Journal of Interventional Cardiac Electrophysiology (2019) 54:109–118
https://doi.org/10.1007/s10840-018-0452-x

Parameters associated with acute morphometric lesion dimensions created by cryocatheters

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Received: 13 April 2018 / Accepted: 7 September 2018 / Published online: 25 September 2018

Abstract

Purpose Despite the wide use of cryoenergy, there is a paucity of data regarding the impact of certain ablation parameters on lesion size. Specifically, this study sought to evaluate the impact of catheter type, ablation time, heat load, and tip orientation on lesion dimensions using a porcine thigh model with focal cryoablation catheters.

Methods In 6 pigs, 251 lesions were created on thigh muscle with parameter permutations to compare the acute impact of catheter type (electrode tip sizes 4, 6, and 8 mm), ablation time (2, 2 × 2, 3, 4, and 2 × 4 min), heat load (1 and 2 L/min), and tip orientation (perpendicular or parallel) on lesion dimensions (length, depth, and cross-sectional area) immediately post-ablation. As a sub-study to evaluate the importance of tissue contact during the cryoablation procedure, a 1-min freeze was performed without tissue contact until an ice ball formed, followed by an additional 2–3 min freeze.

Results The linear regression model revealed that catheter type ($p < 0.0001$) and the interaction between catheter orientation and catheter type ($p = 0.027$) were significantly associated with lesion cross-sectional area. Lesion length and depth, but not cross-sectional area, are significantly impacted by the catheter type ($p < 0.0001$; $p = 0.003$) and orientation ($p < 0.0001$; $p < 0.0001$), respectively. Compared to parallel catheter placement, lesions created with the perpendicular orientation were deeper using 4-mm ($p = 0.136$), 6-mm ($p = 0.005$), and 8-mm tip catheter ($p = 0.004$). Lesion creation with an ice ball significantly reduced lesion depth compared to lesions made without an ice ball ($p < 0.05$). In contrast, ablation time ($p = 0.097$) and heat load ($p = 0.467$) were not significantly associated with lesion size. Additionally, there was no statistical significant difference in lesion size between 2 × 2 and 4 min ablation times.

Conclusions The present study demonstrated that lesion size was significantly impacted by catheter type and catheter tip orientation and that maintaining tissue contact prior to applying cryoenergy is essential.

Keywords Cryoablation · Cryothermal energy · Cryoenergy · Catheter ablation

Abbreviation

TTC 2,3,5-Triphenyltetrazolium chloride

1 Introduction

Over the past 30 years, radiofrequency has been the main energy source to treat cardiac arrhythmias and has been studied extensively. The advent of cryoenergy for cardiac ablation is relatively new and has recently gained momentum as a safe and effective energy source to treat patients. An advantage of using cryoenergy is the ability to predict the outcome of a lesion before irreversible damage is done [1]. However, the use of cryoenergy for ablative purposes is a highly complex process, and many factors play a role in the efficacy of lesion formation [2]. For example, selection of the appropriate catheter tip size was shown to be essential for creating desired lesion sizes in myocardial tissue in dogs [3]. Further basic research is still required to better understand the impact of specific ablation-related parameters on lesion size. The purpose of this study was to evaluate lesion sizes created using different cryoablation catheter types (4-, 6-, and 8-mm tips) and parameters (ablation time, tip orientation, and heat load) to advance knowledge to generate specific lesion sizes for different targets in cryoablation.

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2 Methods

2.1 Animal preparation and apparatus setup

Six female Yorkshire pigs (age 4.1–5.1 months, 70.2–81.4 kg) underwent acute thigh prep procedures as described in Tse et al. [4].

The pig specimens were housed and received humane care in accordance with the Guide for Care and Use of Laboratory Animals and the Animal Welfare Act as amended. The study protocol was reviewed and approved by the Medtronic PRL IACUC (Institutional Animal Care and Use Committee). The pigs were pre-medicated with a cocktail of xylazine (2–3 mg/kg), butorphanol (0.3–0.4 mg/kg), and midazolam (0.3–0.4 mg/kg). Buprenorphine (0.6–0.9 mg) was given an as analgesic, and isoflurane (1–3%) was used for induction and maintenance of general anesthesia. The pigs were intubated and ventilated with 100% oxygen. Succinylcholine (40–80 mg) was given as needed to reduce muscle spasm. Heparin (300 units/kg) was administered 5 min prior to euthanasia via pentobarbital euthanasia solution (390 mg/kg) while monitoring vital signs. The skin and fascia were dissected to expose the thigh muscle. Care was taken to avoid nonhomogeneous areas or areas with fat inclusions for ablation targets.

