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Shorter cryoballoon applications times do effect efficacy but result in less phrenic nerve injury: Results of the randomized 123 study

Marleen M.D. Molenaar MSc[1,2] | Carl C. Timmermans MD, PhD[3] | Tim Hesselink MD[1] | Marcoen F. Scholten MD, PhD[1] | Rachel M.A. ter Bekke MD, PhD[3] | Justin G.L.M. Luermans MD, PhD[3] | Marjolein Brusse-Keizer PhD[1] | Karin Kraaier MD, PhD[1] | Bernard ten Haken PhD[2] | Jan G. Grandjean MD, PhD[1] | Kevin Vernooy MD, PhD[3] | Jurren M. van Opstal MD, PhD[1]

[1]Medisch Spectrum Twente, Enschede, The Netherlands

[2]Universiteit Twente, Enschede, The Netherlands

[3]Maastricht University Medical Centre, Maastricht, The Netherlands

Correspondence
Marleen M.D. Molenaar, MSc, Thorax Centre Twente, Medisch Spectrum Twente, Koningsplein 1, 7512 KZ Enschede The Netherlands.
Email: mmdmolenaar@gmail.com

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Abstract

Background: The second-generation cryoballoon significantly improves outcome of pulmonary vein isolation (PVI) but may cause more complications than the first generation. Currently, no consensus regarding optimal cryoballoon application time exists. The 123-study aimed to assess the minimal cryoballoon application duration necessary to achieve PVI (primary endpoint) and the effect of application duration on prevention of phrenic nerve injury (PNI).

Methods: Patients <75 years of age with paroxysmal atrial fibrillation, normal PV anatomy, and left atrial size <40 cc/m^2 or <50 mm were randomized to two applications of different duration: "short," "medium," or "long." A total of 222 patients were enrolled, 74 per group.

Results: Duration per application was 105 (101-108), 164 (160-168), and 224 (219-226) s and isolation was achieved in 79, 89, and 90% ($P < 0.001$) of the PVs after two applications in groups short, medium, and long, respectively. Only for the left PVs, the success rate of the short group was significantly less compared to the medium- and long-duration groups ($P < 0.001$). PNI during the procedure occurred in 19 PVs (6.5%) in the medium and in 20 PVs (6.8%) in the long duration groups compared to only five PVs (1.7%) in the short duration group ($P < 0.001$).

Conclusions: Short cryoballoon ablation application times, less than 2 min, did affect the success for the left PVs but not for the right PVs and resulted in less PNI. A PV tailored approach with shorter application times for the right PVs might be advocated.

KEYWORDS
atrial fibrillation, cryoballoon, phrenic nerve injury, pulmonary vein isolation, safety

1 | BACKGROUND

Since the observation of pulmonary vein (PV) ectopy as the origin for atrial fibrillation (AF), pulmonary vein isolation (PVI) has become the cornerstone for nonpharmaceutical AF treatment. Continuous improvement of ablation devices made catheter ablation the first-line therapy in selected patients, where radiofrequency (RF) and cryoballoon PVI are the recommended methods of the use.[1]

Cryoballoon ablation has shown to be safe and efficacy rates are noninferior to RF ablation.[2] The re-engineered second generation cryoballoon (Arctic Front Advance, Medtronic Inc., Minneapolis, MN, USA) resulted in more homogeneous cooling, which improved the

procedure efficacy.[3,4] However, this higher efficacy goes hand-in-hand with an increased risk of complications. In particular, when compared to the first generation, the second-generation cryoballoon is associated with a higher incidence of right phrenic nerve injury (PNI), which remains a considerable drawback of this technique.[4–7]

Historically, 4-min applications with a bonus application after isolation have been the standard for the first-generation cryoballoon. With the aim to reduce complications without affecting treatment efficacy, the dosing of cryoenergy becomes of critical importance for the second-generation cryoballoon. So far protocols with shorter ablation times have shown encouraging results.[8–10] However, this has been only demonstrated in retrospect and at this moment, uniform consensus regarding the optimal cryoballoon application time is lacking.

The main objective of the present study was to prospectively assess, in a randomized fashion, the minimal cryoballoon application time necessary to achieve PVI and the effect of minimizing application duration on preventing PNI.

