Iron-Sensitive Cardiac Magnetic Resonance Imaging for Prediction of Ventricular Arrhythmia Risk in Patients With Chronic Myocardial Infarction 
Early Evidence

Ivan Cokic, MD*; Avinash Kali, MS*; Hsin-Jung Yang,* MS; Raymond Yee, MD; Richard Tang, MD; Mourad Tighiouart, PhD; Xunzhang Wang, MD; Warren S. Jackman, MD; Sumeet S. Chugh, MD; James A. White, MD†; Rohan Dharmakumar, PhD†

**Background**—Recent canines studies have shown that iron deposition within chronic myocardial infarction (CMI) influences the electric behavior of the heart. To date, the link between the iron deposition and malignant ventricular arrhythmias in humans with CMI is unknown.

**Methods and Results**—Patients with CMI (n=94) who underwent late-gadolinium-enhanced cardiac magnetic resonance imaging before implantable cardioverter-defibrillator implantation for primary and secondary preventions were retrospectively analyzed. The predictive values of hypointense cores (HIC) in balanced steady-state free precession images and conventional cardiac magnetic resonance imaging and ECG malignant ventricular arrhythmia parameters for the prediction of primary combined outcome (appropriate implantable cardioverter-defibrillator therapy, survived cardiac arrest, or sudden cardiac death) were studied. The use of HIC within CMI on balanced steady-state free precession as a marker of iron deposition was validated in a canine MI model (n=18). Nineteen patients met the study criteria with events occurring at a median of 249 (interquartile range of 540) days after implantable cardioverter-defibrillator placement. Of the 19 patients meeting the primary end point, 18 were classified as HIC+, whereas only 1 was HIC−. Among the cohort in whom the primary end point was not met, there were 28 HIC+ and 47 HIC− patients. Receiver operating characteristic curve analysis demonstrated an additive predictive value of HIC for malignant ventricular arrhythmias with an increased area under the curve of 0.87 when added to left ventricular ejection fraction (left ventricular ejection fraction alone, 0.68). Both cardiac magnetic resonance imaging and histological validation studies performed in canines demonstrated that HIC regions in balanced steady-state free precession images within CMI likely result from iron depositions.

**Conclusions**—Hypointense cores within CMI on balanced steady-state free precession cardiac magnetic resonance imaging can be used as a marker of iron deposition and yields incremental information toward improved prediction of malignant ventricular arrhythmias. (Circ Cardiovasc Imaging. 2015;8:e003642. DOI: 10.1161/CIRCIMAGING.115.003642.)

**Key Words:** arrhythmias, cardiac ■ iron ■ myocardial infarction ■ tachycardia, ventricular ■ ventricular fibrillation

Received January 26, 2015; accepted July 21, 2015.
From the Department of Biomedical Sciences, Biomedical Imaging Research Institute (I.C., A.K., H.-J.Y., R.T., R.D.), Biostatistics and Bioinformatics Research Center (M.T.), and Cedars-Sinai Heart Institute (X.W., S.S.C., R.D.), Cedars-Sinai Medical Center, Los Angeles, CA; Department of Biomedical Engineering, University of California, Los Angeles (A.K., H.-J.Y., H.-J.Y.); Department of Medicine, London Health Sciences Centre, London, Ontario, Canada (R.Y.); Heart Rhythm Institute, University of Oklahoma, Oklahoma City (W.S.J.); Department of Medicine, David Geffen School of Medicine, University of California, Los Angeles (S.S.C., R.D.); and Stephenson Cardiac Imaging Centre, Department of Cardiac Sciences, University of Calgary, Calgary, Alberta, Canada (J.A.W.).

*Dr Cokic, A. Kali, and H.-J. Yang contributed equally to this work.
†Drs White and Dharmakumar are co-senior authors.

Correspondence to Rohan Dharmakumar, PhD, Department of Biomedical Sciences, Cedars-Sinai Medical Center Biomedical Imaging Research Institute, 8700 Beverly Blvd, Suite G-140D Los Angeles, CA 90048. E-mail rohandkumar@csmc.edu

© 2015 American Heart Association, Inc.

Circ Cardiovasc Imaging is available at http://circimaging.ahajournals.org
DOI: 10.1161/CIRCIMAGING.115.003642
However, only 20% to 30% of these patients have been shown to benefit from this therapy; the remaining 70% to 80% not experiencing mVA. Moreover, of those experiencing SCA, at least 65% were not eligible for primary prevention ICD because they did not have a conventional risk predictor (LVEF≤55%). Hence, there is a critical need to develop and validate novel approaches for ventricular arrhythmia risk prediction incremental to the assessment of left ventricular function. To date, several risk predictors have been explored, including, infarct size, type of infarction, infarct transmurality, gray-zone (GZ)/border-zone signal, surviving bundles, and surface ECG parameters. Although these have demonstrated value for the prediction of mVA over conventional LVEF-based stratification, it is also apparent that arrhythmia substrate is complex and is not adequately characterized by the markers explored to date.