A custom-designed flow cell that recreates the heart's heat load conditions was placed on the thigh muscle, and saline was circulated to simulate blood flow (Fig. 1) similar to the apparatus described in Picher et al. [5]. A fluid dynamics analysis was conducted to determine the optimal flow in the flow cell. Specifically, the 8-mm catheter was modified to operate at a low flow and placed into the porcine left atrium without tissue contact at the roof and mitral isthmus lines, as these represent the most extreme areas of lowest and highest flow. Refrigerant was injected at 650 sccm, which represents highest flow with stable

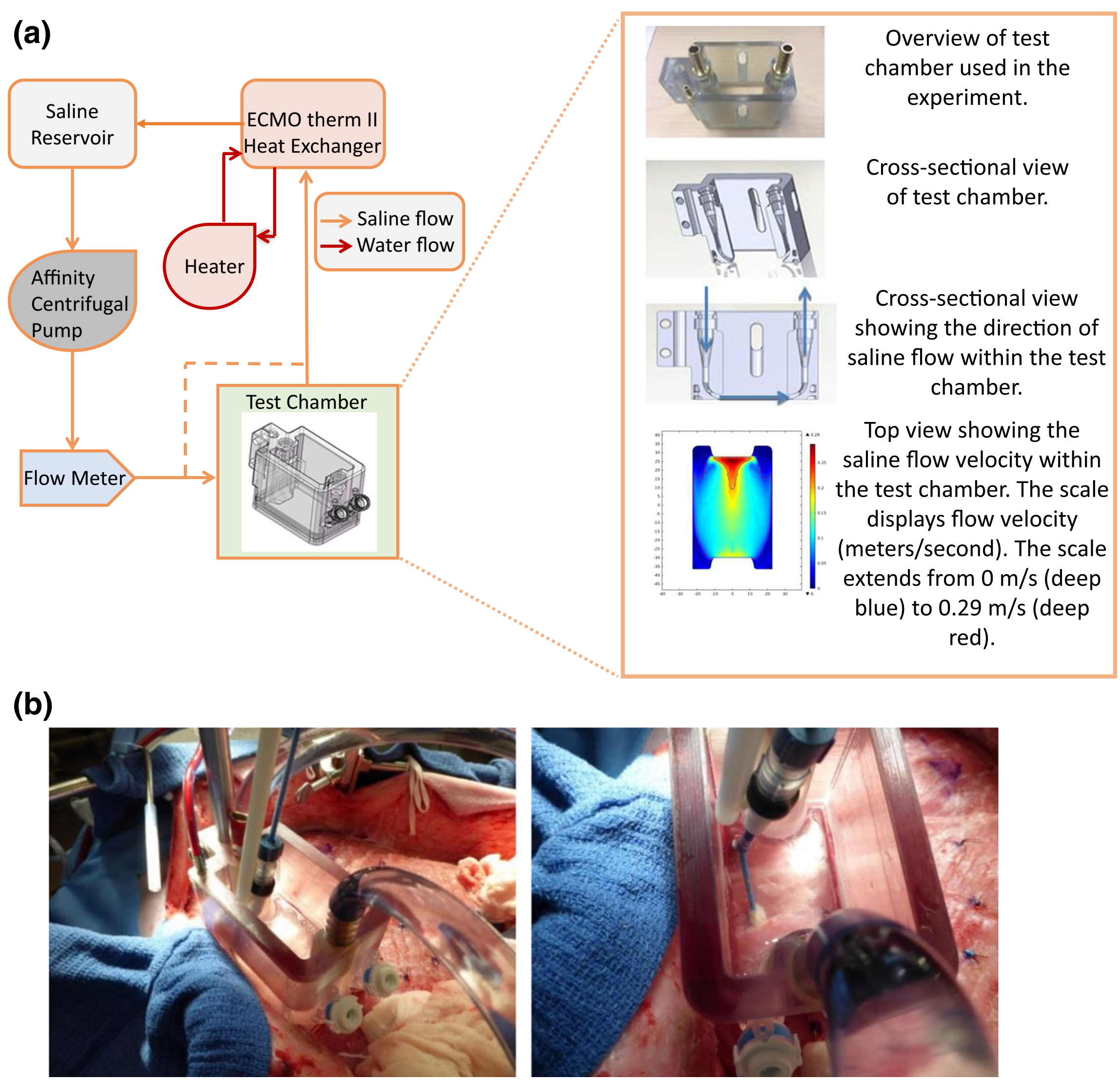

Fig. 1 **a** Schematic diagram of custom flow cell setup apparatus. **b** Image of thigh prep with flow cell placement (left) and placement of catheter during ablation (right)

temperature, and the tip temperature (2 × 4 min) was recorded at each location. The average temperature was recorded for minutes 2, 3, and 4. Next, the flow cell was moved to the thigh model. The saline flow in the flow cell was adjusted to get the same tip temperature as in the porcine left atrium. Ultimately, this analysis concluded that the settings of 1 L/min and 2 L/min in the flow chamber approximately simulate the worst-case convection factors for the left atrium in a pig model and these settings were used during the experiment (Table 1). Markers were placed in the lesion; locations were planned prior to the procedure and were tagged with markers.