2 | METHODS

2.1 | Study design

This clinical trial was designed as a patient-blinded, randomized, dual center (Medisch Spectrum Twente [MST], Enschede and Maastricht University Medical Centre+ [MUMC], Maastricht, the Netherlands) study. The study was approved by both local institutional ethical committees on human research and was registered in the clinicaltrial.gov database (no. NCT02074566).

2.2 | Study population

Patients fulfilling the following inclusion criteria were enrolled in the study: paroxysmal AF eligible for PVI according to the ESC guidelines and an age <75 years.[1,11] Patients were excluded if they had undergone a prior PVI; if the life expectancy was <12 months; if there was pregnancy at the time of the procedure, contrast allergy, creatinine clearance <60/mL/min/1.73 m^2, left ventricular ejection fraction (LVEF) <40%, left atrial (LA) volume >40 cc/m^2, or LA diameter >50 mm, or aberrant PV anatomy.

2.3 | Randomization

After written informed consent, patients were randomized to "short," "medium," or "long" application duration protocols in a 1:1:1 fashion. Patients received two applications of 1 (short), 2 (medium), or 3 (long) min after reaching maximal N_2O cooling flow; hence, this study was called "the 123-study." Maximal N_2O flow is achieved at 72 Standard Cubic Centimeters Minute (SCCM) for the 28-mm balloon and 56 SCCM for the 23-mm balloon and the time to reach maximal flow may differ per application. The study flow chart is illustrated in Figure 1.

2.4 | PVI procedure

The procedures were performed under conscious sedation at the MUMC site and under general anesthesia, with continuous blood pressure monitoring, at the MST site. Heparin was administered to achieve an activated clotting time of >300 s during the procedure. Pacing on the Achieve (Medtronic Inc.) mapping catheter in the left superior PV prior to cryotherapy was performed to exclude left-sided phrenic capture. During cryoablation of the right-sided PVs, the right phrenic nerve was continuously stimulated by pacing from the superior caval vein or the right subclavian vein. When diminished diaphragm excursion was noted during cryotherapy, the application was stopped immediately using the double stop technique.[12] If a premature termination caused by diminished diaphragm excursion had to be performed, this was qualified as "PNI." At the end of a procedure in which PNI occurred, recovery of phrenic nerve function was tested by the stimulation.

Applications were performed using a double-walled cryoballoon (Arctic Front Advance [$n = 208$] or Arctic Front Advanced ST [$n = 14$], Medtronic Inc.). The preference was to use the 28-mm balloon. If the PV diameter was too small to occlude, we used the 23-mm balloon. Quality of PV occlusion was visualized by contrast administration during fluoroscopy and scored on a semiquantitative scale with grades 1 (very poor) to 4 (excellent), as described previously.[13] An occlusion of 4 was aimed for in every application. The operators were not blinded for the application duration. The second application started at least 1 min after deflation of the cryoballoon of the first application. The Achieve mapping catheter was inserted through the inner lumen of the balloon to assess PV signals before, during, and after every application and to guide the positioning of the balloon. The initial application sequence was left superior PV (LSPV), left inferior PV (LIPV), right superior PV (RSPV), and right inferior PV (RIPV) for the first 11 patients. The sequence was adjusted to LSPV, LIPV, RIPV, and RSPV after the occurrence of PNI in the RSPV in two patients, whereafter the RIPV could not be safely isolated anymore because the phrenic nerve could not be stimulated. Time to isolation (TTI), if visible, was measured for the last 170 patients.

Primary successful isolation was defined as an isolation of that particular PV, proven by an entrance and exit block, after two applications. Primary unsuccessful isolation was defined if there was no PV isolation after two applications. In PVs with primary unsuccessful isolation, supplementary applications were applied to ensure complete isolation for all the PVs. The number and duration of these supplementary application(s) was determined by the operating physician. In PVs, extremely challenging to isolate the Achieve catheter was placed in other PV branches of the same PV or the pull-down maneuvre was used.[14]

2.5 | Statistical analysis

The study was designed to assess three noninferiority hypotheses comparing the different groups (short, medium, long) independent of each other. Prior to the study, a noninferiority margin of 10% was determined and a power calculation that assumes success was performed. Based on this calculation, a total of 222 patients (3×74) were enrolled. Data analysis were performed using SPSS (version 22; IBM Corp., Armonk, NY, USA).

