Impact of Lesion Placement on Efficacy and Safety of Catheter-Based Radiofrequency Renal Denervation

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ABSTRACT

BACKGROUND Insufficient procedural efficacy has been proposed to explain nonresponse to renal denervation (RDN).

OBJECTIVES The aim of this study was to examine the impact of different patterns of lesion placements on the efficacy and consistency of catheter-based radiofrequency RDN in pigs.

METHODS The impact of increasing number of lesions versus location of RDN was investigated in a porcine model (Group 1; n = 51). The effect of treating the main artery, the branches, and the 2 combined was compared in Group 2 (n = 48). The durability of response and safety of combined treatment of the main artery plus branches was examined in Group 3 (n = 16). Renal norepinephrine (NE) tissue content and renal cortical axon density were assessed.

RESULTS Increasing the number of RF lesions (4, 8, and 12) in the main renal artery was not sufficient to yield a clear dose-response relationship on NE content and axon density. In contrast, targeted treatment of the renal artery branches or distal segment of the main renal artery resulted in markedly less variability of response and significantly greater reduction of both NE and axon density than conventional treatment of only the main renal artery. Combination treatment (main artery plus branches) produced the greatest change in renal NE and axon density with the least heterogeneity. The changes were durable through 28 days post-treatment.

CONCLUSIONS These data provide the rationale for investigation of an optimized approach for RDN in future clinical studies. This may have profound implications for the clinical application of RDN, as this approach may not only achieve greater reductions in sympathetic activity but also reduce treatment effect variability. (J Am Coll Cardiol 2015;66:1766–75) © 2015 by the American College of Cardiology Foundation.

Catheter-based renal denervation (RDN) was developed to offer patients with uncontrolled hypertension a new treatment option (1–3). The underlying pathophysiological concept is well established: targeting the renal sympathetic nerves reduces sympathetic efferent and sensory afferent signaling to and from the kidneys (4–6).

However, the clinical evidence supporting RDN as an effective interventional technique to disrupt the renal sympathetic nerves in patients with resistant hypertension is conflicting (7). A number of observational studies and randomized, controlled trials (8–10) support the concept, but some smaller studies and the large, single-blind, randomized, sham-controlled SYMPLICITY HTN-3 (Renal Denervation in Patients With Uncontrolled Hypertension) trial (11) failed to prove the efficacy of RDN. Many hypotheses have been proposed to explain these differences, including an insufficient RDN procedure, as potential causes for nonresponse to treatment (7,12,13). Although numerous efficacy markers have shown some promising results in smaller studies, including baroreflex function (14) and electric stimulation of the renal artery (15), there are no means of determining...
the effectiveness or completeness of renal sympathetic nerve destruction intraprocedurally with the currently available systems. This may lead to insufficient ablation if the depth and width of the ablative lesions are insufficient to engage the majority of renal nerves, perhaps explaining the highly variable treatment results (16,17).

Recently, variations in the distribution and density of the renal sympathetic nervous system, RDN’s ultimate target, have been assessed in human tissue (18). The mean distance from renal artery lumen to nerve location is least in the distal segments compared with the proximal and middle segments, although fewer nerves surround the renal artery in the distal segments versus the proximal and middle segments (18,19). These findings support the notion that more consistent ablation of sympathetic renal nerves might be achieved with ablation in distal positions, potentially in the branches along the renal vascular architecture rather than in mid or proximal positions (7).

However, few studies have assessed the role of different techniques for applying radiofrequency (RF) ablation along the renal arteries, with special emphasis on the branches (7,20). The aim of the present study was to investigate the impact of different patterns of lesion placements on the efficacy and consistency of catheter-based RF RDN in pigs.