2.2 Ablation protocol

Once the thigh model was prepped and the flow cell model was optimized, ablations were performed on the thigh muscle with all permutations on parameters as per Table 1 to compare the impact of electrode-tip size [4-mm tip (7Fr, Freezor), 6-mm tip (7Fr, Freezor Xtra), and 8-mm tip (9Fr, Freezor Max)], ablation time (2, 2 × 2, 3, 4, and 2 × 4 min), heat load (1 and 2 L/min), and tip orientation (perpendicular or parallel) on lesion size. To further evaluate the role of tissue contact during cryoablation procedures, a sub-study was performed with the 8-mm tip catheter only; first, cryoenergy was applied for 1 min without tissue contact until an ice ball formed on the catheter, followed by an additional 2–3 min freeze to determine the impact on lesion formation.

A total of 18 to 27 lesions per thigh were required to create all permutations and the sub-study. During the ablation, the operator attempted to maintain a constant contact force using a Shimpo™ digital force gauge (Fig. 2a–c). For each lesion, the parameters were recorded.

2.3 Morphometric analysis and data handling

After all ablations were completed, the animal was heparinized and euthanized per institutional protocol. Death was confirmed by absence of vital signs (pulse, cardiac electrical activity, pressure). The muscle portion containing all ablations was excised in total with wide margins and adequate depth. The muscle slab was immersed in 1% 2,3,5-triphenyltetrazolium Chloride (TTC) at 38 °C. The viable myocardium is stained red by the TTC and leaves damaged myocardium (lacking dehydrogenase enzymes) unstained (Fig. 2). The surface dimension (width) was measured with a caliper in instances where the ablation was sufficiently visible. Ablations were then cut in half, perpendicular from the surface, and parallel to the longest diameter. Each cryoablation was measured for maximum lesion length and depth by the pathologist, who was blinded to the parameter settings. Representative cross sections were imaged with a macro lens and with a ruler in the image. The lesion size outcomes were recorded, including length, depth, and cross-sectional area of a half-ellipsoid (= π × length/2 × depth/2).

Due to the gradual nature of the lesion border, there exists some degree of subjectivity where the lesion begins and ends; to control for potential measurement bias, a multi-step process was created. First, any lesions that were not completed per protocol due to documented system notices during the freeze, or any lesions that the pathologist was unable to measure accurately for length and depth due to insufficient ablation visibility, were removed from the dataset. Next, all lesion measurements were compared manually to corresponding lesion images by a second blinded reviewer; any discrepancies were reviewed by the principal investigator and a larger team and provided recommendations for data handling, including re-measurement of the tissue sample by the pathologist. Finally, if the new measurement was ± 10% different than the original measurement, the value was updated to the secondary measurement. If evidence was not found to update and/or discard data, the original measurement was maintained.

2.4 Statistical analysis

Descriptive statistics (mean, standard deviation, minimum and maximum) for unadjusted lesion dimensions were calculated for each parameter. To test the association of the measured parameters with lesion size, a linear regression analysis was performed for each outcome (length, depth, and cross-sectional area), adjusting for the effects of the other variables. The model was fitted using the least squares method. Interaction effects between catheter type and catheter orientation, catheter type and ablation time, and catheter orientation and ablation time were included in the model initially but were removed if statistical significance was > 0.1. A two-sided *p*

Table 1 Parameters tested

Parameter	Values
Catheter type (electrode tip size)	Freezor (4 mm), Freezor *Xtra* (6 mm), or Freezor *MAX* (8 mm)
Ablation time*	2 min, 2 × 2 min, 3 min, 4 min, 2 × 4 min (Freezor and Freezor *Xtra* only)
Heat load*	1 L/min, 2 L/min
Tip orientation	Perpendicular, parallel
Catheter force*	Constant for all catheters (30 ± 10 g, suggested)

*Nominal values. Actual values varied based on experimental circumstances and were recorded