Continuous variables are summarized by mean ± standard deviation for normally distributed variables or median with interquartile ranges for non-normally distributed variables. Categorical variables

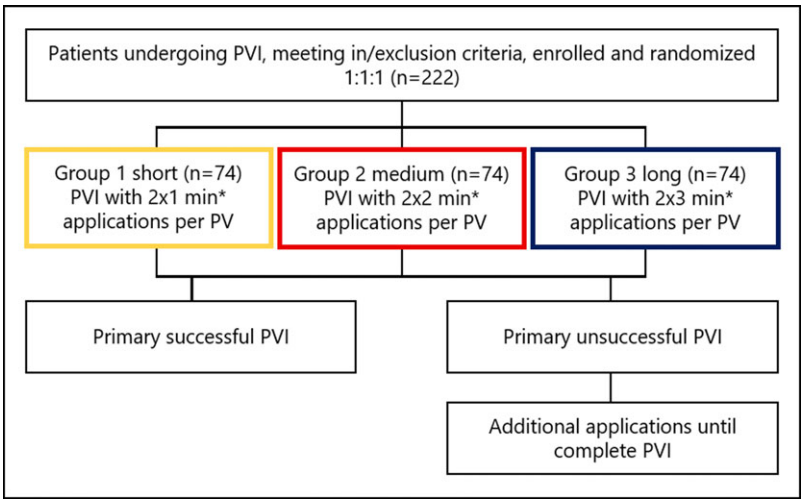

FIGURE 1 123-Study flowchart. PV = pulmonary vein; PVI = pulmonary vein isolation. *Time starts after maximal N_2O flow is reached [Color figure can be viewed at wileyonlinelibrary.com]

are expressed as numbers and percentages. Statistical differences for continuous normally distributed variables were analyzed using ANOVA for three groups. In case the analysis of variance (ANOVA) showed significant differences, post-hoc tests according to the method of Tukey's Honestly Significant Differences were performed. For non-normally distributed variables, the Kruskal–Wallis test was used to test for differences between the three groups with post-hoc Mann-Whitney U tests using Bonferroni-Holm correction. Statistical differences between the groups for categorical variables were analyzed using Chi-square or Fisher exact tests, as appropriate. In case significant differences between categorical variables were observed, post-hoc tests according to Bonferroni-Holm were performed. Also, 95% confidence intervals (CIs) to assess noninferiority for the success percentages were calculated using Epi Info.

All procedural characteristics are reported per patient, while success is mentioned per PV. Complication rates are reported per patient, as well as per PV. Balloon temperature, occlusion grade, thaw time, and application duration are reported per application.

3 | RESULTS

3.1 | Patient characteristics

Between July 2014 and August 2017, a total of 222 patients, 74 patients in each group, were enrolled. As shown in Table 1, there were no significant differences between the baseline patient characteristics for the three groups.

3.2 | Procedural characteristics

Table 2 shows the procedural characteristics for the three groups. As the consequence of the study design, the mean application duration was significantly different for the three groups. The time to reach maximal N_2O cooling flow was 44 [40-48] s and did not differ between the groups ($P = 0.21$, known for all but 167 applications). The procedure time was significantly shorter in the short group when compared to the long group.

3.2.1 | PVI success

Eight out of 888 PVs were not targeted for PVI with the cryoballoon. Applications could not be delivered due to technical malfunction of the cryo console ($n = 2$), the occurrence of PNI after ablation of the ipsilateral PV ($n = 2$), PV anatomy in which it appeared to be possible to occlude the ostium of the LSPV and LIPV simultaneously ($n = 3$), and impossibility to occlude the PV using the cryoballoon ($n = 1$).

The remaining 880 out of 888 PVs were targeted for PVI with the cryoballoon. The number of primary successful isolated PVs was significantly lower in the short group (79% [CI, 74-83%]) when compared to the medium group (89% [CI, 85-92%]) and the long group (90% [CI: 86-93%]) (Figure 2), but did not exceed the predefined margin of 10% regarding the noninferiority analysis. A subanalysis, comparing the successes for the groups for the four different PVs, showed that the significant difference between the groups was only present for the left PVs and there was no significant difference in success between the groups for the right PVs (Figure 3). The success after a single application differed significantly between the short (62%) and medium (75%) groups and between the short and long (78%) groups. An occlusion grade of 4/4 was achieved in 77% of all applications. Occlusion grades of 3, 2, and 1/4 were achieved in 15, 8%, and <1%, respectively, of all applications and occlusion grades did not differ significantly between the three groups ($P = 0.71$). The RIPV and LSPV were the most difficult PVs to isolate.

