Remote heating of metal placed near a source of radiofrequency energy within the heart has been previously described. This phenomenon has led to some safety concerns (MRI) and for those undergoing catheter ablation. However, for patients undergoing cardiac magnetic resonance imaging and iron staining. Irrigated ablation in the regions with greater FeO, after FeO infusion and magnetic guidance, created larger lesions without a greater incidence of steam pops.

Conclusions—Metal nanoparticle infiltration resulted in significantly larger ablation lesions with altered electric and thermal conductivity. In vivo magnetic guidance of FeO nanoparticles allowed for facilitated radiofrequency ablation without direct infiltration into the targeted tissue. Further research is needed to assess the clinical applicability of this ablation strategy using metallic nanoparticles for the treatment of cardiac arrhythmias. (Circ Arrhythm Electrophysiol. 2016;9:e003820. DOI: 10.1161/CIRCEP.115.003820.)

Key Words: cardiac arrhythmias ■ iron ■ magnet ■ magnetic resonance imaging ■ nanoparticle

Therefore, we sought to characterize ablation lesion formation characteristics using tissue sensitized to radiofrequency energy after treatment with different metals. We also utilized magnets to guide the distribution of systemically administered iron oxide (FeO) nanoparticles for in vivo facilitated ablation.

Methods

Ex Vivo Model

Experimental protocols have been approved by the Institutional Animal Care and Use Committees of the University of Colorado. An

Received August 21, 2015; accepted February 25, 2016.

From the Section of Cardiac Electrophysiology, Division of Cardiology, Department of Medicine, University of Colorado, Aurora.


Correspondence to William H. Sauer, MD, Section of Cardiac Electrophysiology, University of Colorado, 12401 E 17th Ave, B136, Aurora, CO 80045.

E-mail william.sauer@ucdenver.edu

© 2016 American Heart Association, Inc.

Circ Arrhythm Electrophysiol is available at http://circep.ahajournals.org

DOI: 10.1161/CIRCEP.115.003820
ex vivo model consisting of viable bovine myocardium, a circulating saline bath at 37°C, a submersible load cell, and a deflectable sheath was assembled. The circulating bath utilized a perfusion pump designed for cardiac bypass and circulated fluid in a saline bath at a rate of 5 L/min. A load cell was submersed in the bath and contained a section of viable bovine ventricular myocardium excised within 1 hour of experimentation. This load cell measured force applied to the overlying myocardial tissue and was used to standardize application of energy. This ex vivo model has been validated and described in further detail elsewhere.13 A 4-mm nonirrigated radiofrequency ablation catheter (Biosense-Webster, Diamond Bar, CA) was positioned with 10 g of force in a perpendicular position using a deflectable sheath (Agilis; St. Jude Medical, St Paul, MN).

**Metal Nanoparticle Preparation**

FeO nanoparticles are superparamagnetic, dispersed in normal saline, and are 10 to 15 nm in length (Nanomaterials, Inc, TX). Titanium (Ti) nanoparticles are 30 to 50 nm in length and were obtained from US Research Nanomaterials, Inc; copper nanoparticles (CuNPs) are 5 to 7 nm in length and are dispersed in an organic medium, phosphate ester (SkySpring Nanomaterials, Inc, TX). Nanoparticles were suspended or further diluted in normal saline or water.

**Liposomal Iron Preparation**

Liposomal FeO nanoparticles were commercially obtained (Avanti Polar Lipids, Alabaster, AL). To prepare the formulation, the lipid components were measured in chloroform solutions of known concentration or weighed in dry solid state and dissolved in minimal chloroform. Chloroform was evaporated from the mixture using a gentle stream of nitrogen, thus leaving a thin lipid film on the walls of the bottle. The dry lipid film was dissolved in cyclohexane and then allowed to freeze at −50°C. The frozen lipid mixture was freeze-dried for approximately 60°C for a total of 2 passes to yield an effective diameter of 188.7 nm (SD, 1.4 nm), polydispersity of 0.336 (SD, 0.004). Gravimetric analysis presented a concentration of liposomal iron nanoparticles at 1.48 mg/mL. To assess for uniformity, particle size was measured using a dynamic light scattering instrument, which reports an average particle size.

