freezing cycles were applied per PV. The balloon was repositioned for each application, preferably with the guidewire

situated in a different branch of the PV in order to maximize the effect of freezing at different aspects of the ostium. Before the start of ablation of the septal veins, a quadripolar catheter was placed in the superior caval vein, where constant capture of the right phrenic nerve could be achieved. One stimulus at maximal output was delivered every 5 s, with manual assessment of the diaphragmatic movement. The freezing cycle was terminated immediately if loss or weakening of the diaphragm response occurred. PVI was assessed on the basis of the signals recorded by the Achieve wire.

2.4.2. Phased radiofrequency ablation

The catheter was advanced through the FlexCath sheath over a 0.032-inch guidewire (BARD Electrophysiology Inc., Lowell, MA, USA), which was positioned selectively in each PV. The electrical conduction properties of the PV were assessed on the basis of the signals recorded by the PVAC electrodes after placement inside the ostium. Before the first RF application at each PV, the positions of the electrodes relative to the PV ostium were always confirmed by means of contrast injection through the FlexCath sheath. Care was always taken to apply the RF outside the PV in the antral region, targeting potentials of high amplitude on as many electrodes as possible for each application. RF energy was applied for 60 s, usually 3-4 times per PV, until PVI was achieved. The target temperature was 60 °C, measured separately for all bipoles. Any electrode pair that failed to reach at least 50 °C during RF delivery was switched off to avoid ineffective ED due to improper contact at the electrode-tissue interface. Common ostia were isolated by inserting the guidewire into the different side branches and ablating subsequent segments of the targeted veins. The PV conduction was reassessed after each RF application, the electrodes being advanced inside the ostium. The endpoint of the procedure was the electrical isolation of all PVs, as confirmed by an entrance block.

2.4.3. Ablation with the novel Multipolar Irrigated Radiofrequency Ablation catheter

The nMARQ™ (Biosense Webster, Inc., Diamond Bar, CA, USA) is a multi-electrode, irrigated RF ablation catheter with a uni-directional deflectable tip. The distal tip section is circular or semilunar and contains 10 platinum ring electrodes with irrigation holes at both ends for stimulation, recording and ablation. The diameter of the circular or semilunar loop is variable to accommodate the LA anatomy. The catheter is integrated with the CARTO 3 electroanatomical mapping system, which provides mapping and real-time navigation capabilities. For ablation, the catheter is used in conjunction with the nMARQ™ Multi-

Channel RF Generator, which is able to deliver RF simultaneously to multiple electrodes in unipolar or bipolar mode.

The circular-tip catheter was used exclusively in this study. The catheter was advanced to the LA through the FlexCath guiding sheath. The 3-dimensional map of the LA was constructed by using the CARTO™ electroanatomical mapping system (Biosense Webster, Inc., Diamond Bar, CA, USA) equipped with Cartomerge™ software (Biosense Webster, Inc., Diamond Bar, CA, USA). The pre-ablation cardiac CT image of the LA with the proximal portions of the PVs was imported and registered for real-time mapping, focusing on the PVs and the PV antra of the LA. In addition, the positions of the electrodes relative to the PV ostium were always confirmed before the first RF application at each PV, by means of contrast injection through the FlexCath sheath. Care was taken to apply the RF outside the PV in the antral region, targeting potentials of high amplitude on as many electrodes as possible for each application. RF energy was applied for up to a maximum of 60 s at a maximum of 20 W/electrode in the unipolar mode, with the target temperature set at 43 °C. Electrodes not demonstrating the desired rise in temperature up to 40 °C during energy delivery were switched off. The irrigation flow rate between RF applications was 4 ml/min, which was increased to 60 ml/min, starting 5 s before the onset of RF application and maintained until 5 s after termination. The electrical conduction properties of the PVs were assessed via the recordings through the electrodes of the nMARQ catheter. The endpoint of the procedure was the electrical isolation of all PVs, as confirmed by an entrance block.