The number of supplementary cryoballoon applications per PV, needed to achieve complete PVI when isolation was primary unsuccessful, were 1.6 ± 1.1 for group 1, 1.6 ± 1.3 for group 2, and 1.8 ± 2.1 for group 3 and did not differ between the groups ($P = 0.87$).

In six of 880 PVs (six patients) complete isolation could not be achieved with the cryoballoon due to the occurrence of PNI ($n = 3$) or due to a PV anatomy, which only allowed poor occlusion ($n = 3$). In two patients with a PV anatomy, which only allowed poor occlusion, isolation was ultimately achieved using point-by-point cryo or RF ablation. Therefore, eventually in all but four patients a successful PVI was performed.

TABLE 1 Baseline patient characteristics

	Total ($n = 222$)	Short group ($n = 74$)	Medium group ($n = 74$)	Long group ($n = 74$)	*P* value
Age (years)	58 ± 9	58 ± 10	59 ± 9	57 ± 9	0.40
Gender (male,%)	143 (64)	46 (62)	47 (64)	50 (68)	0.78
CHA_2DS_2VASC score	1.2 ± 1.1	1.3 ± 1.2	1.2 ± 1.2	1.0 ± 1.0	0.34
Hypertension (%)	73 (33)	26 (35)	21 (28)	26 (35)	0.60
Diabetes mellitus 2 (%)	7 (3)	3 (4)	1 (1)	3 (4)	0.70
Prior revascularisation (%)[a]	7 (3)	3 (4)	3 (4)	3 (4)	1.00
EHRA class	2.4 ± 0.5	2.4 ± 0.5	2.4 ± 0.5	2.4 ± 0.6	0.90
Body mass index (kg/m^2)	27 ± 3	27 ± 3	27 ± 3	26 ± 3	0.48
Left atrial volume(cc/m^2)[b]	30 ± 8	29 ± 8	29 ± 10	30 ± 8	0.72

Data are expressed as mean ± standard deviation or *n* (%), unless otherwise indicated. EHRA = European Heart Rhythm Association.
[a]Revascularization by percutaneous coronary intervention and/or coronary artery bypass grafting.
[b]Left atrial volume in cc/m^2 was not available in 16 patients due to a limited echocardiographic window or missing echocardiography recordings. In these patients left atrial volume in mm was available (37 ± 6).

TABLE 2 Procedural characteristics

	Total ($n = 222$)	Short group ($n = 74$)	Medium group ($n = 74$)	Long group ($n = 74$)	*P* value
Duration per application (s) (median [IQR])	163 [107-219]	105 [101-108]	164 [160-168]	224 [219-226]	<0.001[a]
Lowest balloon temperature per application (°C)	−47 ± 8	−45 ± 8	−47 ± 8	−48 ± 7	<0.001[b]
Thaw time per application (s)	9 ± 5	8 ± 4	9 ± 5	9 ± 5	<0.001[c]
Balloon 23 mm (%)	5 (2.3)	1 (1.4)	2 (2.7)	2 (2.7)	0.52
28 mm (%)	212 (96)	70 (95)	70 (95)	72 (97)	
23 + 28 mm (%)	5 (2.3)	3 (4.1)	2 (2.7)	0 (0)	
Procedure time (hh:mm)	1:28 ± 0:24	1:22 ± 0:23	1:29 ± 0:27	1:32 ± 0:20	0.04[d]
Contrast (mL)	67 ± 38	69 ± 36	70 ± 44	63 ± 32	0.49
Radiation time (hh:mm)	0:22 ± 0:10	0:22 ± 0:10	0:23 ± 0:11	0:22 ± 0:10	0.65
Radiation dose area product ($mGy\ cm^2$) (median [IQR])	14.8 [8.5-22.4]	13.9 [7.3-19.7]	13.7 [9.1-22.5]	15.2 [9.8-23.9]	0.26

Data are expressed in mean ± standard deviation or *n* (%) unless stated otherwise. IQR = interquartile range.
[a]Significant between all groups.
[b]Significant between short and medium and short and long group.
[c]Significant between short and medium and short and long group. Known for all but 300 applications.
[d]Significant between short and long group.