**Delivery of Radiofrequency Energy Applied to Infiltrated Myocardium**

A series of ablation lesions using low power (20 W) and high power (50 W) were created on recently excised bovine myocardium. Immediately before radiofrequency energy delivery, the myocardium was infiltrated using a 29-gauge hypodermic needle at a depth of 5 mm with 1 mL of solution containing 100 μg of CuNP, 100 μg of FeO, or 100 μg of TiNPs. Separate ablation lesions on the same myocardial tissue were created using sham injection or 1 mL of injections of 0.9% saline for comparison. The number of lesions applied per ventricular section depended on the available endocardial surface. No lesions were placed over or in immediate proximity (5 mm) to papillary muscles or other lesions. Furthermore, no lesions were placed within 1 cm of section edge.

**Tissue Temperature Analysis**

T-type thermocouple wires were inserted horizontally into myocardium at 3 and 5 mm depths and perpendicular to the ablation surface. Thermocouple analogue inputs were converted to digital signals using LabView software (version 7.0). Temperatures were recorded in a continuous fashion throughout the 60 s of radiofrequency application at a rate of 5 Hz. Peak tissue temperature was defined as the maximum temperature reading during radiofrequency application. Radiofrequency applications that generated steam pops were excluded from temperature curve analysis.

**In Vivo Magnet-Guided Ablation**

Yorkshire pigs (n=4) were anesthetized and bilateral femoral artery access was obtained. Porcine thigh preps were prepared bilaterally and modified from previously described canine thigh preps.16 In brief, the skin and connective tissue were dissected to expose the underlying muscle. The skin was raised, to form a cradle, and heparinized; warmed porcine blood was circulated at 350 mL/min. Before performing ablation studies, methylene blue infusion was performed via the ipsilateral femoral artery to demonstrate that there was methylene blue distribution in the ipsilateral thigh prep.

An ablation catheter was placed perpendicular to the muscle surface. Ablations were delivered at 30 W for 20 seconds with the same amount of force, as measured by a force sensing, open irrigated-tip radiofrequency catheter (SmartTouch Thermocool; Biosense-Webster); ablation lesions were tagged by the electroanatomic mapping system and averaged between 10 and 20 g of force. Animals were pretreated with 10 mg of dexamethasone and 100 μg epinephrine to prevent potential FeO-induced anaphylaxis and hypotension. During ablation, infusion of 200 mg of FeO nanoparticles (dispersed in 20 mL and infused at 1 mL/s) or 0.9% normal saline was performed via the ipsilateral femoral artery. In addition, magnets (neodymium with 310 pound pull force; CMS Magnetics, Inc, TX) were placed immediately adjacent to the ablation catheter with an orientation parallel to the tissue plane to attract nanoparticle aggregation as close as possible to the targeted ablation site.

**In Vivo Ablation After Liposomal Infusion**

In separate thigh preps, and before liposomal iron infusion, intravenous dexamethasone was given to prevent possible anaphylactic reaction to the liposomes. Control ablations, using an irrigated catheter at 30 W for 60 seconds with 10 to 20 g of force, were performed on thigh preps before infusion of liposomes. Next, liposomal iron (10 mg/mL) was infused at a rate of 1 mL/min for 30 minutes before ablation. Ablation was then performed on thigh preps using the...
same parameters as control ablations. Ablation lesion data, including force, impedance, and power, were recorded by the electroanatomic mapping system. Of note, we had used 20 seconds for the ipsilateral arterial thigh infusion of FeO because this was a higher concentration infusion of pure FeO nanoparticles and hence we expected the effect to be larger and, therefore, would require less ablation time to see an effect. With systemic liposomal FeO infusion (given intravenously), we expected that a smaller amount (relative to the direct arterial injection) of liposomal FeO would reach the thigh and, therefore, we wanted to have a longer time (60 seconds) for radiofrequency to heat the liposomes to release their contents in order for them to have an effect.

### Magnetic Resonance Imaging

After animals were euthanized, thigh preps were resected and placed in 0.9% saline for immediate imaging. MRI was performed using a 3.0T Skyra (Siemens Medical, Erlanger, Germany) to identify scar and iron deposition. Susceptibility-weighted imaging was performed using a swi3Dir sequence with repetition time of 28 ms and echo time of 20 ms at a field of view of 270x384 mm and slice thickness of 1.2 mm to assess for Fe deposition. Using this method, tissue injury seems as bright signal intensity, and Fe deposition seems dark; specific characteristics related to ablation can thus be assessed. For MRI sections, iron signal intensities, expressed as a percentage, were measured by dividing the sum of all dark iron intensities from all sections by the sum of ablation lesion areas from all sections and multiplying by 100.