METHODS

All animal studies were performed in accordance with the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health (Publication Number 85-23, revised 1996). Male (castrated) or female (nulliparous) Yorkshire domestic farm swine, weighing between 35 and 50 kg, were used for all studies. All animal experiments were performed at Synchrony Labs (Durham, North Carolina) and adhered to the Guide for the Care and Use of Laboratory Animals under an approved Institutional Animal Care and Use Committee protocol, in compliance with the Animal Welfare Act and the U.S. Food and Drug Administration regulations and their amendments. These studies were conducted in 3 parts to assess: 1) the relative influence of dose and anatomic location on ablative success; 2) whether the lesion placement strategy may be optimized; and 3) long-term durability. The efficacy of RDN was determined by a statistical decrease in renal cortical norepinephrine (NE) and renal cortical axonal density values compared with untreated naïve kidneys in the same subject. Response of arterial tissue to RF energy was evaluated through histopathologic analysis of treated vascular regions. Experimental arms for each study are outlined in Table 1. The same anesthesia protocol was used for all study arms.

COMPARISON OF DOSE AND LESION PLACEMENT. The study was designed to examine the impact of increasing numbers of RF lesions in the main renal artery as well as targeted application of RF lesions to either the renal artery branches or circumferential application of RF lesions in the distal main renal artery within 1.5 cm of the renal branch ostium. To control for potential variations in NE concentration and cortical axon density between swine, ablative treatments were performed only in the right renal arteries; thus, the untreated left artery and kidney would serve as the control within the test system. Once angiography was performed, a multielectrode RDN catheter (Spyral; Medtronic Cardiovascular, Santa Rosa, California) was advanced into the right renal artery, and treatment ablations were performed as follows: in Group 1, 1 60-s treatment in the main right renal artery (4 lesions; n = 12); in Group 2, 2 60-s treatments in the main right renal artery (8 lesions; n = 12); in Group 3, 3 60-s treatments in the main right renal artery (12 lesions; n = 12); and in Group 4, 1 60-s treatment in each branch of the right renal artery (4 lesions each; n = 15). Multiple cycles of catheter treatments were performed by positioning initially in the distal portion of the main right renal artery. After 1 60-s cycle of treatment was performed, a second cycle of treatment was delivered by moving the catheter proximally by approximately 2 mm and rotating about 45° before the application of energy in Group 2.
TABLE 1  
Animal Study Design

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Objective</th>
<th>Termination</th>
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<tr>
<td>Study 1</td>
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<tr>
<td>1) 4 lesions/main artery</td>
<td>12</td>
<td>Compare impact of number of lesions vs. location of lesions</td>
<td>Day 7</td>
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<tr>
<td>2) 8 lesions/main artery</td>
<td>12</td>
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<tr>
<td>3) 12 lesions/main artery</td>
<td>12</td>
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<tr>
<td>4) 4 lesions/branches</td>
<td>15</td>
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<td>Study 2</td>
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<tr>
<td>I) 4 lesions/main artery</td>
<td>12</td>
<td>Compare effect of combined treatment of main with branches vs. main alone</td>
<td>Day 7</td>
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<tr>
<td>II) 4 lesions/branches</td>
<td>12</td>
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<tr>
<td>Y) 4 lesions/main + 4/branch</td>
<td>12</td>
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<tr>
<td>Y2) 8 lesions/main + 4/branch</td>
<td>12</td>
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<tr>
<td>Study 3</td>
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<tr>
<td>1) 8 lesions/main + 4/branch</td>
<td>8</td>
<td>Durability of response</td>
<td>Day 28</td>
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<tr>
<td>2) Naive</td>
<td>8</td>
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Likewise, in Group 3, this process was repeated to provide a third cycle of treatment in the main renal arteries. Seven days after the RF ablation treatments were performed, all test animals underwent follow-up angiography to check for vessel patency, and tissues were collected from all animals following termination for histological and bioanalytical analysis.