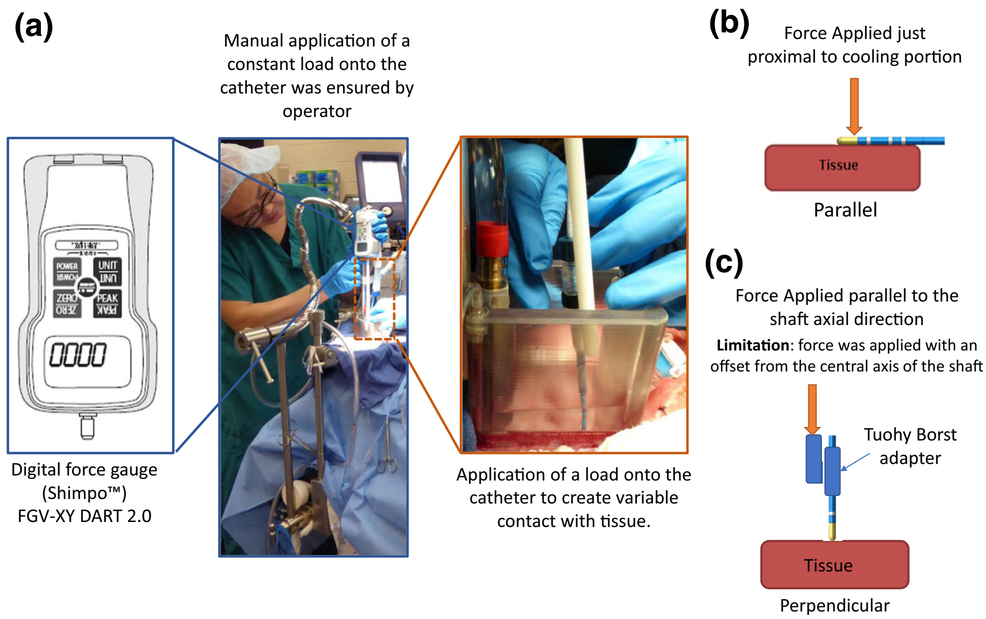

Fig. 2 Setup of methods to maintain contact force using the Shimpo force gauge images (**a**) and diagrams depicting the force applied in parallel (**b**) and perpendicular (**c**) axes

value < 0.05 was considered statistically significant (apart from the interaction effects). Statistical significance was not adjusted for multiple comparisons. The linear regression model assumes independence between each lesion. Because multiple lesions were created on each pig thigh, a random effects regression model was also utilized to account for multiple lesions within an animal.

Apart from the primary objectives to characterize lesion dimensions and testing associations between parameters using the linear regression model, several hypotheses were specified after the experiment, but before data analysis. Related to dosing, it was hypothesized that the depth and length of lesion for 2 × 2 min would be greater than 4 min for both orientations due to the benefits of the thawing cycle (Handler et al.). Additionally, it was hypothesized that the lesion dimensions for 2 × 4 min would be greater than 4 min freeze due to the longer duration of ablation. It was also hypothesized that a larger catheter tip (8 mm) would result in a larger lesion than a smaller tip (4 mm). Finally, it was hypothesized that perpendicular catheter orientation would increase lesion depth, as this orientation would likely radiate the cryoenergy more directly downward. For this post-hoc hypothesis testing, the lesion size differences were evaluated using *t* tests based on the final linear regression models for length, depth, and cross-sectional area.

For the ice ball sub-study, a two-sample *t* test was used to test for difference in means. The *F* test for inequality in variances was not significant for any outcome or hypothesis; therefore, pooled two sample *t* tests were applied.

3 Results

3.1 Ablation lesion

A total of 251 lesions (7 34, 40, 40, 43, 43, and 51 lesions per pig) were created during the study. From this group, 47 lesions were removed from analysis: 37 lesions where length/depth could not be measured, 9 not created per protocol, and 1 lesion with documented measurement issues. Furthermore, 10 lesions with ice ball freezing were excluded from the main analysis since they were only a part of the sub-study. Excluded lesions distributed roughly proportionally across all pigs, catheter tip sizes, and freeze times. The remaining 194 lesions were used for the main analysis (Fig. 3).

There were 66 lesions created by the 4-mm tip catheter, 68 lesions by the 6-mm catheter, and 60 lesions by the 8-mm catheter in the analysis cohort. The mean ± standard deviation lesion dimensions were 8.2 ± 2.2 mm in length, 4.9 ± 1.3 mm in depth, and 32.1 ± 12.3 mm^2 in cross-sectional area for 4-mm tip group; 9.1 ± 2.2 mm in length, 4.7 ± 1.3 mm in depth, and 34.4 ± 14.0 mm^2 in area for 6-mm tip group; and 9.8 ± 3.2 mm in length, 5.4 ± 1.2 mm in depth, and 41.4 ± 15.6 mm^2

in area for 8-mm tip group. The lesion dimensions are summarized by all tested parameters (Table 2).

Lesion ablation to euthanasia time (time between creation of the lesion and euthanasia of the animal) ranged from 7 to 440 min, with an average of 201 min across the 194 analyzed lesions.

3.2 Associations between ablation parameters and lesion size

The lesion length linear regression model demonstrated that catheter type ($p < 0.0001$), catheter orientation ($p < 0.0001$), ablation to euthanasia time ($p < 0.0001$), and the interaction between catheter type and catheter orientation ($p = 0.0006$) were significantly associated with lesion length (Table 3). The lesion depth linear regression model demonstrated that catheter type ($p = 0.003$) and catheter orientation ($p < 0.0001$) were significantly associated with lesion depth. The cross-sectional area linear regression model demonstrated that catheter type ($p < 0.0001$), ablation to euthanasia time ($p < 0.0001$), and the interaction between catheter orientation and catheter type ($p = 0.027$) were significantly associated with lesion cross-sectional area (Table 3; Fig. 4). The effect of ablation to euthanasia time (min) was estimated to be 0.03. When using random effect regression models to test the effects of independent variables on lesion length, depth, and cross-sectional area while also accounting for multiple lesions performed within an animal, the interpretation of all three models was the same as the corresponding linear regression models described above due to low variability from animal to animal. As such, only the original linear regression models are presented in Table 3.

The linear regression models detected no statistically significant effect of the pre-specified parameter of heat load or ablation time on length ($p = 0.789$, 0.32), depth ($p = 0.252$, 0.33), or cross-sectional area ($p = 0.467$, 0.10), respectively.