### Histology and Microscopy

Tissues containing the control and FeO ablation lesions were collected immediately after ablation, to minimize a diffusion effect of FeO nanoparticles, and samples were snap frozen in liquid nitrogen. The tissue was sectioned, at 50-μm thickness, using a microtome and then fixed with 10% buffered formalin. Sections were then stained with Prussian blue and counterstained with nuclear fast red. A Zeiss Axiovert 200 mol/L microscope (Carl Zeiss MicroImaging, Inc, Thornwood, NY) was used for imaging. Stained slides were examined under light microscopy, digitized using a high-resolution scanner, and analyzed using Photoshop CS image analysis software (Adobe Systems Inc, San Jose, CA). Iron staining and total area of ablation lesions, for all sections, were manually traced in the digital images and automatically calculated by the software.

### Ablation Lesion Volume Measurements

Tissue sections were analyzed and lesions were measured with a digital micrometer by a blinded observer, who was blinded to the treatment group. The detection of metallic nanoparticles is only possible with examination of MRI or special histological stains; there is no method to identify scar and iron deposition. Susceptibility-weighted imaging was performed using a swi3Dir sequence with repetition time of 28 ms and echo time of 20 ms at a field of view of 270x384 mm and slice thickness of 1.2 mm to assess for Fe deposition. Using this method, tissue injury seems as bright signal intensity, and Fe deposition seems dark; specific characteristics related to ablation can thus be assessed. For MRI sections, iron signal intensities, expressed as a percentage, were measured by dividing the sum of all dark iron intensities from all sections by the sum of ablation lesion areas from all sections and multiplying by 100.

### Statistical Analysis

IBM SPSS Statistics and Microsoft Excel were used to perform statistical calculations. The t test was used to compare continuous variables (lesion characteristics, measured impedances, and temperatures) between different groups with equal variances within the groups not assumed. The χ² test was used for dichotomous comparisons of ablation lesion characteristics from metal nanoparticle–treated versus untreated tissue. When comparisons were made using pooled results from the same animal or slab, hierarchal analysis with adjustment for possible bias because of clustering was performed.

### Results

#### Effect of Metal Nanoparticle Infiltration on Ablation Lesion Characteristics and Electric Properties Using Low- and High-Power RF Energy

At both low and high powers, ablation lesions in myocardium treated with CuNP, FeO, or TiNP were significantly larger compared with tissue infiltrated with saline. Table 1 shows the differences in ablation lesion characteristics, using high-power radiofrequency (50 W) applied to the different myocardium treatments. Ablation after metal nanoparticle infiltration resulted in larger impedance changes compared with ablation of tissue infiltrated with untreated (sham) or saline. Figure 1 details the mean starting and ending impedance measurements in untreated control (sham), saline-infiltrated, and metal nanoparticle–treated myocardial tissue before and after radiofrequency ablation at 50 W.

#### Effect of Metal Nanoparticle Infiltration on Myocardial Tissue Temperature Dispersion

For all metals, the peak temperatures recorded at the ablation catheter tip were significantly higher in metal-treated myocardium than in saline-infiltrated myocardium using power-control mode, at both 20 and 50 W. When measuring temperatures at 3- and 5-mm depths, there were greater tissue temperatures in CuNP- and FeO-treated tissues than in sham- or saline-infiltrated tissue (Table 2). Figure 2 displays the mean temperature dispersion at 3- and 5-mm depths in FeO nanoparticle–treated myocardium compared with saline infiltration and untreated myocardium after radiofrequency at 50 W.

#### In Vivo FeO Delivery to Ablation Sites Within Thigh Preparations

Liposomal iron and FeO nanoparticles were directed to an area of interest using magnets, as shown in Movie I and Figure I in the Data Supplement. Staining of ablation lesion sections from porcine thighs also confirmed the presence of iron within tissues ablated after FeO infusion. The levels of iron were significantly greater, with 10.2% of tissues staining for iron, in tissues ablated after FeO infusion, compared with <1% background iron staining (from ruptured red blood cells) of control sections; *P*<0.001 (Figure 3A and 3B are representative slides).