2.5. Follow-up after phased radiofrequency ablation

Patients were usually discharged within 2 days after the ablation. Following the procedure a vitamin K antagonist was continued for at least 3 months. Patients taking an AAD before the procedure continued the medication for 3 months post-ablation. It was then discontinued if the patient was free of an AF relapse. Follow-up visits were scheduled at 6 weeks, and 3, 6, 9 and 12 months post-procedure, and every 6 months thereafter. 12-lead ECG was performed at each follow-up. 24-hour Holter was performed at both the 3- and at the 9-month follow-up. In addition, patients were provided with transtelephonic event recorders and asked to transmit their ECG at least twice a day (while having no symptoms) for 3 weeks at 6 and 12 months postablation. Follow-up visits were scheduled at every 6 months after 1 year, and included 12-lead ECG recording and 24-hour Holter recordings. Event recorders after 1 year were used if any symptom emerged which could potentially be attributed to AF

recurrence. The follow-up regime was the same following the initial (cryoballoon) and the redo (phased radiofrequency) ablation procedures. Arrhythmia-free survival was calculated with a 3-month blanking period post-ablation.

2.6. Transcranial Doppler recording and evaluation of the microembolic signal count

TCD recording was performed throughout the whole period of LA access (from the transseptal puncture to the removal of all catheters and sheaths from the LA). The transducer was held in place by a proprietary headpiece supplied with the system. The MCAs were bilaterally insonated from transtemporal windows by using a multifrequency Doppler (Multi Dop T digital, DWL, QL software 2.8) which insonates simultaneously with frequencies of 2 and 2.5 MHz. The system is capable of the automatic online identification of true MESs with a sensitivity of 100% and a specificity of 99.3%, and also of discrimination between gaseous and solid emboli with a specificity of 96.5%. The identification of true MESs with parallel artefact rejection is possible by implementing an event detector system, using a previously published algorithm to detect high-intensity signals due to emboli. This step is followed by a second algorithm using data from the dual-frequency insonation to determine whether the MESs are to be attributed to a solid or to a gaseous embolus. Differentiation is possible because the reflection of ultrasound power is dependent not only on the size of the embolus, but also on its composition and the insonating frequency used: solid emboli reflect more ultrasound at 2 MHz than at 2.5 MHz, whereas the opposite is true for gaseous emboli. TCD parameter settings as recommended by the consensus criteria [24] were kept constant during the procedures. The insonation depth was 45-55 mm, the sample volume was 8 mm, and the power was 60-100 mW. An example of a TCD record is given in Figure 1. MES counts were collected and evaluated separately during different stages of the procedure, as follows:

1. Transseptal puncture: the 30-s period after crossing the interatrial septum with the transseptal needle.
2. PV angiography: contrast injection through the injection port of the CryoCath catheter or the transseptal sheath during PVAC or nMARQ ablation.
3. Energy delivery: from the start until 15 s after the termination of ED

4. The remainder of the procedure: that part of the LA access period during which none of the aforementioned maneuvers were performed.

As bilateral insonation of the MCA could not be achieved in all patients for technical reasons, the results were provided as MES count per MCA, i.e. either the mean of bilateral recordings or the number from the unilateral recording.

2.7. Recording of ablation parameters for evaluation of role of the role of ongoing rhythm and the site of ablation

The ongoing rhythm (AF or SR) and the targeted vein were recorded during each ED. In those rare events when the rhythm changed during the ED, the longer-lasting rhythm was registered as the ongoing rhythm.

Generator files for each ablation during the procedure were collected from the GENius 14.4 ablation generator. The files included information on the power for each electrode sampled at 1 Hz, and the temperature for each electrode sampled at 8 Hz. These data were analyzed together with the ongoing rhythm and the target PV.

2.8. Statistical analysis

2.8.1. Statistical analysis for long-term clinical success of phased radiofrequency ablation for arrhythmia recurrence after a failed cryoballoon ablation

The post-operative survival until the onset of arrhythmia was compared across levels of categorical factors and across terciles derived from continuous explanatory variables through the use of log-rank tests. Survival tendencies were visualized by graphing Kaplan-Meier survival functions against the follow-up time. In order to assess whether the difficulty in achieving isolation of a PV with the CB was predictive of the difficulty during PVI with the PVAC, patients were categorized into 2 groups: those requiring ≤ 3 , and those requiring ≥ 4 cryo applications per PV. So as to address the presence of multiple measurements clustered within patients, multilevel mixed-effects linear regression was used to compare these groups in terms of continuous outcome variables. Outcome variables were transformed to improve normality if necessary. Cryo groups were compared in terms of categorical outcome variables through the use of multilevel mixed-effects logistic regression.