3.3 Post-hoc hypothesis testing

Several hypotheses were specified after the experiment, but before data analysis. There was no statistically significant difference between 2 × 2 min and 4 min ablation times for length ($p = 0.553$), depth ($p = 0.377$), or cross-sectional area ($p = 0.409$) using the linear model. Lesion depth for the 2 × 4 min ablation time was 0.6 mm deeper on average than lesions with a single 4 min freeze (95% CI −0.02–1.2; $p = 0.058$), but the lesion length was similar ($p = 0.260$). Cross-sectional area for the 2 × 4-min lesions was also 7.13 mm^2 larger on average than lesions with a 4-min application (95% CI 0.6–13.7; $p = 0.034$). There was no difference between 2 min and 4 min ablation times in terms of lesion length ($p = 0.610$), depth ($p = 0.971$), or cross-sectional area ($p = 0.581$).

Lesions created in the parallel orientation by the 8-mm tip catheter had 18.66 mm^2 greater cross-sectional area than lesions created by 4-mm (95% CI 11.8–25.5; $p < 0.0001$) and 19.20 mm^2 greater cross-sectional area than lesions created by 6-mm tip catheters (95% CI 12.7–25.7; $p < 0.0001$). Lesions created in the perpendicular orientation by the 8-mm tip catheter had 7.39 mm^2 greater cross-sectional area than lesions created by the 4-mm tip catheter (95% CI 0.4–14.4; $p = 0.042$) but were not significantly different from those created by the 6-mm tip catheter (mean difference 2.26 mm^2; 95% CI −4.87–9.38; $p = 0.54$). (Fig. 5).

Lesions created in the parallel orientation were longer than lesions created in the perpendicular orientation when using 4-mm ($p = 0.0006$), 6-mm ($p = 0.072$), and 8-mm tip catheter ($p < 0.0001$), whereas lesions created in the perpendicular orientation were deeper than lesions created with the parallel orientation using 4-mm ($p =$

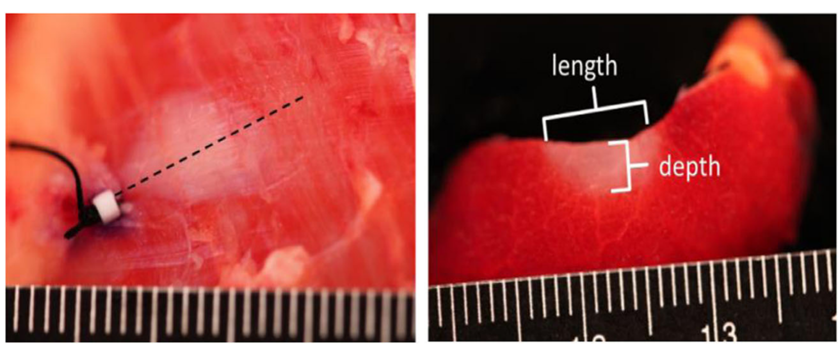

Fig. 3 Representative images of lesion sectioned along the length and stained with TTC. Approximate measurements of width, length, and depth are illustrated

Table 2 Summary of unadjusted lesion characteristics by freezing times, catheter orientation, and ice ball formation, including all experimental parameters

	Number	Length (mm) Mean (standard deviation)	Depth (mm) Mean (standard deviation)	Cross-sectional area (mm^2) Mean (standard deviation)
Catheter tip size				
4-mm (Freezor)	66	8.2 (2.2)	4.9 (1.3)	32.1 (12.3)
6-mm (Freezor Xtra)	68	9.1 (2.2)	4.7 (1.3)	34.4 (14.0)
8-mm (Freezor Max)	60	9.8 (3.2)	5.4 (1.2)	41.4 (15.6)
Ablation time				
2 min	39	9.1 (2.9)	4.8 (1.3)	34.2 (13.9)
3 min	42	8.7 (2.4)	5.1 (1.5)	35.1 (14.2)
4 min	42	9.1 (2.2)	4.8 (1.3)	35.2 (14.5)
2 × 2 min	44	9.2 (2.8)	5.1 (1.1)	37.0 (13.7)
2 × 4 min	27	9.1 (3.0)	5.2 (1.4)	38.1 (17.3)
Heat load				
1 L/min	99	9.2 (2.6)	5.1 (1.4)	37.5 (15.4)
2 L/min	95	8.9 (2.6)	4.9 (1.3)	34.0 (13.2)
Catheter orientation				
Parallel	98	10.0 (3.0)	4.6 (1.1)	36.9 (15.3)
Perpendicular	96	8.0 (1.7)	5.4 (1.4)	34.6 (13.5)
Ice ball formation and ablation time				
1 min ice ball +2 min freeze	5	10.4 (3.4)	3.8 (0.7)	31.9 (14.1)
2 min	15	9.9 (3.5)	5.1 (1.0)	39.2 (15.0)
1 min ice ball + 3 min freeze	5	9.7 (2.3)	4.0 (1.0)	30.5 (10.6)
3 min	15	8.9 (3.0)	5.6 (1.4)	38.6 (15.2)

0.136), 6-mm ($p = 0.005$), and 8-mm tip catheter ($p = 0.004$).