In addition, MRI immediately after ablation procedures with liposomal iron infusion also demonstrated that iron was preferentially deposited in tissue adjacent to which a magnet had been applied at the time of ablation, and within the ablation target areas. Quantitative analysis from MRI sections show that iron signal intensities were significantly greater after a magnet was applied during liposomal iron infusion, covering 32.4% of total ablation lesion areas. This was in comparison with signal intensities covering only 10.3% of ablation lesion areas after liposomal iron infusion without use of a magnet, and <1% in controls; *P*<0.001. Figure 4A shows iron deposition at the thigh preparation surface and at a depth of 5 mm from the thigh preparation surface. Iron susceptibility seems dark on...
MRI, within areas of ablation where magnets were placed after systemic liposomal iron infusion. These dark intensities were significantly less in areas ablated after systemic liposomal iron was infused but magnets were not applied (Figure 4B).

**In Vivo Ablation Facilitated by Iron Delivered Directly or Via Iron-Containing Liposomes**

Iron delivered via FeO nanoparticle infusion in the ipsilateral femoral artery during porcine thigh ablation with irrigated-tip catheters, and directed by magnets to the areas of ablation, led to larger ablation lesions compared with untreated controls (201.5 versus 139.1 mm³; \(P < 0.001\)). There was no statistical difference in the average contact force between control and nanoparticle groups. Table 3 provides ablation lesion characteristics for control and magnet-guided FeO nanoparticle infusion.

Irrigated-tip ablation after iron delivery via systemic liposomes also led to larger lesions compared with control (379.7 mm³ versus 189.5 mm³; \(P < 0.001\)), and lesion characteristics are summarized in Table 4.

**Discussion**

**Study Results**

In this study, we have shown that a low dose of metallic nanoparticles (100 μg) infiltrated directly into myocardial

---

**Table 1. Myocardial Ablation Lesion Characteristics After Radiofrequency Energy Applied at 50 W for 60 s**

<table>
<thead>
<tr>
<th>Group</th>
<th>Average Maximum Depth, mm</th>
<th>Diameter Maximum, mm</th>
<th>Max Surface Diameter, mm</th>
<th>Steam Pop</th>
<th>Volume (mm³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sham (n=30)</td>
<td>6.0±0.3</td>
<td>10.8±0.8</td>
<td>6.5±0.4</td>
<td>4/98</td>
<td>267.5±43.7</td>
</tr>
<tr>
<td>NS (n=30)</td>
<td>6.1±0.2</td>
<td>10.7±0.6</td>
<td>7.5±0.9</td>
<td>2/66</td>
<td>261.6±31.8</td>
</tr>
<tr>
<td>100 μg Cu (n=30)</td>
<td>7.2±0.4</td>
<td>11.7±0.6</td>
<td>8.6±0.5</td>
<td>5/47</td>
<td>355.7±45.4</td>
</tr>
<tr>
<td>100 μg FeO (n=30)</td>
<td>6.3±0.4</td>
<td>11.4±1.1</td>
<td>7.5±0.8</td>
<td>3/48</td>
<td>302.5±56.2</td>
</tr>
<tr>
<td>100 μg Ti (n=30)</td>
<td>6.7±0.4</td>
<td>11.1±0.8</td>
<td>7.8±0.8</td>
<td>2/50</td>
<td>311.5±41.9</td>
</tr>
<tr>
<td>NS vs sham (P) value</td>
<td>0.092</td>
<td>0.671</td>
<td>&lt;0.001</td>
<td>0.725</td>
<td>0.556</td>
</tr>
<tr>
<td>100 μg Cu vs saline (P) value</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>0.985</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>100 μg FeO vs saline (P) value</td>
<td>0.014</td>
<td>0.008</td>
<td>0.890</td>
<td>0.407</td>
<td>0.001</td>
</tr>
<tr>
<td>100 μg Ti vs saline (P) value</td>
<td>&lt;0.001</td>
<td>0.045</td>
<td>0.105</td>
<td>0.777</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Cu indicates copper; FeO iron oxide; and Ti, titanium.

---

Figure 1. Mean starting and ending impedance measurements in untreated control (sham), saline-infiltrated, and metal nanoparticle-treated myocardial tissue before and after radiofrequency ablation at 50 W. The statistically significant \(P\) values (<0.05) are in bold. Cu indicates copper; FeO, iron oxide; and Ti, titanium.
tissue significantly altered tissue properties and the effects of applied RF ablation energy. Specifically, in our experiments, ablation of myocardial tissue infiltrated with metal nanoparticles resulted in an increase in ablation lesion size and improved tissue heating. In vivo, magnets were able to guide FeO nanoparticles and iron-containing liposomes to the site of targeted ablation, resulting in iron-facilitated ablation with increased lesion sizes.

