Safety and Feasibility of Closed Chest Epicardial Catheter Ablation Using Electroporation

Kars Neven, MD, PhD; Vincent van Driel, MD; Harry van Wessel, BSc; René van Es, MSc; Bastiaan du Pré, MD; Pieter A. Doevendans, MD, PhD; Fred Wittkampf, PhD

Background—Permanent coronary artery damage is a hazardous complication of epicardial radiofrequency ablation. Irreversible electroporation (IRE) is a promising nonthermal ablation modality able to create deep myocardial lesions. We investigated the effects of epicardial IRE on luminal coronary artery diameter and lesion depth.

Methods and Results—In 5 pigs (60–75 kg), the pericardium was exposed using surgical subxiphoidal epicardial access. A custom deflectable octopolar 12-mm circular catheter with 2-mm ring electrodes was introduced in the pericardium via a steerable sheath. After coronary angiography (CAG), the proximal, mid, and distal left anterior descending, and circumflex coronary arteries were targeted with a single, cathodal 200 J application. CAG was repeated after IRE and after 3 months follow-up. Using quantitative CAG, the minimal luminal diameter at the lesion site was compared with the average of the diameters just proximal and distal to that lesion. Intimal hyperplasia and lesion size were measured histologically. CAG directly postablation demonstrated short-lasting luminal narrowing with normalization in the targeted area, suggestive of coronary spasm. After 3 months, all CAGs were identical to preablation CAGs: mean reference luminal diameter was 2.2±0.3 mm, mean luminal diameter at the lesion site was 2.1±0.3 mm (P=0.35). Average intimal hyperplasia in all arteries was 2±4%. Median lesion depth was 6.4±2.6 mm.

Conclusions—Luminal coronary artery diameter remained unaffected 3 months after epicardial IRE, purposely targeting the coronary arteries. IRE can create deep lesions and is a safe modality for catheter ablation on or near coronary arteries. (Circ Arrhythm Electrophysiol. 2014;7:913-919.)

Key Words: catheter ablation ■ coronary arteries ■ electroporation ■ pericardium ■ safety

Before radiofrequency catheter ablation was introduced in cardiac electrophysiology in the late 1980s, direct current catheter ablation was used to treat cardiac arrhythmias. This method caused severe and hazardous adverse effects by creation of an electrically isolating vapor globe. This led to a spark (arching), an explosion, and a pressure wave.1 During the past 20 years, radiofrequency catheter ablation has become the standard ablation technique for cardiac arrhythmias.2 Radiofrequency causes heat damage to all tissue near the ablation site. Ablation near coronary arteries can, therefore, have hazardous adverse effects such as coagulation of blood inside the vessel and vessel stenosis with subsequent myocardial infarction.3-5 In addition, the cooling effect of arterial and endocardial blood flow may limit lesion formation and success of the procedure.6,7

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Recently, Lavee et al,3 Hong et al,9 and Wittkamp et al10 published feasibility of epicardial nonthermal electroporation ablation of myocardial tissue, demonstrating that an energy level lower than the arcing threshold could successfully create large myocardial lesions without hazardous adverse effects. du Pré et al11 showed that epicardial electroporation ablation over coronary arteries, with a follow-up of 3 weeks, has a low risk of coronary damage and that the use of this technique near or even on large coronary arteries is relatively safe. In addition, electroporation ablation did not seem to be affected by the presence of arterial blood flow.

The purpose of the present study was to investigate the safety and feasibility of catheter ablation using electroporation in the pericardial space. More specifically, the long-term (3 months) effects of electroporation ablation on the coronary arteries (safety) and the ability to create myocardial lesions by electroporation ablation from the pericardium (feasibility) were subject to investigation.