3.4 Sub-analysis of ice ball formation

The formation of an ice ball prior to 2 min of freezing reduced lesion depth by 1.3 mm compared to freezing without an ice ball ($p = 0.018$; Table 2). Similarly, the formation of an ice ball prior to 3 min of freezing reduced lesion depth by 1.55 mm compared to freezing without first forming an ice ball ($p = 0.031$; Table 2).

4 Discussion

This pre-clinical study evaluated the impact of cryoablation catheter type, ablation time, catheter tip orientation, and heat load on lesion size using a porcine thigh model. The results of this study add to previous literature by demonstrating that

Table 3 Overall model effects of explanatory variables for lesion length, depth, and cross-sectional area

Effect	Degrees of freedom	Length *F* test statistic (*P* value)	Depth *F* test statistic (*P* value)	Cross-sectional area *F* test statistics (*P* value)
Ablation time	4	1.18 (0.32)	1.17 (0.33)	2.00 (0.10)
Catheter type	*2*	*14.29 (< .0001)*	*6.01 (0.0030)*	*15.49 (< .0001)*
Catheter orientation	1	*49.88 (< .0001)*	*17.25 (< .0001)*	3.58 (0.06)
Catheter orientation × catheter type	*2*	*7.77 (0.0006)*	*N/A*	*3.67 (0.03)*
Heat load	1	0.07 (0.7891)	1.32 (0.25)	0.53 (0.47)
Ablation to euthanasia time (min)	*1*	*27.14 (< .0001)*	0.43 (0.51)	*18.63 (< .0001)*