### Potential Mechanisms of Metal Nanoparticle Facilitation of RF Ablation

In our ex vivo model, infiltration of myocardial tissue with metallic nanoparticles resulted in significantly larger ablation lesions. There are several potential mechanisms for metallic nanoparticle facilitation of radiofrequency ablation. Copper, iron, and titanium are metals that can conduct electricity with extremely high efficiency, thereby possibly allowing for deeper penetration of radiofrequency current through the circuit established by the catheter and grounding patch, if there is an uninterrupted connection between the nanoparticles and the ablation catheter tip. In addition, these metals exhibit extremely efficient thermal conductivity, which is another possible contributing factor to their enhancement of thermal injury in tissues receiving radiofrequency energy. Finally, metallic nanoparticles can affect radiofrequency current to allow for a greater surface area of the metal–tissue interface and thus resistively heated tissues exposed to the metallic nanoparticles. The presence of the metal functioning as a “lightning rod” within the tissue, therefore, affects radiofrequency current to cause resistive heating of tissue at the metal–tissue interface. This unique combination of favorable electrical and thermal conductive properties enhances the thermal injury to myocardial tissue treated with metallic nanoparticles when RF energy is applied to it. The enhanced effects of nanoparticles is likely due a combination of these effects beyond that of direct electric conduction because their effects are greater than those of a nonmetallic, ionic medium (normal saline) that was used during control ablations. The results of our experiments do not prove the exact relative contribution of each of these potential mechanisms.

In previous studies where saline infusion was utilized to create larger lesions, Sapp et al described the delivery of radiofrequency during the infusion of saline over the duration

### Table 2. Mean Maximum Temperature Dispersion at Surface, 3- and 5-mm Depths for 20 and 50 W

<table>
<thead>
<tr>
<th></th>
<th>Tip–Tissue Peak Temperature</th>
<th>3-mm Depth Peak Temperature</th>
<th>5-mm Depth Peak Temperature</th>
<th>Tip–Tissue Peak Temperature</th>
<th>3-mm Depth Peak Temperature</th>
<th>5-mm Depth Peak Temperature</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated (n=30)</td>
<td>51±6</td>
<td>58.5±3.8</td>
<td>50.1±3.0</td>
<td>50±3</td>
<td>74.0±3.5</td>
<td>63.9±2.8</td>
</tr>
<tr>
<td>Saline (n=30)</td>
<td>52±8</td>
<td>59.5±3.9</td>
<td>51.7±2.9</td>
<td>51±6</td>
<td>74.7±3.8</td>
<td>64.0±1.8</td>
</tr>
<tr>
<td>100 μg FeO (n=30)</td>
<td>54±9</td>
<td>61.2±3.6</td>
<td>53.2±3.1</td>
<td>50±5</td>
<td>77.9±4.3</td>
<td>67.1±4.5</td>
</tr>
<tr>
<td>100 μg Cu (n=30)</td>
<td>51±7</td>
<td>64.9±3.7</td>
<td>56.0±3.4</td>
<td>56±3</td>
<td>81.2±2.6</td>
<td>69.8±2.8</td>
</tr>
<tr>
<td>100 μg titanium (n=30)</td>
<td>52±8</td>
<td>61.5±2.6</td>
<td>55.8±3.3</td>
<td>49±3</td>
<td>79.7±3.8</td>
<td>68.7±6.4</td>
</tr>
<tr>
<td>Saline vs untreated P value</td>
<td>0.685</td>
<td>0.501</td>
<td>0.174</td>
<td>0.217</td>
<td>0.593</td>
<td>0.927</td>
</tr>
<tr>
<td>100 μg FeO vs saline P value</td>
<td>0.384</td>
<td>0.243</td>
<td>0.207</td>
<td>0.561</td>
<td>0.039</td>
<td>0.027</td>
</tr>
<tr>
<td>100 μg Cu vs saline P value</td>
<td>0.568</td>
<td>&lt;0.001</td>
<td>0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>100 μg titanium vs saline P value</td>
<td>0.960</td>
<td>0.126</td>
<td>0.001</td>
<td>0.037</td>
<td>0.001</td>
<td>0.016</td>
</tr>
</tbody>
</table>

Cu indicates copper; and FeO, iron oxide.