3.3 | PNI

In 39 of 222 patients (18%), PNI occurred (44/880 PVs [5%]). PNI occurred significantly more in the medium group ($n = 19$ PVs [6.5%]) and in the long group ($n = 20$ PVs [6.8%]) when compared to the short group ($n = 5$ PVs [1.7%] $P = 0.006$) (Figure 4). PNI did also differ significantly between the four PVs ($P < 0.001$) and only occurred in the RIPV ($n = 10$ [1.1%]) and RSPV ($n = 34$ [3.9%]). There was no significant difference in minimal balloon temperature in the right PV applications in which PNI occurred when compared to the right PV applications in which PNI did not occur ($P = 0.71$). In 27 of 39 (69%) patients with PNI, phrenic nerve function had recovered completely at the end of the procedure.

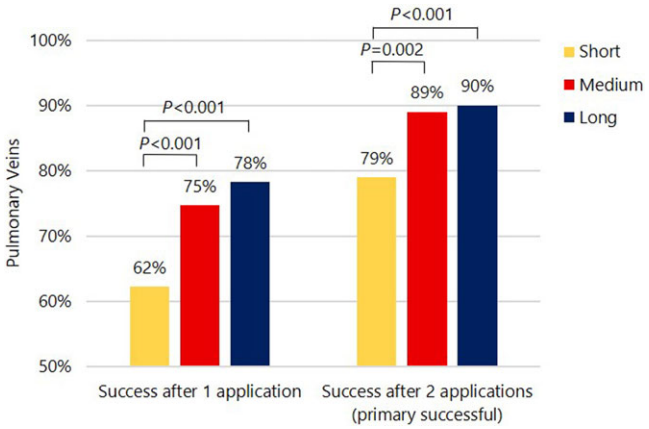

FIGURE 2 Percentage of successfully isolated pulmonary veins after one and two application(s) [Color figure can be viewed at wileyonlinelibrary.com]

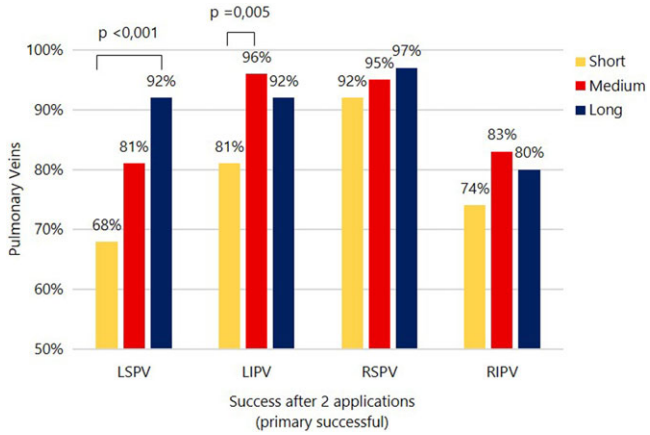

FIGURE 3 Percentage of successfully isolated pulmonary veins after two applications per PV. LIPV = left inferior PV; LSPV = left superior PV; PV = pulmonary vein; RIPV = right inferior PV; RSPV = right superior PV [Color figure can be viewed at wileyonlinelibrary.com]

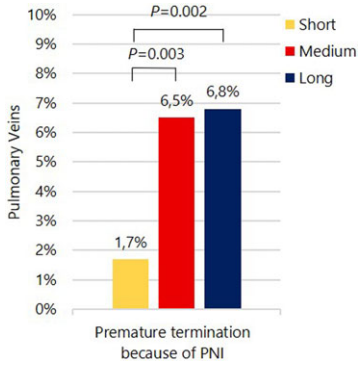

FIGURE 4 Percentage of pulmonary veins in which applications were prematurely terminated because of PNI. PNI = phrenic nerve injury [Color figure can be viewed at wileyonlinelibrary.com]

3.4 | Other complications

Three patients (one patient in every group) had vascular complications, all treated conservatively. One patient (medium group) experienced a minor transient ischemic attack. In one patient (medium group), ventricular fibrillation occurred during cryotherapy delivery in the RIPV with ST-segment elevation. A coronary angiogram showed no coronary artery occlusion, the ST-segment elevation resolved spontaneously, and the patient was discharged without sequela. No other major procedural complications occurred.

3.5 | Time to isolation

In 670 applications in which isolation was achieved, an attempt to measure TTI was made. The TTI could be assessed in 316 of 670 (47%). Mean TTI was shorter in PVs with persisting isolation at the end of the procedure compared to PVs showing early reconduction during the procedure (48 ± 30 s vs 62 ± 38 s; $P = 0.006$). Mean balloon temperature at the time of isolation was 35 ± 9°C and did not differ between these two groups ($P = 0.31$).