2.8.2. Statistical methods for transcranial measurement of cerebral microembolic signals during pulmonary vein isolation performed with single-shot techniques

Statistical analysis was performed by using IBM SPSS 20. Data are reported as means and standard deviation (SD) (with 95% confidence intervals). The normality of continuous variables was evaluated by means of the Kolmogorov- Smirnov test. Statistical differences between groups were determined by using analysis of variance when the data showed normal distribution and homoscedasticity (Levene's statistic); otherwise, the Kruskal-Wallis test was performed. For comparison of the MES counts obtained during CB ablation versus those during the other 4 techniques, the Mann-Whitney test was used. In all statistical tests a p value below 0.05 was considered as significant.

2.8.3. Statistical analysis for evaluation of role of the role of ongoing rhythm and the site of ablation

Gaseous and solid signal counts were summed (Total MES). As bilateral recording of MCA was not possible in all cases, the mean MES count was calculated, using either the mean of the bilateral counts when both sides were measured, or the unilateral data when only one side was available. To improve normality, signal count data were natural log-transformed.

Unadjusted comparisons of ablation factor categories in terms of continuous variables were based on Student's t tests or Wilcoxon's rank-sum tests, subject to normality assumptions being satisfied. Linear regression with robust standard errors based on the clustering of observations within patients was used to evaluate the effects of ablation parameters on mean MES count. Models were adjusted for total energy delivered and average temperature. Interaction terms were used to assess effect heterogeneity across levels of potential effect-modifying factors. Fixed effects were expressed as estimated differences in the log-transformed outcome, 95% confidence intervals, and p values. Model checking was based on inspection of the normality of residuals.

p values less than 0.05 were interpreted as indicating statistical significance. The statistical package Stata (StataCorp 2009. Stata Statistical Software: Release 11., College Station, TX: StataCorp LP) was used for statistical analysis.

3. Results

3.1. Phased radiofrequency ablation after initial Cryoballoon ablation

Between September 2008 and December 2010, a total of 87 consecutive patients were treated with CB ablation in our center for drug-resistant paroxysmal or persistent AF. 34 of the 87 (11 women, mean age 57 (SD:11) years) underwent a 'redo' procedure involving the use of phased RF and the PVAC.

In the course of initial CB ablation the mean procedure, fluoroscopy and ablation times were 159.4 (SD: 36.7), 33.7 (SD: 10.2) and 41.9 (SD: 14.3) min, respectively. After a mean of 2.6 (SD: 1.2) cryoenergy applications per vein, 116 (90.6%) of the 128 PVs were isolated. The mean temperature reached was -41.2 (SD:6.6) °C.

During the procedure, transient phrenic nerve palsy occurred in 3 (8%) and groin hematoma in 1 patient (3%).

During repeat procedure with phased RF and the PVAC, the mean time to repeat ablation after the first procedure was 13.7 (SD: 11.7) months. Electrical reconnection was observed in 80(68.9%) of the 116 previously isolated PVs, comprising 17 right superior PVs (51.5%), 11 right inferior PVs (33.3%), 22 left superior PVs (88%), 19 left inferior PVs (76%), 2 accessory PVs on the right side (100%), 1 right common ostium (100%) and 8 left common ostia (89%). The mean number of reconnected PVs per patient was 2.4±1. Electrical conduction was demonstrated in all 14 PVs which were not isolated successfully during the initial CB ablation.

The mean procedure, fluoroscopy and ablation times were 104.7 (SD: 38.5), 26.1(SD: 16.2) and 12.8(SD: 6.1) min, respectively. The mean numbers of RF applications were 14.06 (SD: 6.7) per patient and 4.4(SD: 3.2) per vein. Acute isolation was achieved in all PVs in all patients. No significant difference was found in the numbers of RF applications required for full isolation between the groups of patients who needed ≥ 4 versus those who needed ≤ 3 cryoapplications during the initial procedure($p=0.723$). Likewise, there was no significant difference was found in the rate of PV reconnection between the these two groups ($p=0.263$).

Apart from 2 cases of groin hematoma (treated conservatively) no significant periprocedural complication occurred.

Following a blanking period of 3 months, 27 (79%) of the 34 patients remained free of atrial arrhythmia during a mean 21.3 (SD:12) months after the repeat ablation. 25 of 26 patients who were free of arrhythmia recurrence 12 months after the redo ablation remained free of atrial arrhythmias during a mean follow-up of 26.4 (SD:8.7) months. Arrhythmia recurred within 1 year after PVAC ablation in 6 patients, and at 14 months in 1 patient.