Methods

All studies were performed after prior approval from the Animal Experimentation Committee of the University Medical Center Utrecht, Utrecht, The Netherlands, and were performed in...
The study was performed in 6 pigs (weight 60–75 kg). Amiodarone therapy was started 1 week before the index procedure (400 mg once daily) to prevent procedure-related arrhythmias. Carbasalate calcium (80 mg once daily) and clopidogrel (75 mg once daily) therapy was started 1 week before the index procedure (400 mg once daily) to prevent procedure-related arrhythmias. Carbasalate calcium and clopidogrel were continued throughout the index procedure. Carbasalate calcium and clopidogrel therapy was continued after the index procedure. Carbasalate calcium and clopidogrel therapy was started 1 week after the index procedure (400 mg once daily) to prevent procedure-related arrhythmias. Carbasalate calcium and clopidogrel therapy was continued throughout the index procedure. Carbasalate calcium and clopidogrel therapy was started 1 week after the index procedure (400 mg once daily) to prevent procedure-related arrhythmias.

**Measurement of Coronary Diameters**

Luminal diameters of the coronary artery, proximal and distal to the application site, and the minimal diameter at the application site were measured using quantitative CAG. The latter value was then compared with the average value of the diameters proximal and distal to the application site.

**Histological Evaluation**

After fixation, multiple 3- to 4-mm-thick segments were dissected from each lesion to facilitate measurement of lesion width and depth. All sections were taken perpendicular to the epicardial surface and to the main course of the targeted coronary artery. Paraffin-embedded segments were sectioned and stained with H&E and elastic–van Gieson. All histological sections were scanned with a ScanScope XT scanner (Aperio Technologies, Inc, Vista, CA) and analyzed using Imagescope (Aperio Technologies). Lesion depth was measured in each section. Of all coronary arteries and branches, the luminal area, the area encompassed by the internal elastic lamina (IEL area), and the area encompassed by the external elastic lamina (EEL area) were measured. The intimal area was calculated by subtracting the luminal area from the IEL area. All arteries with an EEL area >0.15 mm² were considered clinically relevant and were included in the study. Coronary damage was defined as intimal hyperplasia, and percentage stenosis because of intimal hyperplasia was calculated as follows: (IEL area–luminal area)/IEL area×100. From these data, the median values of each lesion were calculated. Subsequently, the mean value of all medians was calculated.

**Measurement of Lesion Depth**

Lesion depth was measured in each histological section. Large lesions often showed tissue shrinkage as also seen after myocardial infarction. When sufficient undamaged myocardium was present in the histological section, the estimated original epicardial contour was used to measure lesion depth. In case the lesion was transmural, the estimated original endocardial contour was used to measure lesion depth. From these data, the median depth of each lesion was calculated. Subsequently, the mean value of all median lesion depths was calculated. An ablation lesion was considered to be transmural when transmurality was observed in at least 2 consecutive histological sections.

**Statistical Analysis**

Differences in lesion depth and coronary artery luminal diameters were examined with a Wilcoxon signed rank test. These analyses are lesion-based and not pig-based. Continuous variables were expressed as mean±SD. Statistical significance was defined as P≤0.05 (2-sided).

![Figure 1. Circular electroporation ablation catheter. The custom circular electroporation ablation catheter that was used for this safety and feasibility study. The distal circular, 12-mm diameter segment of the deflectable 7F catheter contains 8 electrodes 2 mm in length.](http://circep.ahajournals.org/)

![Figure 2. Coronary angiography (CAG) before and after catheter ablation. CAG before (A), directly after (B), and 3 mo after (C) electroporation catheter ablation using a steerable sheath. Note the coronary spasm of the left anterior descending artery (LAD) after electroporation ablation over the LAD (arrow in B) and complete normalization after 3 mo (C).](http://circep.ahajournals.org/)
Results

Five animals survived the index procedure and the 3-month follow-up without complications.

Acute Death

One animal suddenly developed cyanosis with prolonged, untreatable hemodynamical instability after the end of the index procedure, ≈7 hours after the last ablation. This animal was euthanized acutely.

The procedure of the animal that died was complicated by multiple episodes of catheter-induced, hemodynamically unstable ventricular tachycardia requiring acute electrocardioversion. This already happened before the first electroperoration application, despite the pretreatment with amiodarone to prevent occurrence of tachyarrhythmias as much as possible. After each electrocardioversion, the animal was allowed to recover for 20 to 30 minutes. This was uneventful. After the 3 planned electroperoration applications (following protocol), the animal was hemodynamically stable. There were no signs of upcoming complications. Seven hours after the last application, after the end of the procedure, the animal suddenly developed cyanosis and severe dyspnoe. Despite basic emergency medical care, the clinical situation did not stabilize. Therefore, the animal was euthanized following protocol.