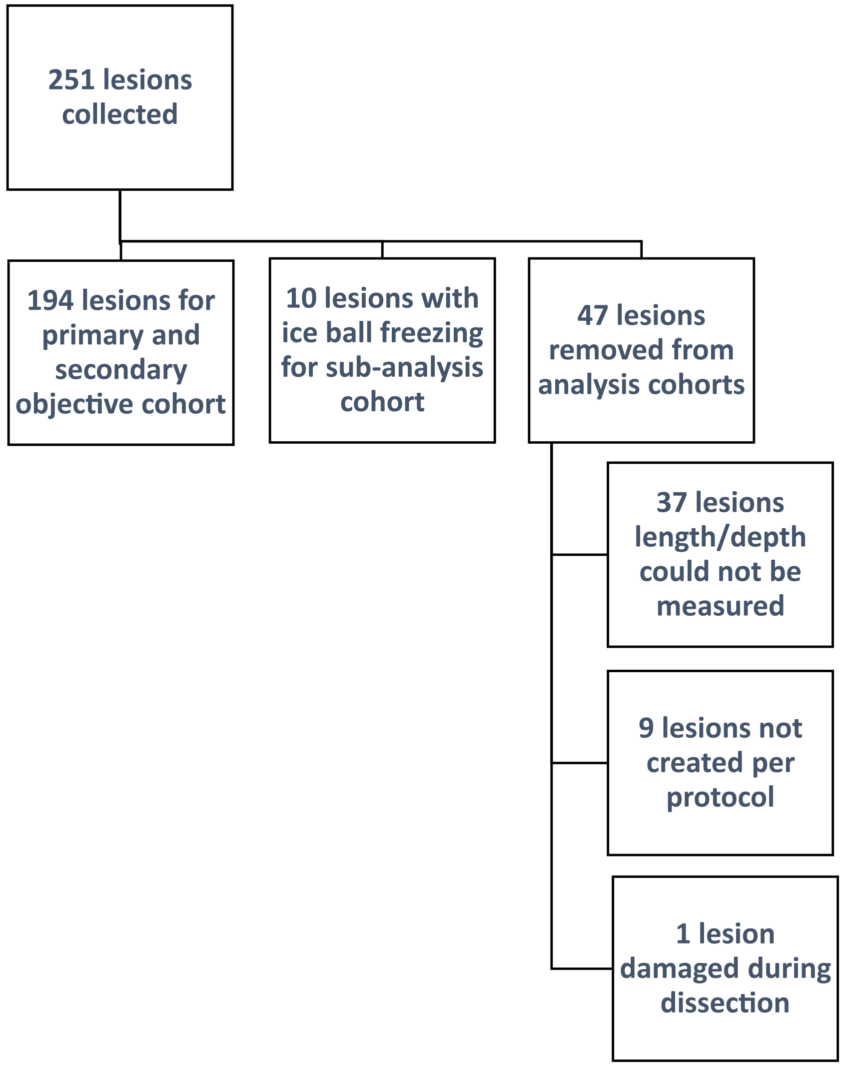

Fig. 4 Study lesion disposition

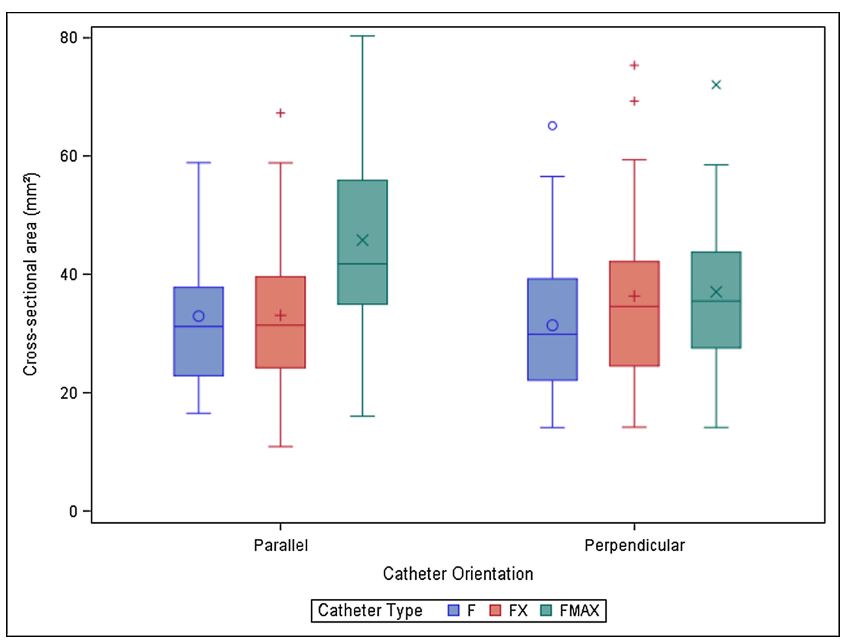

Fig. 5 Lesion cross-sectional area by orientation and catheter type box plot, including outliers. F Freezor (4 mm tip), FX Freezor Xtra (6 mm tip), FMax Freezor Max (8 mm tip)

cryoablation catheter type (electrode tip length) and tip orientation significantly impact lesion size. In addition, the presence of an ice ball prior to ablation due to a lack of tissue contact significantly reduces lesion depth compared to direct tissue application.

Lesion dimensions observed in the present study are similar to previous reports that use a variety of catheters [6–11]. Specifically, the present study mean ± SD depths of 4.9 ± 1.3 mm, 4.7 ± 1.3 mm, and 5.4 ± 1.2 mm for the 4 mm, 6 mm, and 8 mm tip catheters, respectively, which are aligned with the depth of 4.9 ± 1.7 across both 7F and 9F catheters reported previously by Khairy et al. [6]. Although the present study only evaluated lesions produced by cryoablation catheter, present study results with the 4-mm tip (length 8.2 mm, width 4.9 mm) were slightly larger; the observed lesion dimensions in the present study with a 4-mm tip cryoablation catheter were slightly larger than lesions produced by radiofrequency catheters in a porcine thigh model with an average length (7-mm), width (4.5-mm), and depth (4.5-mm) as reported by Tse et al. [10].

4.1 Significant factors associated with lesion dimension

The results of this study showed that the cryoablation catheter type significantly impacted lesion length, depth, and cross-sectional area. The catheter with the larger electrode tip created larger and deeper lesions than the catheter with the smaller electrode tip. Khairy et al. showed in the porcine heart that the surface area, but not depth, of lesions was significantly different between catheter type [11]. In contrast, the significant effect of catheter type on lesion depth in the present study may be due to the different model tissue utilized (thigh vs. beating heart) or inconsistencies in tip orientation or tissue contact that were not specifically controlled in their study. Additionally, the main driver of lesion depth is the catheter footprint and refrigerant flow, which is similar for 4-mm and 6-mm tip catheters, whereas the 8-mm tip catheter has a larger footprint and higher flow. Our study results, together with published data, suggest that catheter selection is essential to control the lesion size in cryoablation.

Using linear regression modeling, this study also demonstrated that lesion length and depth, but not cross-sectional area, are significantly impacted by the orientation of the catheter tip. Each of the three catheters has electrodes that are greater in length than in tip diameter. Thus, it was hypothesized that the lesion lengths would be greater when the tip was in the parallel orientation to the tissue compared to the perpendicular orientation, which was confirmed in our study. In addition, the results demonstrate that lesions performed with catheters in the perpendicular orientation create significantly deeper lesions than when in the parallel orientation. Interestingly, catheter tip orientation did not significantly impact lesion cross-sectional area. This may be the result of increased lesion length in the parallel orientation contrasted by increased lesion depth in the opposite perpendicular orientation, resulting in similar cross-sectional areas for each orientation. Therefore, in addition to the selection of catheter tip diameter, care in handling catheter tip orientation during the procedure is required to create the desired lesion size. For example, one may want to create shallower lesions around the pulmonary veins for gap ablation, whereas maximal depth with minimal width may be desirable for ablations near the AV node.