The baseline clinical and procedural parameters, including age, sex, type of AF, LA short diameter, atrial flutter in the history, flutter ablation in the history, hypertension, heart failure, coronary disease, valvular disease, diabetes, previous electrical or pharmacological cardioversion, the procedure time, the fluoroscopy time and the number of RF applications, were not predictive of the clinical outcome after the redo procedure.

3.2. Transcranial measurement of cerebral microembolic signals during pulmonary vein isolation performed with single-shot techniques

A total of 89 patients were enrolled in the study. Regarding demographic and clinical parameters on the patients in the 5 treatment groups, there were no significant intergroup differences.

Procedural data are listed in Table 2. The total procedure, fluoroscopy, ED and LA access times demonstrated significant differences. The acute success rates were 100% in the 3 PVAC Groups, and 98% in both the CB and the nMARQ groups. The shortest LA access time was achieved in PVAC Group III, while the shortest ED time was obtained during ablation with the nMARQ. There were no significant differences between the intraprocedural ACT values. Symptomatic thromboembolic event did not occur in any patient.

The MES count detected with the CB was used as a reference for comparison with the other technologies. No significant difference was found between the CB group, PVAC Group II ($p=0.543$) and PVAC Group III ($p=0.317$) in the total cerebral MES counts. However significantly higher MES counts were demonstrated in PVAC Group I ($p=0.005$) and with the nMARQ technique ($p=0.007$). The ratios of gaseous versus solid MESs detected in the 5 treatment groups did not differ significantly ($p=0.688$).

A relatively even distribution of embolus formation was observed across the whole LA access time in the CB group, and in PVAC Groups II and III, whereas, microemboli were detected mostly during ED in PVAC Group III and with nMARQ.

3.3. Effect of the ongoing rhythm and the site of energy delivery on cerebral microembolization during phased radiofrequency ablation

Ablations of 48 consecutive patients assigned to PVI with the PVAC catheter were analyzed.

A total of 730 EDs were analyzed. 410 EDs were performed at the ostia of the left-sided PVs: 204 at the left superior PV (LSPV), 174 at the left inferior PV (LIPV), and 32 at left-

sided PVs with common ostia (LC). No intermediate PV was found on the left side. A total of 320 EDs were performed at the ostia of the right-sided PVs: 188 at the right superior PV (RSPV), 131 at the right inferior PV (RIPV) and 1 at the right intermediate PV (R Int. PV). No common ostia was found on the right side.

The MES count was significantly higher during ablations of the left- than of the right-sided PVs (mean MES count: 34.5 SD:48.8 vs. 19.5 SD:33.6; $p<0.0001$) No difference was found between the upper and the lower PVs (mean MES count: 31.7 SD:47.1 vs. 23.5 SD:39.6, respectively; adjusted $p=0.159$). Importantly, no difference was seen in the average temperature and average power between ablations of the left- and right-sided PVs. After adjustment for average temperature and total energy delivered the MES count remained significantly higher during the ablations of the left-sided PVs.

The temperature was significantly higher and the power significantly lower during ablations in AF than those in SR (52.8 SD:3.5 °C during ED in AF vs. 51.3 SD:3.3 °C during ablation in SR; $p<0.0001$) (5.6 SD:1.6 W vs. 6.2 SD:1.4 W, respectively unadjusted $p<0.0001$, robust unadjusted $p=0.0499$).

The relationship between the MES count and the ongoing rhythm was dependent on the temperature achieved during ED: while no difference in MES count was recorded during ED in AF versus SR when the temperature remained <56 °C, a significantly lower MES count in SR relative to that in AF was measured at temperatures >56 °C.

4. Discussion

4.1. Phased radiofrequency ablation for atrial fibrillation recurrence after cryoballoon ablation

Several studies have been published on repeat ablation for AF. The majority of these data refer to circumferential point-by-point RF ablation and the recurrence of arrhythmia was generally attributed to reconnection in some of the previously isolated PVs, due to electrically active gaps.