![Figure 2](http://circagr.aahajournals.org/content/10/1/96/F2.large.jpg)

**Figure 2.** Mean temperature dispersion at 3- and 5-mm depths in FeO nanoparticle–treated myocardium, compared with saline infiltration or no treatment after radiofrequency at 50 W. FeO indicates iron oxide.
of time that radiofrequency energy was delivered through a deployed and electrically active needle. This is in contrast to our ex vivo experiments where radiofrequency is delivered on the surface after an injection of a relatively small amount of saline that is unlikely to have an effect similar to what has been described by Sapp et al.12 with needle ablation. This small amount of saline, which was preinjected and not concurrent with ablation, would have minimal cooling effect, as it did in the studies by Sapp et al.12

We have also evaluated the infusate volume and geometry (Figure II in the Data Supplement). The infusate volume is mostly oval, instead of spherical, in shape, and this is likely related to the fact that gravity tends to pull the infusate down, with likely greater density toward the inferior portion of the infusate geometry. This shape is fairly consistent across the various infusates. The infusate volume can impact the ablation lesion volumes. This is most apparent for the 50 W lesions, but not for the 20 W lesions. However, as the infusate geometry is similar across infusates, the infusate geometry alone is unlikely to account for the difference in lesion sizes. Rather, at 50 W, the nanoparticle components within their infusate volume tend to increase the lesion size toward where the nanoparticle was injected and had settled, which we hypothesize is related to an ohmic heating mechanism. This increase in lesion size toward the site of nanoparticles within the infusate geometry is not as prominent for control, nonmetallic infusates.

Myocardium infiltrated with Cu nanoparticles had a higher impedance, compared with control myocardium. We think this is because of the fact that it was suspended in a nonconductive organic solvent (phosphate ester), which was subsequently diluted in saline, but the effects may have remained. In addition, injection of the copper nanoparticles into the tissue may have resulted in some oxidation altering cellular structure and acidic environment of the tissue, thereby possibly affecting electric current. With regards to the temperature at the catheter tip–tissue interface, which was also increased for copper nanoparticles at 50 W, we think that this reflects the increase in resistive heating near the ablation tip overwhelming any potential passive cooling or diffusion of heating deeper into the tissue (which may also have been inhibited by the solvent). The tip–tissue temperatures for iron and titanium were not higher, and for titanium, it was mildly lower than control at 50 W.

**Previous Investigations Into Ablation of Nanoparticle-Treated Tissue**

We have previously demonstrated that radiofrequency ablation near metallic objects can lead to collateral heating near the metal.13 We demonstrate in this article that a similar phenomenon can be utilized to enhance ablation efficacy.

Studies in cancer therapeutics have utilized various metals to improve thermal energy destruction of tumor cells. We first applied this concept to cardiac ablation using carbon nanotubes, which are not metallic but can enhance thermal conductivity within the target tissue and increase lesion size.14 The use of carbon nanotubes in humans has not been demonstrated and we, therefore, explored the use of gadolinium, a commonly used chelated metal in medical procedures, to facilitate ablation.5 We showed that ablation after in vivo injection of gadolinium led to increased lesion sizes in a porcine model. However, the high doses of gadolinium that would need to be directly injected to achieve sufficiently high myocardial distribution may limit its widespread utility.15 Hence, in this study, we demonstrated that small amounts of metals in the bloodstream can enhance ablation lesion sizes, allowing for the possibility of systemic administration.

In this study, we further established that a novel strategy using magnetic guidance can effectively direct distribution of metal nanoparticles to a targeted site of ablation. Iron was chosen to facilitate ablation because it has been used in humans, and FeO nanoparticles have been studied in cancer therapeutics.16,17 In addition, magnetic guidance of nanoparticles has been previously shown as a viable drug delivery system, including its use to direct ganglionic plexus toxicity.18 Heat-sensitive liposomes can be utilized to systemically
administer a pharmacological payload but yet have a localized effect by only releasing that payload on heat activation. Liposomes are biologically inactive and have minimal toxic effects in humans; in our porcine model, animals were pretreated with medications because pigs may have an allergic reaction to the liposomes. However, for humans, by encasing iron nanoparticles within a liposome, systemic side effects can be mitigated, although there may be some exposure to the metals once they are released when exposed to heat. Magnetic navigation of the FeO liposomes further enhances the localized effect of the metallic agents on ablation lesion formation. Our study demonstrates the potential utility of magnetically directed liposomal agents for augmenting radiofrequency ablation.