Analysis of the data unexpectedly identified that the time between lesion creation and euthanasia of the animal was associated with lesion size, which has not been previously described as a variable associated with lesion size. As such, this was not pre-specified as a controlled variable in the experiment. Importantly, the linear regression model accounted for this post-ablation variable and adjusted for the impact on lesion size to evaluate the effect of the intended tested parameters during ablation. This interaction should be closely considered in future experiments of acute lesions to proactively control lesion ablation to euthanasia time where possible.

4.2 Factors not associated with lesion dimension

Interestingly, in this porcine thigh model, the ablation time did not significantly impact lesion length, depth, or cross-sectional area. Previously, Tse et al. demonstrated that increased cryoablation times and the use of multiple freezes vs. a single freeze resulted in increased lesion size [4]; however, minimal differences in lesion size were observed when comparing 2 min vs. 4 min or 2 × 2 min vs. 4 min ablation times in the present study. Tse's study results [4] are incongruent not only to the present study but also to other published studies as well, which may be due to difference in study models (e.g., flow cell apparatus settings) and catheter tip size (e.g., 4-mm, 6-mm, 8-mm vs. 6.5 mm). Andrade et al. showed that 2 and 4 min of ablation produced similar lesion size [12], and Takami et al. reported that 3 and 4 min of ablation produced similar lesion size at PV antrum [13]. Using an in-silico model, Handler et al. showed that the influence of multiple freeze-thaw cycles on lesion size varies according to the duration of the initial freezing phase and also the duration of the thawing period, with a longer thawing period resulting in different lesion depths [14]. In the current study, the impact of the ablation time on lesion size may have plateaued by 2 min. Our study results suggest that in clinical practice, bonus applications close to the successful application site might be recommended rather than repeat applications (freeze-thaw-freeze) at the same site or prolongation of application after 2 min of successful application.

Although it was hypothesized that a high heat load which models for areas of high blood flow in the heart would result in a significant decrease in lesion size, the data demonstrate that the catheters maintain heat transfer from the tissue in both low and high heat load conditions.

4.3 Ice ball sub-study

Results from the ice ball formation sub-analysis experiments demonstrated that the presence of an ice ball prior to ablation due to a lack of tissue contact significantly reduces lesion depth compared to direct application to the tissue. Conversely, the presence of an ice ball showed no significant differences in lesion length. In clinical practice, ice balls may form when there is not sufficient contact. We observed that if there is not sufficient contact and an ice ball is formed consequently, lesion depth is reduced significantly. Therefore, ensuring adequate contact is essential for increasing the likelihood of a lesion with optimal depth. However, a lesion can still be created even after ice ball formation, which might support the “pull-down technique” during cryoballoon ablation in challenging cases [15].

4.4 Study limitations

First, while the thigh model is advantageous for comparing multiple parameters as in this study, there may be factors affecting ablation in the heart that is not fully accounted for in this model. The thigh model was developed originally in canines by Nakagawa et al. [7] in 1998, and a porcine thigh model has been used in multiple assessments of ablation catheter lesion dimension assessments [4, 8–10]. While not set up to exactly compare myocardium tissue vs. thigh models, Wijffels et al. found similar results in lesion characteristics between the two models [9]. Second, assessment of lesion morphology was only performed acutely, without additional evaluation of electrophysiological findings and lesion durability at the chronic phase. Thus, while the present findings provide insight into the acute effects of various ablation parameters on lesion size, potential changes in lesion characteristics over longer time periods that are not accounted for in this model should be considered [1]. In addition, despite efforts to manually maintain a constant load using a digital force gauge, there were challenges in providing a consistent heat load that may have impacted the fidelity of the measurements for the heat load parameter. Specifically, the pump and reservoir for the saline are optimized for one flow cell. However, two flow cells were often employed in this study, which may have resulted in heat load variance. Additionally, the force was applied with an offset from the central axis of the shaft in the perpendicular position versus just proximal to the cooling portion in the parallel orientation (Fig. 2c), so we cannot be certain that the force was exactly the same in both orientations. Future work should take heat load variance and methods to maintain contact force into consideration. Also, the number of lesions performed in the ice ball sub-study was small; however, were no outliers that appeared to significantly drive the observed significant difference of depth between ablations with ice ball formation vs. those without. Finally, since this study only utilized focal cryoablation catheters, results may not be applicable to the more commonly used cryoballoon catheters.

5 Conclusions

Results from this study demonstrate that lesion size is significantly impacted by catheter type and catheter tip orientation. Catheters with larger electrode tips created larger and deeper lesions. Catheters in the perpendicular orientation deliver deeper lesions whereas the parallel orientation creates longer lesions. Additionally, this study suggests that catheter selection and tip orientation are more important in determining lesion size than ablation time when utilizing ablation time greater than 2 min. Maintaining tissue contact prior to applying cryoenergy is essential.

Funding information This work was financially supported through a Medtronic research grant.

Compliance with ethical standards

The pig specimens were housed and received humane care in accordance with the Guide for Care and Use of Laboratory Animals and the Animal Welfare Act as amended. The study protocol was reviewed and approved by the Medtronic PRL IACUC (Institutional Animal Care and Use Committee).

Conflict of interest The authors declare that they have no conflict of interest.

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