Atrial arrhythmia recurrence after a CB ablation has been reported in up to 50% of the cases. Similarly as after focal RF ablation, this recurrence has been ascribed to the electrical reconnection of the previously isolated PVs. Fürnkranz et al. demonstrated that the inferior segments of the PVs and the ridge between the LA appendage and the left-sided PVs are predilection sites of reconnection after CB procedures. Sharp catheter angulations with loss of

the central CB alignment required to reach inferior PVs have also been associated with a higher reconnection rate. In agreement with these observations, retrospective analysis¹³ of the shape and the angle of the PV ostia confirmed a significant relationship with the degree of occlusion achieved with the CB. Positioning of the CB with good circumferential tissue contact at the PV ostium, as indicated by no or only minimal leakage of contrast from the vein, is critical for the achievement of an effective lesion. This implies the limited ability of the pre-shaped CB device, currently available in 2 fixed sizes, to adapt to a variety of LA and PV anatomies. Although special maneuvers have been advocated to overcome these difficulties, CB energy applications need to be supplemented by focal (cryo or RF) lesions in order to achieve complete isolation in a significant proportion of PVs. As the reconnection of the PVs is related to a geometrical mismatch between the CB and the size, shape or angle of the PV ostium, it could be argued that the same technology may not be the optimal choice for a redo procedure. Data from randomized comparisons between different ablation techniques after CB are not available; only a few single-center studies have been performed with either CB or focal RF during the repeat procedure.

Our study is the first to assess the results of two single-shot techniques applied in sequence: CB as the initial procedure and PVAC ablation as a redo in the event of arrhythmia recurrence. We assumed that use of a different type of energy and a catheter with different maneuverability and a better or at least different ability to accommodate to the anatomy might be supplementary to the previous CB procedure. Our results seem to support this rationale: the difficulties encountered during CB ablation, as indicated by a relatively high number of energy applications to achieve complete PVI, were not predictive either of a higher number of recurrences in these PVs or of a higher number of RF sessions required for isolation with the PVAC. Furthermore, the procedure parameters and the long-term success achieved in our patients are similar to those reported following the use of focal RF, and compare favorably with the results of redo ablation with the CB.

There are important differences to consider when selecting a focal RF catheter versus the PVAC for a redo ablation. Focal RF ablation after previous PVI includes a careful mapping to find the sites of electrical gaps along the old ablation lines. This might well be time-consuming, though the known predilection sites of conduction recovery after CB ablation may facilitate the process. Once the gaps have been identified, a relatively low number of energy applications are required to close them. Important features of this technique includes double transseptal punctures, the use of a circular mapping catheter in the PVs and an electroanatomical mapping or navigation system. In our practice, mapping with the circular

PVAC was confined to the assessment of PV conduction, and RF energy was then applied to all electrodes demonstrating a good tissue contact around the circular catheter in order to close potential gaps and to consolidate the lesion around the PV ostia. Nonetheless, the number of RF pulses in our study (mean: 14) was less than the numbers reported during PVAC ablations carried out as an initial procedure (mean: 24-27).

It is an important point that both CB and PVAC ablations are single-shot techniques which share critical elements of the procedure and require similar operator skills. The over-the-wire design, the possibility to use the same steerable sheath, and to create circumferential lesions around the PV ostia with a few energy applications after appropriate positioning of the device are common features of the two techniques, while point-by-point ablation requires skills, experience and a learning curve that differs considerably. This could make PVAC ablation after a failed CB procedure an attractive and logical choice, especially for operators who have limited experience with focal RF ablation for AF.

4.2. Comparison of different single-shot ablation techniques: evaluation of safety

In reference to safety, major differences have been reported in the incidence of SCI after AF ablation with various technologies and with the different methods and criteria applied to evaluate cerebral lesions. The diversity of the periprocedural anticoagulation routine in the various studies further hampers a direct comparison of the different AF ablation techniques. In the present study, we compared different single-shot ablation tools designed for PVI without additional LA ablation (eg. creating long linear lesions, or ablating complex fractionated electrograms) in patients with similar baseline characteristics. Further, periprocedural anticoagulation was homogeneous across all treatment groups with uninterrupted VKA administration and therapeutic INR level before the procedure and iv heparinization to a target ACT above 300 s in agreement with current recommendations. Any differences demonstrated in the number of microemboli are therefore truly inherent to the ablation techniques compared.