Although there was no ECG monitoring anymore at the time the adverse event happened, we suspect that the animal had again developed a sustained, hemodynamically unstable ventricular tachycardia. At autopsy, no pericardial effusion or trauma other than the ablation lesions was found. Gross inspection of other organs also showed no abnormalities.

Pericardial Ablation

In the 5 surviving animals, a total of 13 (median, 3; range, 2–3) 200 J electroperoration applications were delivered over the LAD and RCx arteries. No arcing or barotrauma was seen during any of the applications. Frequently, noise on 1 or several local bipolar electrograms suggested the presence of air in the pericardial space between the pericardium and the epicardium, possibly because of the pericardial incision and the supine position of the animal (Figure 3).

Coronary Angiography

All preablation coronary angiograms were normal. CAG postablation demonstrated short-lasting (<30 minutes) luminal narrowing with subsequent normalization in the targeted area, suggestive of coronary spasm. After 3-month survival, all coronary angiograms were identical to the preablation coronary angiograms.

Table. Lesion Depth, Coronary Artery Diameter, and Patency

<table>
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<tr>
<th>Pig #</th>
<th>Median Lesion Depth, mm</th>
<th>Transmurality of Lesion</th>
<th>Coronary artery</th>
<th>Proximal Reference Diameter, mm</th>
<th>Distal Reference Diameter, mm</th>
<th>Minimal Diameter in Lesion, mm</th>
<th>Median Stenosis, %</th>
<th>Maximal Stenosis, %</th>
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Results per lesion after 3 mo follow-up. Median stenosis (%) is median value of luminal stenosis in all sections of the respective lesion. Maximal stenosis (%) is the absolute value of the maximal luminal stenosis in all sections of the respective lesion. In the cross-sections of pig 2, lesion 3, no arteries were visible. The angiographic caliber of the coronary artery was too small to analyze for the third electroperoration application of pig 2. Therefore, no information about coronary diameters was available for this application. LAD indicates left anterior descending artery; NA, not available; and RCx, circumflex coronary artery.
angiograms: mean reference luminal diameter was 2.2±0.3 mm, mean luminal diameter at the lesion site was 2.1±0.3 mm ($P=0.35$; Figure 2 and Table).

**Acute Epicardial Lesions**

Although 1 animal died shortly after the end of the procedure, already the four 7-hour-old lesions on the epicardium could clearly be identified. Next to the imprint of the separate electrodes of the 12-mm circular ablation catheter, a ≈22-mm-wide whitish coloration of the surrounding epicardium could be identified (Figure 4).

**Macroscopic Findings**

Careful inspection of the organs adjacent to the pericardium showed no abnormalities in any animal. No macroscopic signs of bleeding, scarring, or excessive fibrotic tissue proliferation were found.

**Lesion Depth and Coronary Arteries**

In 5 animals, 104 cross-sections from 13 electroporation lesions, with a median of 9 (range 2–10) cross-sections per lesion, were analyzed. Using 200 J electroporation applications, there was no central surviving area of myocardial tissue visible in any lesion. Transmurality of the ablation lesion was seen in 4 of 13 (31%) lesions and significant shrinkage because of scar contracture was obvious. Mean value of the median lesion depths was 6.4±2.6 mm (range, 0.0–10.4 mm; Table). Arterial branches were predominantly located epicardially, very close to the application site. A total of 167 arterial branches with an EEL area >0.15 mm$^2$ were found. These arteries were divided into 154 arterial sections that were surrounded by a lesion and 13 that were located outside a lesion. None of the arteries inside the lesion was surrounded by intact myocardial tissue.