The relative impact of targeted treatment on the main renal artery (Group I), renal artery branches (Group V), or combined main and branch artery treatments (Group Y) was compared as follows: in Group I, 1 60-s catheter treatment in the main right renal artery (4 lesions; n = 12); in Group V, 1 60-s treatment in each branch of the right renal artery (4 lesions each; n = 12); in Group Y, 1 60-s treatment in the main right renal artery (4 lesions) combined with 1 60-s treatment in each branch of the right renal artery (4 lesions each; n = 12); and in Group Y2, 2 60-s treatments in the main right renal artery (2 cycles, 8 lesions) combined with 1 60-s treatment in each branch of the right renal artery (4 lesions each; n = 12). RF treatments were performed only in the right renal arteries, leaving the untreated left artery and kidney to serve as the control. Seven days after the RF ablation treatments were performed, all test animals underwent the same follow-up and tissue collection outlined earlier (Table 1).

Long-term efficacy was assessed on day 28 by examining the morphological and physiological effects of the Spyral catheter treatments on the renal nerves (21). Bilateral treatment was applied to the main renal arteries (2 cycles, 8 lesions) and renal artery branches (4 lesions each) in 8 swine; they were compared with 8 swine used as a naive control group. RDN was quantified on the basis of renal tissue NE concentration and renal sympathetic axon density at 7 or 28 days following the procedure.

ASSESSING RENAL NE AND CORTICAL AXON DENSITY. Tissue samples were obtained from each kidney adjacent to those taken for axon density and kept frozen at −80 °C. The samples were homogenized in 0.4 mol/l perchloric acid and centrifuged to produce a clear supernatant; the supernatants were analyzed for norepinephrine (NE) content using high-performance liquid chromatography. Quantitative determination of NE was performed as follows. A mobile phase solution was run with the samples using the following parameters: Dionex WPS-3000TBS set at 4 °C pump and autosampler (Thermo Fisher Scientific, Waltham, Massachusetts), ESA Coulochem III detector (Thermo Fisher Scientific), and Gemini C18 (Phenomenex, Torrance, California) 5 μm 150 × 3.0 mm column at a flow rate of 1 ml/min and an average run time of 16 min.

The function of the renal sympathetic nerves was assessed by quantitative immunohistochemistry (IHC) to determine the density of terminal axons in the renal cortex. Eight embedded tissue samples from each kidney were cut on a rotary microtome at 5 to 6 μm. All sections were stained immunohistochemically for tyrosine hydroxylase as an indicator of viable sympathetic nerves. All sections were digitized by scanning on Aperio ScanScope AT (Leica Biosystems, Buffalo Grove, Illinois) and saved as whole-slide images. The number of intact (tyrosine hydroxylase positive) renal nerves was assessed on the whole-slide images using Aperio ImageScope software and a customized positive pixel count algorithm (Leica Biosystems). The terminal sympathetic axons were quantified in the renal cortex as the density of positive staining in the observed sections normalized to observed area.

STATISTICAL ANALYSIS. All reported values are expressed as mean ± SD calculated from the raw data. Normalized differences between the control and treatment groups were calculated and are reported for convenience of relative comparison; however, all statistical analysis was performed on non-normalized values. Differences in renal NE concentration and axonal density were evaluated using unpaired Student t tests with Welch’s correction. P values for all comparisons were 2 sided, and values obtained from different tissues of the same animal were considered independent for statistical purposes.

RESULTS

Renal cortical NE concentrations were significantly reduced (Figure 1A) following main artery treatments performed with the Spyral catheter in response to
4 lesions (150 ± 112 pg/mg; p = 0.0017), 8 lesions (203 ± 89 pg/mg; p = 0.0124), and 12 lesions (125 ± 72 pg/mg; p = 0.0005), respectively. Although delivery of RF energy to the main renal artery significantly decreased cortical NE concentrations, a clear dose-response relationship to increasing numbers of lesions in the main renal artery was not apparent. In contrast, renal cortical tissue collected on day 7 revealed a significant decrease (Figure 1A) in NE concentration in kidneys receiving treatment in the renal artery branches compared with untreated naive kidneys (46 ± 48 pg/mg vs. 266 ± 63 pg/mg; p = 0.0001). Renal artery branch treatment resulted in significantly greater reductions in renal NE than a single-cycle (4-lesion) treatment of the main renal artery (46 ± 48 pg/mg vs. 150 ± 112 pg/mg; p = 0.0093).