4 | DISCUSSION

The 123-study was designed to assess the minimal cryoballoon application duration for PVI while improving the safety. Patients were randomized to three different groups comparing standard to reduced application durations. The ultimate goal was to find an optimal freezing duration preserving the efficacy and improving the safety by avoiding PNI.

This prospective randomized study shows that (1) there is no difference in isolation rate between two applications of medium or long duration but two applications of short duration results in significantly less isolation, albeit that there is no proven superiority. (2) The significant differences in isolation rate between the three groups exist only for the left but not for the right PVs. (3) PNI occurred significantly more in the groups with medium and long freeze durations.

Historically, 4-minute applications with a bonus application after isolation were the standard for the first-generation cyroballoon.[15] With the advent of the second generation cryoballoon, efficacy increased but it caused a decrease in safety, especially for right PNI.[3,4] As a result, the paradigm shifted from efficacy to safety and, in recent years, the use of single applications and applications without bonus applications after successful isolation has been explored.[8,9,16–18] The omission of bonus applications did not compromise the efficacy of PVI and, depending on the PV, the mean number of applications needed for isolation was found to be between 1 and 2.[8,16,18,19] Also, shorter application durations have been suggested and adopted, shortening the standard application duration in many centers to 180 s,[8,9] recently followed by studies that include TTI of a PV, if recordable during ablation.[10,20–22] However, despite the encouraging results in terms of efficacy no previous study was performed prospectively in a randomized fashion comparing the standard to new dosing protocols.

4.1 | Efficacy

To eliminate the difference in effective ablation time within the three groups, due to differences in time needed to reach maximal N_2O freezing flow, application durations for the three groups were defined as 1, 2, and 3 min after reaching maximal N_2O flow. This resulted in a mean application duration of 104 ± 13 s for the short group, 159 ± 19 s for the medium group, and 212 ± 35 s for the long group. Therefore, the long group may be compared to the historical standard of 4-min (240 s) applications.

The present study demonstrates that 2 × 2 min of ablation after reaching maximal N_2O freezing flow does not impair the efficacy. Further shortening of the application duration significantly reduced results for the left PVs, although no inferiority was proven. In contrast, for the right PVs, even 2 × 1 min of freezing after reaching maximal N_2O freezing flow showed similar isolation rates compared to longer cryo application durations.

The difference between the left and right PVs may be explained by the left lateral ridge, a fold of the atrial wall protruding into the endocardial LA surface between the orifices of the left PVs, and the ostium of the left atrial appendage characterized by thick muscular tissue and known to be associated with isolation gaps.[23,24] Because of its

thickness, it may take longer for cryoenergy to disperse and form a transmural lesion, therefore, a longer ablation duration may be needed to achieve isolation in the left PVs.

In this study, the number of successfully isolated PVs after a single application is similar to earlier reports using 180 and 240 s application durations.[8,9,17–19]

4.2 | Safety

In general, the complication rate in this study was similar to comparable previous studies. In our study, premature termination because of PNI occurred in 5% of the PVs and was limited to the right PVs. Also, the RIPV accounted for a substantial amount of these premature terminations. Compared to the short group, premature termination because of PNI was more common in the groups with longer application durations.

The incidence of PNI has been reported to increase from 2-11% in the first-generation balloon[4,15,25,26] to 3-24% in the second-generation balloon.[4,25–27] Previous study showed that the mean time to PNI was 123.6 ± 47 s (median: 131), indicating that shorter application duration might prevent the occurrence of this complication.[27]

This study showed that shortening application duration reduces the incidence of PNI while, for the right PVs, even the ablation protocol with the shortest duration did not impair efficacy. Therefore, a PV-tailored approach with shorter application duration for the right PVs may be recommended. This is especially important as deterioration of diaphragm contractions sometimes already indicates irreversible or long-lasting nerve damage.