Intimal hyperplasia was observed in 66 of 154 arteries inside lesions and in 1 of 13 arteries outside lesions (Figure 5). The single affected artery outside a lesion had an EEL area of 0.99 mm$^2$. This artery was located 1.6 mm from the lesion border and was surrounded by several smaller and larger unaffected arteries. Mean value of median luminal stenosis in all arteries was 2±4% (range, 0–61%), whereas mean value of median luminal stenosis of affected arteries...
was 8±5% (range, 1–61%; Table). Arteries with intimal hyperplasia located inside a lesion were similar in size to arteries inside a lesion without intimal hyperplasia (mean EEL area of 1.04±0.73 versus 1.01±0.81 mm², respectively; P=0.86). Lesion depth measured in cross-sections with arteries showing intimal hyperplasia was greater than lesion depth in cross-sections with arteries showing no intimal hyperplasia (6.9±2.7 versus 4.4±3.3 mm, respectively; P<0.0001).

Discussion
This is the first study investigating the effect of epicardial irreversible electroporation by an ablation catheter placed in the pericardial space.

In recent years, the possibilities and number of catheter ablation procedures of cardiac arrhythmias have skyrocketed. Endocardial ablation of ventricular arrhythmias is performed in a large number of centers, but some ventricular arrhythmias can better be ablated from the epicardial side. An epicardial approach, however, is associated with severe complications like cardiac tamponade, ventricular arrhythmias, phrenic nerve damage, and damage to the coronary arteries. Based on available data and experience, a distance of >5 mm between the ablation catheter and an epicardial artery is commonly recommended when radiofrequency ablation is considered.

In this study, we simulated epicardial ventricular catheter ablation in humans. Because of our inexperience with porcine pericardial puncture and to minimize occurrence of periprocedural complications, such as cardiac tamponade, we made a small (<10 mm) pericardial window to obtain pericardial access.

Acute Outcome
One animal died because of unstable ventricular tachycardia ≈7 hours after ablation. Unfortunately, pigs are susceptible to developing hemodynamically unstable ventricular arrhythmias and the success rate of resuscitation of a pig after prolonged unstable ventricular tachycardia is known to be disappointing. Visual inspection of the epicardium clearly revealed epicardial lesions, ≈22 mm in diameter despite a only 12-mm circular ablation catheter. This raises the question how fast the effect of electroporation ablation takes in. In a study by Hong et al., sheep hearts were ablated with electroporation. They proved conduction block directly after ablation. With a maturation period in the animal before euthanization as short as 1 hour, the lesions were in general not visible grossly, but could be detected via histology on lesion cross-sections.

Pericardial Ablation
No complications occurred during or after pericardial access. With a deflectable sheath, the steerable ablation catheter could easily be moved toward target areas. The pericardial window resulted in a layer of air in the pericardial space in some animals. Therefore, tissue contact of the ablation catheter may sometimes have been suboptimal.

Coronary Angiography
In this study we purposely targeted the main coronary arteries. Apart from short-lasting (<30 minutes) coronary spasm, no long-term luminal narrowing was seen; after 3 months follow-up, the luminal diameters of the main coronary arteries were identical to the baseline luminal diameters. This suggests that the patency of the main coronary arteries is not affected by irreversible electroporation. These data support the findings of du Pré et al. who found similar results after a shorter follow-up period of only 3 weeks.

Lesion Size and Coronary Arteries
One of the limiting factors of conventional catheter ablation is the inability to create transmural left ventricular lesions. With epicardial electroporation ablation, transmural lesions were easily created. These deep lesions did not come at the cost of major damage to the coronary arteries. Intimal hyperplasia was observed in 67 of 167 arteries. Mean values of median luminal stenosis in all arteries were 2±4%; mean values of median luminal stenosis of the arteries showing any intimal hyperplasia were 8±5%. There were no occluded main arteries. These results are again in line with the findings of du Pré et al. who found similar results after a follow-up period of only 3 weeks.

This could be a major breakthrough in the treatment of epicardial ventricular arrhythmias, because no other ablation technique can create deep myocardial lesions very close to or on top of the coronary arteries without causing significant damage to them.