**Potential Clinical Implications for Metal Nanoparticle–Facilitated Ablation**

The goal of our study was to explore the possibility of improving the efficacy and safety of cardiac ablation using metal nanoparticles and specifically FeO in vivo. The use of metal nanoparticles in myocardial tissue offers potential in other aspects of electrophysiology. To realize the clinical applications for copper or titanium facilitation of radiofrequency ablation, a reliable delivery mechanism into targeted myocardial tissue would be needed. These agents lack the high ablating metallic nanoparticles through their binding to specific agonists for cardiac-specific receptors. Further research is needed to develop these potential strategies for metallic nanoparticle delivery into the myocardium, which is essential for the exploration of the clinical application of our observations. Establishing a dose response and potential risks of toxicity are important next steps and, before any consideration for clinical use, further animal studies exploring different doses to determine efficacy and toxicity in survival models are required.

**Limitations**

We attempted to control for variations in ablation lesion dimensions caused by external variables such as circulation rate, passive catheter cooling, and catheter contact by standardizing these variables on repetitive energy delivery. These limitations have been well described in previous studies using ex vivo and thigh preparations. To reduce the impact of variable lesion size, we repeated energy delivery under similar conditions for each radiofrequency application and applied the mean ablation lesion size when comparing the effects of metal nanoparticle infiltration. In addition, the variable conditions that may exist would be nondifferential among the individual experiments and is unlikely to explain our results.

This study used bovine myocardium in an ex vivo model and a porcine thigh preparation in an in vivo model to approximate RF ablation lesion characteristics produced during indicated procedures performed in patients with cardiac arrhythmias. It is impossible to reliably apply our findings to this clinical setting with any degree of accuracy. However, we think that the biophysical principles of RF energy delivery that we describe in both our ex vivo and in vivo models can be used for further in vivo experimental settings and ultimately in clinical studies.

Although our results are intriguing and have implications for clinically relevant ablation strategies, further studies involving metallic nanoparticle sensitization of myocardium need to be confirmed in other animal studies before considering human trials. Given the potential for side effects from metal nanoparticles deposited in myocardial tissues and the challenges to direct them specifically to the myocardium with risks of embolization, these remain as potential limitations in translating these findings to humans. Furthermore, although the research for nanoparticles in cancerous tumor destruction seems promising, there are significant differences in treating

### Table 3. Ablation Lesion Characteristics After Irrigated-Tip Radiofrequency Energy Applied at 30 W for 20 s (In Vivo Thigh Preparations and Iron Infusion)

<table>
<thead>
<tr>
<th></th>
<th>Average Maximum Depth, mm</th>
<th>Diameter Maximum, mm</th>
<th>Maximum Surface Diameter, mm</th>
<th>Volume (mm³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated (n=22)</td>
<td>4.4±0.6</td>
<td>9.6±0.8</td>
<td>7.1±0.9</td>
<td>139.1±30.8</td>
</tr>
<tr>
<td>FeO (n=22)</td>
<td>5.3±0.8</td>
<td>10.5±1.3</td>
<td>7.8±1.0</td>
<td>201.4±59.3</td>
</tr>
<tr>
<td>FeO vs untreated P value</td>
<td>&lt;0.001</td>
<td>0.005</td>
<td>0.026</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

FeO indicates iron oxide.

### Table 4. Ablation Lesion Characteristics After Radiofrequency Energy Applied at 30 W for 60 s (In Vivo Thigh Preparation and Liposomal Iron Infusion)

<table>
<thead>
<tr>
<th></th>
<th>Average Maximum Depth, mm</th>
<th>Diameter Maximum, mm</th>
<th>Maximum Surface Diameter, mm</th>
<th>Volume (mm³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated (n=20)</td>
<td>5.2±0.7</td>
<td>10.1±1.1</td>
<td>7.5±1.0</td>
<td>189.5±54.5</td>
</tr>
<tr>
<td>Liposomal iron (n=23)</td>
<td>7.1±1.1</td>
<td>13.1±1.5</td>
<td>9.1±1.5</td>
<td>379.7±114.8</td>
</tr>
<tr>
<td>Liposomal iron vs untreated P value</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>
cancerous tumors compared with targeting arrhythmogenic myocardial tissues that would need to be considered.

The electrode–tissue temperatures were significantly higher for copper nanoparticles at 50 W, but not for iron or titanium nanoparticles. In addition, there was no significant difference (all $P>0.05$) in steam pop rate between metallic nanoparticle–treated tissue and control. We think that the enhanced thermal conductivity minimizes steam pops despite possible higher tip–tissue temperatures in the case of the copper nanoparticles. External irrigation may further mitigate this risk. Therefore, we do not think that power delivery would be limited by steam pop rates.