There is growing evidence in the literature that postinfarction iron deposition in the myocardium may be arrhythmogenic. Even though it has been long known that abnormal accumulation of iron in the heart has been associated with cardiac conduction abnormalities,20-23 only recently has its relevance come to light in the post-MI setting. Eitel et al24 showed that patients with hemorrhagic infarction have an increased risk for experiencing major adverse cardiac events, including death. Mather et al25 showed that patients with hemorrhagic infarction have prolonged filtered QRS duration, which they associated with increased late-arrhythmogenic risk. Recent canine studies have also confirmed that hemorrhagic infarctions can lead to chronic iron deposition26 and that postinfarction iron can preferentially alter electrophysiology indices within the infarcted heart.27 Moreover, forensic studies using clinical cardiac magnetic resonance imaging (CMR) have shown that SCD victims with chronic MI consistently have hypointense zones in the scarred myocardium in T₂-weighted CMR, which is consistent with iron accumulation. To date, however, there has been no direct evidence for an association between postinfarction iron accumulation and mVAs.

We hypothesized that hypointense cores (HIC) can be identified within CMI using balanced steady-state free precession (bSSFP) CMR at 3.0T, and when used in conjunction with LVEF can markedly improve the prediction of SCA or appropriate ICD therapy. We examined our hypothesis through a retrospective analysis of a CMI patient population undergoing CMR before ICD implantation. Guided by previous theoretical and experimental observations,30,31 we used hypointense territories within CMI on bSSFP CMR at 3.0T as a marker of iron deposition and validated our findings in a canine model of CMI. Specifically, we examined the incremental prognostic value of HIC in bSSFP images, LVEF, scar size, QTc, and GZ on SCD risk using univariable and multivariable analyses.

Methods

Patient Studies

Patient Population

This retrospective cohort study was conducted at a large, tertiary care referral center (London Health Sciences Center, London, Ontario, Canada), between October 2008 and May 2012. The study population consisted of only those patients (n=94) in whom CMR protocol was prescribed and were subsequently instrumented with ICD. CMR imaging was performed in all consenting patients, with the LVEF provided to assist in clinical decision making. Inclusion criteria for the ICD placement were LVEF≤55% estimated by echocardiography, history of prior MI, and appropriate heart failure therapy for ≥3 months. Patients with standard contraindications to late-gadolinium-enhanced (LGE)-CMR, inclusive of estimated glomerular filtration rate ≤30 mL/min per 1.73 m², were excluded. Both verbal and written informed consents were obtained from patients before being studied according to the protocols approved by the Health Sciences Research Ethics Board at the University of Western Ontario. The obtained Research Ethics Board approval also allowed for retrospective analysis of the patient data.

CMR Protocol

Before ICD implantation, all patients underwent CMR in a 3.0T MRI system (MAGNETOM Trio; Siemens Healthcare, Erlangen, Germany) equipped with a cardiac surface receive coil. Cine steady-state free precession images (slice thickness=6 mm, no slice gap, time to echo [TE]=3.0/1.5 ms, flip angle=40°, in-plane resolution=1.8×1.8 mm² to 2.0×2.0 mm², bandwidth [BW]=930 Hz/pixel, and temporal resolution 28–38 ms) were acquired in sequential short-axis views covering the entire LV. LGE imaging was performed 10 to 15 minutes after infusion of gadolinium contrast (0.15–0.2 mmol/kg [IV]; Gadovist, Bayer Inc, Toronto, Canada) with segmented IR-FLASH (inversion-recovery-prepared fast long angle shot) sequence matched to the cine imaging slices triggered at mid diastole (slice thickness=10 mm, no slice gap, repetition time/TE=2.8 ms/1.2 ms, flip angle=20°, in-plane resolution=1.5×1.5 mm² to 1.8×1.8 mm², BW=1500 Hz/pixel). The inversion time was selected empirically by step-wise increase for nulling the remote myocardium.

Image Analysis

All quantitative image analyses were performed using the cmr42 (Circle Cardiovascular Imaging Inc, Calgary, Canada). All qualitative image analysis, including contouring the myocardium, was performed in consensus by 2 blinded reviewers with >3 years experience in reading CMR images on Osirix (version 4.1.2; Pixmeo, Geneva, Switzerland). Remote myocardium was defined as the region showing no hyperintensity on LGE images. A reference region of interest (ROI) was drawn in remote myocardium on LGE images. Infarcted myocardium was defined using the mean±3SD relative to the reference ROI. Infarct GZ was defined as areas with signal intensity between mean±2SD and mean±3SD cutoffs relative to the reference ROI. LV mass, end-diastolic volume, end-systolic volume, and EF were calculated from the cine images and normalized to the body surface area. LV mass was computed from cine images triggered at the same time as LGE images. Infarct volume (%LV) was calculated by normalizing total infarct volume by the total LV myocardial volume.