4.3 | TTI

Most recent studies considering dosing protocols focus on the TTI of a PV.[10,20,22] The TTI in these protocols is used as a surrogate parameter for monitoring tissue physiology. This method has shown to be a predictor for durable lesions and enables a PV- and patient-tailored approach. In line with previous studies, we found a significantly longer TTI in the applications that showed reconduction during the procedure. However, not in every single application is TTI measurable. Therefore, an optimal ablation duration irrespective of TTI is needed. Furthermore, when incorporating TTI as the new standard in ablation protocols, data are needed to determine the optimal duration of an application àfter TTI is reached. Moreover, as recently mentioned, TTI as well as application time are both surrogate parameters.[28] Ideally, a biophysical parameter that directly measures ice formation or, ultimately, irreversible myocardial tissue deconstruction, should be available. Recently, the results of a first promising biophysical parameter that measures ice formation have been presented. This study is a proof of concept performed in an animal model, so we have to wait for clinical applicability.[29]

5 | CONCLUSION

A shortened application duration protocol of medium duration does not impair isolation success compared to the historical standard of long application duration. Further reduction to short application times did significantly impair isolation for the left, but not for the right, PVs. For the right PVs reduction to short application times, less than 2 min, resulted in less PNI.

The 123-study is a randomized study demonstrating that PNI during cryoballoon PVI can be reduced by further shortening the application duration for the right PVs, while maintaining similar efficacy rates. A PV-tailored approach with shorter application times for the right PVs might be advocated.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

ORCID

Marleen M.D. Molenaar MSc
https://orcid.org/0000-0001-5658-8905
Rachel M.A. ter Bekke MD, PhD
https://orcid.org/0000-0003-4278-0132
Marjolein Brusse-Keizer PhD
https://orcid.org/0000-0003-1781-5794
Bernard ten Haken PhD https://orcid.org/0000-0001-8159-7424

REFERENCES

1. Kirchhof P, Benussi S, Kotecha D, et al. 2016 ESC Guidelines for the management of atrial fibrillation developed in collaboration with EACTS. *Eur Heart J*. 2016;50:e1-e88.
2. Kuck K-H, Brugada J, Fürnkranz A, et al. Cryoballoon or radiofrequency ablation for paroxysmal atrial fibrillation. *N Engl J Med*. 2016;374:2235-2245.
3. Liu J, Kaufmann J, Kriatselis C, Fleck E, Gerds-Li JH. Second generation of cryoballoons can improve efficiency of cryoablation for atrial fibrillation. *Pacing Clin Electrophysiol*. 2015;38:129-135.
4. Aytemir K, Gurses KM, Yalcin MU, et al. Safety and efficacy outcomes in patients undergoing pulmonary vein isolation with second-generation cryoballoon. *Europace*. 2014;17:379-387.
5. Fürnkranz A, Bordignon S, Dugo D, et al. Improved 1-year clinical success rate of pulmonary vein isolation with the second-generation cryoballoon in patients with paroxysmal atrial fibrillation. *J Cardiovasc Electrophysiol*. 2014;25:840-844.
6. Guiot A, Savouré A, Godin B, Anselme F. Collateral nervous damages after cryoballoon pulmonary vein isolation. *J Cardiovasc Electrophysiol*. 2012;23:346-351.
7. Fürnkranz A, Bordignon S, Schmidt B, et al. Luminal esophageal temperature predicts esophageal lesions after second-generation cryoballoon pulmonary vein isolation. *Heart Rhythm*. 2013;10:789-793.
8. Ciconte G, de Asmundis C, Sieira J, et al. Single 3-minute freeze for second-generation cryoballoon ablation: One-year follow-up after pulmonary vein isolation. *Heart Rhythm*. 2015;12:673-680.