Cortical axon density closely paralleled renal NE responses (Figure 1B), suggesting that the reduction in cortical NE concentration is linked with an anatomic reduction in the number of viable sympathetic nerves in the renal cortex. Significant reductions in axon density were observed with main renal artery treatment (1 and 3 cycles of treatment). Compared with untreated naive kidneys, renal cortical axon density determined by IHC was significantly reduced in main artery treatments in response to 4 lesions in the main artery (78.8 ± 47.08 µm²/mm²; p = 0.0096) and

![Figure 1](https://example.com/figure1.png)

**Figure 1** Response to Renal Denervation Treatment: Number of Lesions

<table>
<thead>
<tr>
<th>Treatment</th>
<th>NE Concentration (pg/mg)</th>
<th>Cortical Axon Area (µm²/mm²)</th>
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<tr>
<td>Control</td>
<td>600</td>
<td>300</td>
</tr>
<tr>
<td>1 Cycle</td>
<td>400</td>
<td>200</td>
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<tr>
<td>2 Cycle</td>
<td>200</td>
<td>100</td>
</tr>
<tr>
<td>3 Cycle</td>
<td>100</td>
<td>0</td>
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<tr>
<td>Branch</td>
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</table>

Unpaired Student t-test with Welch’s correction vs. matched control
* P=0.0017  # P=0.0124  @P=0.0005  &P=0.0001
Unpaired Student t-test with Welch’s correction vs. Group 1 treatment
$P=0.0093

Response to increasing the number of lesions in the main artery and targeted treatment of branches or distal main artery shows significant reductions in renal norepinephrine (NE) concentration (A) and renal cortical axon density (B). Responses 7 days post-treatment; points represent individual kidneys.
12 lesions in the main artery (70.31 ± 37.52 µm²/mm²; $p = 0.0015$). Treatment with 8 lesions in the main artery produced only a marginally significant decrease in axon density (94.15 ± 38.87 µm²/mm²; $p = 0.0634$). Renal cortical tissue revealed a significant decrease in NE concentration in arteries receiving treatment (all groups; n = 12 each) compared with untreated naive kidneys (26.13 ± 16.38 µm²/mm² vs. 136.89 ± 32.35 µm²/mm²; $p < 0.0001$). Additionally, treatment of the renal artery branches resulted in significantly greater reductions in axon area than a single-cycle (4-lesion) treatment of the main renal artery (26.13 ± 16.38 µm²/mm² vs. 78.8 ± 47.08 µm²/mm²; $p = 0.0026$). Bioanalytical analyses of renal cortical tissue collected on day 7 revealed a significant decrease in NE concentration in arteries receiving treatment (all groups; n = 12 each) compared with untreated naive kidneys (n = 12 each) (Figure 2A).
When the treated groups were compared with the contralateral control, a pronounced and highly consistent reduction in NE concentration was observed: in Group I, 108 ± 95 pg/mg (p < 0.0001); in Group V, 55 ± 71.3 pg/mg (p < 0.0001); in Group Y, 25 ± 30 pg/mg (p < 0.0001); and in Group Y2, 28 ± 36 pg/mg (p < 0.0001). The greatest magnitude response with the least variability was produced with a branch and single-cycle treatment in the main renal artery (Group Y), resulting in significantly greater reduction in renal NE than treatment of the main renal artery alone (Group I; p = 0.0200). Adding a second cycle of treatment to the main renal artery and branch treatment (Group Y) did not improve the magnitude of NE decrease over single-cycle combination treatment (Group Y) but did significantly improve the renal NE reduction compared with main artery treatment alone (p = 0.0269). IHC of the renal cortical tissue revealed a significant reduction in axon density in all treatment groups compared with the corresponding naive kidneys (Figure 2B) (p < 0.0001): in Group I, 79.32 ± 57.2 μm²/mm²; in Group V, 51.76 ± 37.62 μm²/mm²; in Group Y, 34.35 ± 18.20 μm²/mm²; and in Group Y2, 42.61 ± 25.15 μm²/mm². Quantitatively, the greatest magnitude response with the least variability was produced with a branch and single-cycle treatment in the main renal artery, with significantly lower axonal density than observed with treatment of the main artery alone (p = 0.0220). Bioanalytical analyses of renal tissue collected on day 28 revealed a significant decrease in mean NE kidney tissue concentration following main renal artery and branch treatment compared with untreated naive kidneys (48 ± 53 pg/mg vs. 324 ± 88 pg/mg; p < 0.0001), an 85% reduction (Figure 3A). Similarly, IHC of the renal cortical tissue revealed a significant reduction (p < 0.0001) in axon density in arteries receiving treatment (43.44 ± 46.03 μm²/mm² vs. 336.5 ± 91.90 μm²/mm²; p = 0.0001). Mean reduction in renal cortical axon density following treatment was 87% (Figure 3B).