The rate of microembolization during phased RF ablation with the implementation of specific procedural modifications and the use of consecutive software versions of the GENius generator demonstrated a significant decrease in our study. The very same trend was observed in recent DW MRI studies on the rate of new SCI lesions after phased RF ablation. Importantly, no difference was detected in the level of microembolization between the procedure with CB ablation versus that with the PVAC after

procedural and software changes had been implemented (PVAC Groups II and III). In contrast, significantly higher MES counts were recorded during phased RF ablation with the use of the old technique (PVAC Group I) and during ablation with nMARQ.

The significant reduction in microembolization with phased RF was clearly related to the elimination of the MESs during the ED period of the procedure. Similarly to as observed in PVAC Group I, the majority of the microemboli were generated during ED with nMARQ, which also represents a multielectrode RF ablation technology. It is noteworthy that MESs were largely gaseous in nature in all treatment groups.

4.3. Measures related to the reduction in cerebral embolization with phased RF ablation

The significant reduction achieved in clinically silent microembolization, as demonstrated by DW MRI studies and our own results were related to refinements in specific technical elements of the phased RF ablation, based on preclinical data. The observation of enhanced microembolization with blended unipolar:bipolar energy delivered through PVAC electrodes in close proximity to each other was of utmost importance. While this problem remained unknown, this was a likely common scenario, owing to the squeezed situation of the distal PVAC loop. It should be noted that actual physical contact between any two electrodes results in an electrical short circuit and immediate termination of the RF delivery. On the other hand, not contact, but a reduced interelectrode space (less than the fixed 3 mm between two neighboring electrodes mounted on the distal circular segment of the PVAC) may lead to blood and tissue overheating, due to a high current density. Simple measures to avoid this possibility include a careful fluoroscopic assessment of the electrode positions (as we did in PVAC Group II), exclusion of simultaneous RF ED to electrodes 1 and 10, as proposed by several authors and a software modification implemented in the latest version of the GENius generator, which supports simultaneous RF delivery to a maximum of 9 electrodes (software version 15.0; PVAC Group III in our study).

The earlier practice of starting ED in a 4:1 bipolar:unipolar mode at each PV was changed to start with a 2:1 ratio in PVAC Groups II and III. The rationale for this change was to avoid extensive bipolar ED and thereby reduce local heating between adjacent electrodes. Although animal data suggested a tendency for more microemboli with more bipolar RF delivery, the contribution of this change in the reduction of microemboli is uncertain. Nonetheless, starting with the 2:1 instead of the 4:1 ratio is the current practice at other centers.

As demonstrated in an animal model, a potential source of gaseous emboli was the introduction of air into the LA via the transseptal sheath. A significantly larger air volume was measured during the introduction of the PVAC, possibly owing to its more complex shape as compared with a conventional focal RF catheter. To prevent air entrapment around the array, the PVAC was submerged and captured by the introduction device in a saline bath prior to insertion into the FlexCath sheath.

It is noteworthy that, in addition to the procedural changes as described above, the GENius RF generator has also been modified in recent years. An improved energy titration algorithm was implemented starting at version 14.4 (PVAC Group II), which regulates the power delivery to control the maximum, instead of the average temperature, with a target of 60 °C. Further, this software ensures a gradual and limited increase (4 W/s) during variable or intermittent electrode-tissue contact, to avoid high temperature peaks. The importance of the catheter-tissue contact has been highlighted since the introduction of contact force measurement for focal RF ablation catheters. With multipolar ablation, this becomes even more critical, as the maintenance of good contact simultaneously on multiple electrodes can be challenging and the capability of direct contact force measurement with this technology is not yet available. This concept is supported by our previous study: with an analysis of temperature and power data obtained at high resolution sampling from the GENius generator, increased microembolus formation was demonstrated during intermittent contact scenarios when low temperature was compensated by increased power which resulted in a temperature overshoot when the contact was re-established. The latest issue of the GENius software (15.0; PVAC Group III) features the energy titration algorithm of version 14.4, and in addition, RF delivery is limited to 9 electrodes, thereby providing a definite solution for the problem of E1-E10 interaction.