9. Miyazaki S, Hachiya H, Nakamura H, et al. Pulmonary vein isolation using a second-generation cryoballoon in patients with paroxysmal atrial fibrillation: One-year outcome using a single big-balloon 3-minute freeze technique. *J Cardiovasc Electrophysiol*. 2016;27:1375-1380.
10. Chun KRJ, Stich M, Fürnkranz A, et al. Individualized cryoballoon energy pulmonary vein isolation guided by real-time pulmonary vein recordings, the randomized ICE-T trial. *Heart Rhythm*. 2017;14:495-500.
11. John Camm A, Lip GYH, De Caterina R, et al. 2012 focused update of the ESC Guidelines for the management of atrial fibrillation. *Eur Heart J*. 2012;33:2719-2747.
12. Ghosh J, Sepahpour A, Chan KH, Singarayar S, McGuire MA. Immediate balloon deflation for prevention of persistent phrenic nerve palsy during pulmonary vein isolation by balloon cryoablation. *Heart Rhythm*. 2013;10:646-652.
13. Neumann T, Vogt J, Schumacher B, et al. Circumferential pulmonary vein isolation with the cryoballoon technique. Results from a prospective 3-center study. *J Am Coll Cardiol*. 2008;52:273-278.
14. Chun K-RJ, Schmidt B, Metzner A, et al. The "single big cryoballoon" technique for acute pulmonary vein isolation in patients with paroxysmal atrial fibrillation: A prospective observational single centre study. *Eur Heart J*. 2009;30:699-709.
15. Packer DL, Kowal RC, Wheelan KR, et al. Cryoballoon ablation of pulmonary veins for paroxysmal atrial fibrillation: First results of the North American Arctic Front (STOP AF) pivotal trial. *J Am Coll Cardiol*. 2013;61:1713-1723.
16. Heeger C-H, Wissner E, Wohlmuth P, et al. Bonus-freeze: Benefit or risk? Two-year outcome and procedural comparison of a "bonus-freeze" and "no bonus-freeze" protocol using the second-generation cryoballoon for pulmonary vein isolation. *Clin Res Cardiol*. 2016;105:774-782.
17. De Regibus V, Iacopino S, Abugattas JP, et al. Single freeze strategy with the second- generation cryballoon for atrial fibrillation: A multicenter international retrospective analysis in a large cohort of patients. *J Interv Card Electrophysiol*. 2017;49:173-180.
18. Tebbenjohanns J, Höfer C, Bergmann L, et al. Shortening of freezing cycles provides equal outcome to standard ablation procedure using second-generation 28 mm cryoballoon after 15-month follow-up. *Europace*. 2016;18:206-210.
19. Wissner E, Heeger C-H, Grahn H, et al. One-year clinical success of a "no-bonus" freeze protocol using the second-generation 28 mm cryoballoon for pulmonary vein isolation. *Europace*. 2015;17:1236-1240.
20. Aryana A, Kenigsberg DN, Kowalski M, et al. Verification of a novel atrial fibrillation cryoablation dosing algorithm guided by time-to-pulmonary vein isolation: Results from the Cryo-dosing study. *Heart Rhythm*. 2017;14:1319-1325.
21. Rottner L, Fink T, Heeger C-H, et al. Is less more? Impact of different ablation protocols on periprocedural complications in second-generation cryoballoon based pulmonary vein isolation. *Europace*. 2018;20:1459-1467.
22. Reissmann B, Wissner E, Deiss S, et al. First insights into cryoballoon-based pulmonary vein isolation taking the individual time-to-isolation into account. *EP Europace*. 2017;19:1676-1680.
23. Cabrera JA, Ho SY, Climent V, Sanchez-Quintana D. The architecture of the left lateral atrial wall: A particular anatomic region with implications for ablation of atrial fibrillation. *Eur Heart J*. 2008;29:356-362.
24. Sánchez-Quintana D, Doblado-Calatrava M, Cabrera JA, Macías Y, Saremi F. Anatomical basis for the cardiac interventional electrophysiologist. *Biomed Res Int*. 2015;2015:1-24.
25. Fürnkranz A, Bordignon S, Schmidt B, et al. Incidence and characteristics of phrenic nerve palsy following pulmonary vein isolation with the second-generation as compared with the first-generation cryoballoon in 360 consecutive patients. *Europace*. 2015;17:574-578.
26. Martins RP, Hamon D, Césari O, et al. Safety and efficacy of a second-generation cryoballoon in the ablation of paroxysmal atrial fibrillation. *Heart Rhythm*. 2014;11:386-393.
27. Chierchia G-B, Mugnai G, Hunuk B, et al. Impact on clinical outcome of premature interruption of cryoenergy delivery due to phrenic nerve palsy during second generation cryoballoon ablation for paroxysmal atrial fibrillation. *J Cardiovasc Electrophysiol*. 2015;26:950-955.
28. Avitall B. Letter to the editor—in search of cryoablation dose. *Heart Rhythm*. 2017;14:e501.
29. Avitall B, Kalinski A, Kocheril GS, Lizama KS, Coulombe N, Laske T. Characteristics of ice impedance recorded from a ring electrode placed at the anterior surface of the cryoballoon: Novel approach to define ice formation and pulmonary vein isolation. *Circ Arrhythmia Electrophysiol*. 2018;11:1-9.

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