### SAFETY ASSESSMENTS AT DAYS 7 AND 28

Angiography at 7 days post-treatment showed all vessels fully perfused, with no evidence of aneurysm, procedural injury, dissection, appreciable arterial dilation, or filling defects. Semiquantitative histopathologic assessments were used to determine biological effects and healing response to the RF treatment. RF changes were observed in all treated vessels, with no changes observed in the untreated vessels (naive). RF changes were characterized by localized to widespread focal coagulation necrosis in the media (media hyalinization) and complete or near complete endothelial coverage. RF changes variably extended into adjacent tissues, evidenced by adventitial and perivascular collagen hyalinization, adipose tissue necrosis with frequent involvement of small perivascular branches (fibrinoid necrosis of the vessel with minimal to occasionally mild perivascular inflammation and rare thrombosis) or nerves (necrosis and/or inflammation), and inconsistent minor involvement of the lymph node or muscle. The average depth of the RF lesion was comparable in all treatment groups (3.54 ± 3.98 mm).

Arteriography before termination on day 28 revealed no angiographic abnormalities in most treated vessels, with mild narrowing noted in 1 main artery and 1 branch. All treated vessels were fully patent, with complete perfusion and no intraluminal filling defects or aneurysms. RF-related changes were observed histologically in all treated vessels and in both cranial and caudal branches past the main renal bifurcation; no changes were seen in the untreated vessels. The treated vessels showed no procedural injury, dissection, aneurysm formation, thrombosis, or appreciable arterial dilation.

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**Unpaired Student t-test with Welch’s correction to matched control**

* P<0.0001

At 28 days, the combined treatment of both the main renal artery and branches reduced renal tissue norepinephrine (NE) concentration (A) and cortical axon density (B) compared with matched naive swine. Points represent individual kidneys.
with full re-endothelialization at 28 days. Typical mural changes at 28 days consisted of segmental media fibrosis with no or minimal hyalinization, consistent with healing from direct RF injury. There was very little neointimal formation at 28 days; arterial wall media thickness was, on average, marginally increased by approximately 20 μm at the electrode contact point in areas of RF-induced media fibrous healing compared with the opposing wall (272 ± 45 μm vs. 253 ± 114 μm total thickness on average; p = 0.5423) or <10% increase in medial thickness. At 28 days, RF changes were characterized by localized to mild healing coagulation necrosis (hyalinization) and local to widespread...