Limitations
We used only 1 energy setting (200 J). From a previous study, we know that this energy setting is able to create wide and deep lesions. This was also seen in the current study: up to 22 mm wide and 11 mm deep lesions resulted from a single ablation using a 12-mm circular ablation catheter (Figure 6). These large lesions may have a negative effect on total myocardial contractility and ejection fraction, especially when placed at multiple different locations. A possibly proarrhythmic effect of these lesions through creation of a substrate should also be addressed in future studies.

Significant shrinkage because of scar contracture of the 200 J lesions definitively caused underestimation of lesion size after 3 months follow-up.

Lesion size with electroporation ablation will depend on the ultimate catheter design and measures to ensure electrode–tissue contact.

Because of the pericardial window, air entered the pericardial space, and this may have caused suboptimal contact between the ablation catheter and the left ventricular epicardium. Future studies on epicardial electroporation catheter ablation should be performed after subxiphoidal puncture of the pericardium, thereby minimizing the risk of air entrapment in the pericardial space.

In the current study, we used ≈6-week-old pigs, which did not show much epicardial fat at autopsy. In humans, there can be a thick layer of epicardial fat, especially at the basal part of
the ventricles and over the interventricular groove. The influence of (the amount of) epicardial fat on myocardial lesion depth created with electroporation ablation has to be investigated in future studies.

The coronary arteries showed short-lasting (<30 minutes) spasm after electroporation catheter ablation directly on the coronary arteries. It is known that pretreatment with vasodilators can prevent or decrease the occurrence of arterial spasm after insertion of a sheath in the radial artery. In this study, the coronary arteries were not pretreated with vasodilators. Pretreatment of the coronary arteries with vasodilators should be investigated in future studies.

Apart from damage to the coronary arteries, phrenic nerve damage is another possible complication during conventional epicardial ablation. In this study, we did not investigate the effects of electroporation catheter ablation on phrenic nerve function. Additional studies have to investigate whether or not electroporation catheter ablation affects phrenic nerve function.

In this study, the ablation catheter was placed on the left ventricular epicardium. Although we might expect a similar outcome, we do not have information about lesion size or adverse events created by epicardial electroporation ablation in atrial tissue.

Conclusions
The data of this study demonstrate that epicardial catheter ablation using electroporation can create extensive and deep myocardial lesions without significant damage to the coronary arteries after a 3-month follow-up. This effective new ablation technique could possibly solve one of the most important current limitations of epicardial catheter ablation: safe ablation on or near main coronary arteries.

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We thank the staff of the Department of Experimental Cardiology of the University Medical Center Utrecht for technical assistance during the experiments; Aryan Vink, MD, PhD, of the Department of Pathology, University Medical Center Utrecht, for support during analyses of the cross-sections; and Paul Westers, PhD, of the Department of Biostatistics, University Medical Center Utrecht, for assistance with the statistical analyses.

Disclosures
Dr Wittkampf is a consultant for St Jude Medical, Atrial Fibrillation division. Both Dr Wittkampf and H. van Wessel are coinventors of circular electroporation. The other authors report no conflicts.

References


**CLINICAL PERSPECTIVE**

Despite the use of advanced 3-dimensional mapping systems and coronary angiography, coronary artery damage remains one of the potential complications of epicardial ablation using radiofrequency ablation. Not seldomly, the close proximity of the targeted ablation site to a coronary artery forces the investigator to refrain from epicardial ablation. Electroporation ablation is a novel ablation technique. It has been proven that one 6 ms, 200 J electroporation application can create deep and wide myocardial lesions and can isolate a pulmonary vein. However, the safety and efficacy of epicardial electroporation ablation still is unknown. In this porcine model, after surgical subxiphoid access, epicardial electroporation ablation was performed purposely targeting the coronary arteries. Luminal coronary artery diameter remained unaffected 3 months after epicardial electroporation ablation. We demonstrated that epicardial electroporation ablation is able to create deep lesions. We also demonstrated that the coronary arteries are spared from electroporation ablation directly on the coronary arteries. The clinical implementation of subxiphoid epicardial electroporation ablation as a novel, safe, effective and very fast ablation technique could therefore be performed safely, without the risk of damage to the coronary arteries.
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The figure legends for Figures 3 and 4 were erroneously switched.

The compositor apologizes for the error.

The online version of the article has been corrected.
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