4.4. Microembolization during ablations with the Cryoballoon and the nMARQ technique

The use of cryoenergy is generally regarded as a more “tissue-friendly” and safer ablation technology, which is associated with a significantly lower incidence of thrombus formation as compared with RF ablation. The limited data available on microembolus generation with CB ablation demonstrated consistent results. The total MES count during CB ablation in the present study (1057 SD: 784) was reasonably similar (834 SD: 727) to that recorded in our

previous work, and to that reported by Sauren et al. (935 SD: 463). Importantly, we also demonstrated that the microemboli were predominantly gaseous, with an even distribution throughout the LA access period. It should be noted that results obtained on SCI with different AF ablation technologies as evaluated with DW MRI are in agreement with these TCD data.

Although the results obtained with the nMARQ ablation system reflect our initial experience, we found this technology to be very effective with the shortest ED time required for acutely successful PVI in this study. However, this technology was also associated with a high rate of microembolization, of the magnitude of that initially achieved with phased RF (PVAC Group I), and significantly higher than the numbers obtained with modified phased RF (PVAC Groups II and III) and with CB. Recently published results of a German center [32] seem to support our findings: DW MRI demonstrated SCI acutely in 14 (33%) out of 43 patients after PVI with the nMARQ catheter, a ratio similar to the values up to 39% [2,6] in earlier reports on phased RF ablation.

The high microembolization rate was somewhat unexpected after the earlier experience demonstrating lower incidence of SCI and lower microembolus counts with focal irrigated as compared with conventional RF ablation. As a potential explanation, one speculative possibility is the irrigation itself used with the nMARQ catheter, with a very high rate of 60 ml/min during ED. Direct injection of saline into the LA at this rate is known to result in marked bubble formation due to cavitation, which is readily visible on ICE (as we observed in cases when ICE was used during ablations). This mechanism could be responsible for a currently unknown proportion of cerebral microemboli. These microemboli are gaseous, which are generally considered to be less harmful than solid particles, although their significance in the long-term cognitive function is yet to be determined. Other possible causes are likely to be inherent to multielectrode ablation and the high total energy delivered when this catheter is used. By analyzing biophysical parameters during phased RF ablation, our group earlier demonstrated that microemboli generation was related to both the number of active electrodes during ED and the total energy delivered. Further, maintenance of a good contact on multiple poles throughout the entire duration of RF applications might be challenging, and power handling in intermittent contact scenarios could therefore be of significance. There is an urgent need for more data on embolization rates and in-depth investigations into the mechanism with the nMARQ system with the aim of making this otherwise promising technology safer.

4.5. Effect of the ablation site and ongoing rhythm on the MES count

As far as we are aware, this is the first reported demonstration of significant differences in the rate of microembolization according to the site of the ablation. EDs at the left-sided PVs resulted in significantly higher number of MESs as compared with those at the right-sided PVs. Further, the ongoing rhythm during ablation was also found to be a significant predictor of microembolus formation. At temperatures > 56 °C during ablation, significantly lower numbers of MESs were recorded in SR than in AF, whereas no difference was observed at lower temperatures.

Since the first reports were published on the increased risk of SCI specifically related to phased RF ablation, variable mechanisms potentially responsible for this increased thrombogenic potential have been investigated. To date, the following ablation parameters have been identified as contributing to MES formation during PVAC ablations: the interaction between the proximal (E1) and distal (E10) PVAC electrodes, the ablation mode, and the number of enabled electrodes during the ED. Moreover, our group has demonstrated that different variables related to the electrode-tissue contact during ED are also significant predictors of microembolization with this technology. The present study provides additional information concerning the mechanism of microembolus generation during ablation with the PVAC by assessing the roles of the ablation site and the ongoing rhythm.

The more complex anatomy of the left-sided PVs has been recognized as having important implications for transcatheter PVI: the ridge between the left superior PV and the LAA is a known predilection site for both early and late electrical reconnection after either focal RF or a balloon-based ablation. This is probably related to the increased tissue thickness of the ridge, and the difficulties of maintaining a stable catheter-tissue contact at this location with a single-electrode RF ablation catheter, which is even more challenging when a multipolar catheter is used and RF is delivered simultaneously to multiple electrodes. It therefore seems reasonable to assume that our findings of a significantly higher MES count during ablation of the left as compared with that of the right PVs may reflect suboptimal contact, which is a known mechanism of microembolization. Further, the vicinity of the LAA itself, providing a high local flow, may play a significant role in microembolization. In vitro studies with the PVAC demonstrated very high microbubble generation when the electrodes on the ridge were not in proper contact with the tissue and the thermocouples in these electrodes facing the orifice of the LAA were cooled by the flow from the LAA (unpublished

results). It could also be postulated that, while some of the electrodes on the ridge make insufficient contact, others may have too much contact force. This appears to be plausible, especially at the left superior PV, as the direction of the catheter and the sheaths are lined up with the axis of force from the puncture site at the right femoral vein. A higher than optimal force could also result in tissue overheating. These considerations all argue in favor of implementing contact force technology capable of measuring the force separately in all electrodes.