CENTRAL ILLUSTRATION  Optimized Renal Denervation Techniques: Efficacy of Catheter-Based Radiofrequency
Renal Denervation

In this study, we evaluated different patterns involving numbers and locations of lesions to determine efficacy and safety of catheter-based radiofrequency (RF) renal denervation (RDN) in pigs. Renal norepinephrine (NE) concentration decreased in response to increasing number of lesions in the main artery and targeted treatment of branches or distal main artery (Top). Delivery of RF energy to the main renal artery significantly decreased cortical NE concentrations, although a clear dose-response relationship to increasing number of lesions in the main renal artery was not apparent; treating the renal artery branches resulted in even greater NE reductions. (Bottom) The greatest magnitude in response with the least variability was produced with a branch and single-cycle treatment in the main renal artery (Group Y); adding a second cycle of treatment (Group Y2) did not improve the magnitude of NE decrease but also significantly improved the renal NE reduction compared with main artery treatment alone (Group I). Percentage reduction = (mean control NE – test sample NE)/mean control NE × 100.

fibrosis in the media, with complete endothelial coverage and minimal amount of fully mature neointima. RF changes variably extended into adjacent tissues and were largely healed, evidenced by adventitial and perivascular fibrosis with little to no residual collagen hyalinization or adipose tissue necrosis with frequent involvement of nerves (fibrosis and/or residual necrosis and/or inflammation) and inconsistent or minor involvement of the lymph node or muscle. The average depth of the RF lesion was 2.10 ± 0.49 mm, which reflected the ongoing fibrous healing of the RF-exposed perivascular tissue.

**DISCUSSION**

Catheter-based RDN, targeting the renal afferent and efferent sympathetic nerves, has evolved as a new treatment option for patients with hypertension (1-3,22). The available clinical data suggest that it reduces office and ambulatory blood pressure in many, but not all, patients (9,11,22–25). Whether variable clinical response following RDN is a result of incomplete renal nerve ablation is uncertain and further challenged by the absence of practical measures of procedural efficacy. In SYMPLICITY HTN-1 (26), RDN using a single-electrode catheter reduced NE spillover by 40% on average, but the effect was highly variable, ranging from 0% to 80% (16). The same degree of treatment variability also has been documented in pre-clinical studies in pigs (7) and goats (27). However, pre-clinical data, which would allow us to guide and optimize RDN procedures, are incomplete, leading to anecdotal treatment recommendations (19). Herein, we provide evidence that targeted RF treatment on the distal elements of the renal artery (i.e., the distal segment of the main renal artery or the renal artery post-bifurcation) results in significant and relatively uniform reductions in NE and cortical sympathetic axon density and tissue content (Central Illustration). Treatment in these anatomic segments was also associated with a favorable safety profile at 28 days.

The location and distribution of renal sympathetic nerves represents a major underlying substrate for effective RDN procedures. The variation in distribution and density of the renal sympathetic nervous system in 20 human autopsy subjects was recently assessed in detail (17,18). The largest average number of nerves was observed in the proximal and middle segments of the renal artery and the smallest number in the distal segments. The mean distance from the lumen to the nerve was longest in the proximal and shortest in the distal segments. The ablation depth of the currently available RF RDN systems varies between 2 and 4 mm (7,28), thus limiting the accessibility of renal nerves by RF energy delivery in some regions of the renal artery, which suggests that asymmetrical targeting, and most probably distal renal artery targeting, is required to achieve successful denervation of renal afferent and efferent nerves (20). Furthermore, it has been shown that the nerves and ganglia are more abundant at the proximal superior renal artery ostium (19), and while ostial ablations extending up to 5 mm from the lumen affected ≤40% of nerves, a 5-mm ablation applied ≥6 mm from the aorta affected up to 85% of nerves. This implies that, in contrast to early anecdotal experiences, the ostial anatomic site may be unfavorable for intravascular RDN, as the nerves lie beyond the reach of typical ablations. However, it remained unclear whether these anatomic findings should affect clinical practice in performing the procedure. Herein, compared with naive kidneys, RF ablative treatments performed in the main renal artery, the renal artery branches, and a combination of these sites yielded statistically significant reductions in renal NE content and renal cortical axon density, indicating decreased renal sympathetic axons. The greatest magnitude response, though, with the least variability was produced with a branch and single-cycle treatment in the main renal artery (Group Y), resulting in significantly greater reduction in renal NE than main renal artery treatment alone (Group I; p = 0.02). Adding a second cycle of treatment (Group Y2) did not improve the magnitude of NE decrease over single-cycle combination treatment (Group Y) but did significantly improve the renal NE reduction compared with main renal artery treatment alone (p = 0.0269). Quantitatively, the greatest magnitude response with the least variability was seen in Group Y, with significantly lower axonal density than observed in Group I (p = 0.0220).