Conclusions

In both ex vivo and in vivo models, myocardial exposure to metallic nanoparticles resulted in enhanced sensitivity to the thermal destruction of tissue from applied RF energy. Metallic nanoparticles significantly altered the electric properties of targeted myocardial tissue and resulted in an increased sensitivity to radiofrequency heating. Furthermore, metallic nanoparticle facilitated radiofrequency ablation of targeted tissue through resistive heating and improved thermal conductivity, resulting in larger ablation lesions compared with untreated tissue. Metallic nanoparticles can also be directed using magnetic guidance for targeted ablation. Additional research into the safe delivery of metallic nanoparticles into myocardial tissue in an animal model and in pathological models of diseases is required before further consideration to the potential clinical application of our findings.

Disclosures

Drs Sauer and Nguyen receive significant research grants from Biosense-Webster and CardioNXT, Inc, and educational grants from St Jude Medical, Boston Scientific, and Medtronic. The other authors report no conflicts.

References

15. Sacher F, Cochet H. After the fire and ice age, are we entering the metal age? JACC Clin Electrophysiol. 2015;1:185–186.
Enhanced Radiofrequency Ablation With Magnetically Directed Metallic Nanoparticles

Circ Arrhythm Electrophysiol. 2016;9:
doi: 10.1161/CIRCEP.115.003820

Circulation: Arrhythmia and Electrophysiology is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2016 American Heart Association, Inc. All rights reserved.
Print ISSN: 1941-3149. Online ISSN: 1941-3084

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circcep.ahajournals.org/content/9/5/e003820

Data Supplement (unedited) at:
http://circcep.ahajournals.org/content/suppl/2016/05/10/CIRCEP.115.003820.DC1

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation: Arrhythmia and Electrophysiology can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation: Arrhythmia and Electrophysiology is online at:
http://circcep.ahajournals.org/subscriptions/
Supplemental Material

Figure 1. Magnet Guiding Liposomes Containing FeO Nanoparticles. Liposomal FeO NPs can be directed to an area of interest using magnets, as depicted in this figure, where a magnet is attracting FeO-containing liposomes against gravity.
**Figure 2:** Gross histology of infusate geometries and respective ablation lesion geometries at 20W and 50W for untreated, H$_2$O, 0.9% normal saline, iron, titanium, and copper.

<table>
<thead>
<tr>
<th></th>
<th>Infusate Injection</th>
<th>20W Lesion</th>
<th>50W Lesion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated</td>
<td><img src="image" alt="Untreated Infusate Injection" /></td>
<td><img src="image" alt="Untreated 20W Lesion" /></td>
<td><img src="image" alt="Untreated 50W Lesion" /></td>
</tr>
<tr>
<td>H$_2$O</td>
<td><img src="image" alt="H$_2$O Infusate Injection" /></td>
<td><img src="image" alt="H$_2$O 20W Lesion" /></td>
<td><img src="image" alt="H$_2$O 50W Lesion" /></td>
</tr>
<tr>
<td>0.9% Saline</td>
<td><img src="image" alt="0.9% Saline Infusate Injection" /></td>
<td><img src="image" alt="0.9% Saline 20W Lesion" /></td>
<td><img src="image" alt="0.9% Saline 50W Lesion" /></td>
</tr>
<tr>
<td></td>
<td>Iron</td>
<td>Titanium</td>
<td>Copper</td>
</tr>
<tr>
<td>---</td>
<td>------------------------------</td>
<td>-------------------------------</td>
<td>--------------------------------</td>
</tr>
<tr>
<td></td>
<td><img src="#" alt="Image" /></td>
<td><img src="#" alt="Image" /></td>
<td><img src="#" alt="Image" /></td>
</tr>
<tr>
<td></td>
<td><img src="#" alt="Image" /></td>
<td><img src="#" alt="Image" /></td>
<td><img src="#" alt="Image" /></td>
</tr>
<tr>
<td></td>
<td><img src="#" alt="Image" /></td>
<td><img src="#" alt="Image" /></td>
<td><img src="#" alt="Image" /></td>
</tr>
</tbody>
</table>

- Iron:
- Titanium:
- Copper:
Supplemental Video: Liposomal iron oxide nanoparticles are being directed to an area of interest using magnet guidance.