We additionally demonstrated that the ongoing atrial rhythm is also of significance for microembolus generation during ED with the PVAC. It was reasonable to assume that a more stable catheter position can be achieved due to the missing atrial contraction in AF as compared with SR. The significantly higher temperature and lower power attained in AF appeared to support this speculation. On the other hand, the better contact during AF was expected to result in a lower MES count, though this was not confirmed by our results. In fact, similar MES counts were detected, regardless of the ongoing rhythm when the temperature stayed $<56^{\circ}\text{C}$, while the number of microemboli was lower at higher temperature in SR. Our interpretation of these phenomena is that the temperature recorded in AF is not a reliable indicator of the contact, and higher values may rather reflect diminished cooling due to the reduced blood flow with the loss of atrial contraction. On the other hand, the higher temperature reached in SR is a more valid indicator of sufficient tissue contact, which explains the lower MES counts. These data also stress the importance of the implementation of contact force measurement technology in RF catheters used for AF ablation.

Although recorded during phased RF ablation with the PVAC, the implications from our data might be extrapolated to other RF ablation technologies, including multipolar or conventional catheters with or without irrigation. To evaluate the clinical relevance of our results longitudinal assessment of the cognitive function seems justified in these patients. These measurements are performed in our Institute.

4.6. Summary of our results

Our research investigating the long-term success after consecutive use of two different single-shot ablation techniques and cerebral microembolization during pulmonary vein isolation performed with the Cryoballoon, phased-RF and the PVAC or nMARQ was given in this thesis. Our new observations:

Repeat ablation involving the use of phased-RF and the PVAC following CB ablation in cases of AF recurrence is an effective procedure. Long-term clinical success was achieved in 79% of patients at a mean 21-month follow-up.

Microembolus counts were reduced to a level comparable with that observed during CB ablation, when phased RF ablation was implemented with specific procedural modifications with the use of newer generations of the GENius RF generator.

A significantly higher microembolization rate, similar to that recorded with the first version of the GENius generator, was demonstrated with the multipolar irrigated nMARQ RF ablation system.

Both the ongoing rhythm during the ED and the site of ablation influence microembolus generation during PVAC ablation procedures. The higher rate of microembolization during ablation of the left-sided veins and during atrial fibrillation even with temperatures close to the target value, suggest improper tissue contact during energy delivery. This underscores the importance of evaluating the contact during and preferably before energy deliveries.

5. In extenso publication of the author



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Register number: DEENKÉTK/175/2014.
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Candidate: Alexandra Kiss
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List of publications related to the dissertation

1. **Kiss, A.**, Nagy-Baló, E., Hajas, O., Édes, I., Csanádi, Z.: Phased radiofrequency ablation for atrial fibrillation recurrence after cryoballoon ablation.
Exp. Clin. Cardiol. 20 (1), 2438-2443, 2014.
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2. Nagy-Baló, E., **Kiss, A.**, Condie, C., Stewart, M., Édes, I., Csanádi, Z.: Predictors of cerebral microembolization during phased radiofrequency ablation of atrial fibrillation: Role of the ongoing rhythm and the site of energy delivery.
Pacing Clin. Electrophysiol. Epub ahead of print (2014)
IF:1.25 (2013)
3. **Kiss, A.**, Nagy-Baló, E., Sándorfi, G., Édes, I., Csanádi, Z.: Cerebral microembolization during atrial fibrillation ablation: Comparison of different single-shot ablation techniques.
Int. J. Cardiol. 174 (2), 276-281, 2014.
IF:6.175 (2013)





List of other publications

4. Nagy-Baló, E., **Kiss, A.**, Condie, C., Stewart, M., Édes, I., Csanádi, Z.: Predictors of cerebral microembolization during phased radiofrequency ablation of atrial fibrillation: Analysis of biophysical parameters from the ablation generator.
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