Currently available studies suggest that RDN can be delivered safely with minimal procedural complications and no detrimental effect on renal
function (9,11,25,29). In the present pre-clinical study, arteriography before termination on day 28 indicated that all treated vessels were fully patent with complete perfusion and showed no procedural injury, dissection, aneurysm formation, thrombosis, or appreciable arterial dilation. Mild narrowing was noted in 1 main artery and 1 branch. Full re-endothelialization was documented at 28 days, and typical mural changes consisted of segmental media fibrosis with no or minimal hyalinization, consistent with healing from direct RF injury. Despite the favorable safety profile documented herein in pigs, the long-term vascular safety in humans remains to be confirmed, as concerns have been raised that the procedure might induce renal artery stenosis in some patients (30). This is of special importance following treatment in distal renal artery segments, as many of the previous clinical trials did not ablate in this area.

Clinical application of a combination treatment strategy using treatment of the main renal artery and branches is being investigated in clinical trials (NCT02439749 and NCT02439775), with emphasis on long-term efficacy and safety of this new optimized approach.

The presented pre-clinical data provide the rationale for conducting clinical studies to determine the optimal RDN approach. This may have profound implications for the future clinical application of RDN if use of this optimized technique reduces treatment effect variability (17,20).

**STUDY LIMITATIONS.** RDN studies conducted in normotensive, young, healthy porcine models assume that similar renal nerve anatomy and function provide a reasonable simulation to the intended patients. Although these results suggest how the RDN procedure may be optimized in clinical practice, whether this approach will be effective in patients remains unresolved and deserves further investigation. The renal arteries in young, healthy swine may be different from those of the likely human recipient of RDN treatment with regard to atheroma, calcification, fibrosis, mechanical compliance, length, and tortuosity. Additionally, these parameters may also differ in hypertensive swine or humans. As a result, the absolute effectiveness of RF-mediated ablation should be viewed as optimal in the porcine model and may be affected by disease and anatomic limitations in the human setting. In the present study, we did not examine responses beyond day 28, and no conclusions may be made about the durability of the denervation response at longer time intervals (31). Blood pressure monitoring of the normotensive animals was not a part of the present study, and the correlation between reductions in kidney NE tissue concentration and blood pressure, as well as the degree of renal nerve damage to provide meaningful blood pressure changes, remain to be investigated.

**CONCLUSIONS**

Increasing the number of RF lesions in the main renal artery, although consistently effective in reducing renal NE and axon density relative to naive kidneys, was not sufficient to yield a clear dose-response relationship. In contrast, targeted treatment of the renal artery branches or distal segment of the main renal artery resulted in markedly less variability of response and significantly greater reduction of both NE and axon density than conventional treatment of only the main renal artery. Combination treatment of both the main renal artery and associated branches produced the greatest decline in renal NE and reduction of axon density with least variability of the treatments tested being durable through 28 days post-treatment.

**COMPETENCY IN MEDICAL KNOWLEDGE:** One potential reason for lack of response to RDN in patients with hypertension is incomplete ablation resulting from the inconsistent distribution and density of the renal sympathetic nerves in distal versus proximal portions of the main renal artery and its branches. Targeting ablation to the distal segment of the main renal artery and post-bifurcation branches results in greater and more uniform reductions in renal cortical sympathetic axon density and tissue NE levels.

**TRANSLATIONAL OUTLOOK:** The long-term efficacy and safety of this targeted approach to renal sympathetic denervation should be investigated in clinical trials involving patients with hypertension.
REFERENCES