The presence of infarcted myocardial regions with HICs in bSSFP images (HIC+) was identified qualitatively using a binary scoring scheme (present or absent). All of the HIC determinations were blinded to the clinical outcomes. Additional segmental analysis was performed (also in consensus) to determine the fraction of myocardial segments that were HIC+ relative to the scarred segments and its association with ICD events. The image analysis for the identification of patients with HIC+ involved (1) displaying the cine bSSFP and LGE images side-by-side in bipanel views in Osirix, (2) identifying the infarct territory on LGE image and visually mapping it to bSSFP images, and (3) manually windowing when necessary to discriminate the presence of HIC within the LGE-positive territory on bSSFP images. HIC+ subjects were identified as those with ≥28 contiguous neighboring hypointense pixels in the infarct territory that are present in every frame of a given short-axis set of cine bSSFP images. All other subjects that did not have hypointense regions or those that did not conform to the identification criterion (ie, those resembling India-ink artifacts, bSSFP banding artifacts, and hyperintense regions enclosed by finite hypointense lines) were identified as HIC−. The 8-pixel approach adapted here is similar to that used in previous publications to visually guide the detection of a desired image contrast.
ICD Implantation and Clinical Follow-Up

ICDs were implanted by the local clinical electrophysiology service in a standard fashion within a median of 29 (interquartile range, 69) days of the date of CMR. ICD devices were programmed to detect ventricular fibrillation if 18 of 24 R-R intervals were ≤240 ms. Fast ventricular tachycardia (VT) was defined as 18 of 24 consecutive R-R intervals ≤320 ms and slow VT as 16 consecutive R-R intervals ≤400 ms. In patients receiving primary prevention, ICDs were routinely programmed to deliver antitachycardia pacing or shock therapy for ventricular fibrillation or fast VT and to monitor (not deliver therapy) for slow VT. For patients receiving secondary prevention, ICD programming was altered to treat slow VT using both antitachycardia pacing and shock therapies.

Clinical follow-up and device interrogations at 1, 3, 6, and every 6 months thereafter were initiated from the time of CMR imaging. The primary composite end point was defined as the occurrence of appropriate ICD therapy, survived cardiac arrest, or SCD. In patients having ≥2 registered clinical events during follow-up, the time to first clinical event was used for analysis of event-free survival.

At study closure, all device interrogations were deidentified and adjudicated by 2 local electrophysiologists blinded to CMR image analysis. ICD therapy was classified as appropriate when antitachycardia pacing or shock for fast VT (R-R <320 ms) or ventricular fibrillation was delivered. ICD therapy was defined as inappropriate when delivered for nonarrhythmic conditions (eg, sinus or supraventricular tachycardia), T-wave oversensing, or for any device system malfunction (eg, lead conductor wire fracture).

All study subjects were evaluated for the incidence of SCA or SCD at 12-month intervals and at study closure during a median of 698.5 (interquartile range, 689.5) days. This was performed by telephone interview and a review of all medical records. SCD was defined as death occurring within 1 hour of symptom onset.

Validation of Imaging Findings in Canines

Animal Preparation and CMR Protocol

To ascertain the tissue composition of the MI territories that appear as HIC+, CMR studies were performed in a canine model of chronic reperfused MI. Canines (n=18; 20–25 kg) were enrolled and studied according to the protocols approved by the Institutional Animal Care and Use Committee. Left lateral thoracotomies were performed in canines, and MI was induced by occluding the left anterior descending artery for 3 hours followed by reperfusion. After the surgeries, the canines were allowed to recover for 12 weeks before CMR studies.

CMR Studies in Canines

All canine CMR studies were performed on a clinical 3.0T MRI system (MAGNETOM Verio; Siemens Medical Solutions, Erlangen, Germany). Multiple cardiac-gated, breath-held, 2-dimensional images of contiguous short-axis sections covering the entire LV and the 3 long-axis views (2, 3, and 4 chambers) were acquired using cine bSSFP (repetition time/TE=3.1/1.6 ms, flip angle=40°, 20–25 cardiac phases, BW=930 Hz/pixel), T-weighted imaging (T2* acquired 10–15 minutes after intravenous gadolinium infusion) or LGE (IR-FLASH acquired 10–15 minutes after intravenous gadolinium infusion (Magnevist; Bayer Healthcare Pharmaceuticals Inc, Wayne, NJ), optimal TI to null remote myocardium, repetition time/TE=3.0/1.5 ms, flip angle=25°, and BW=936 Hz/pixel), and LGE imaging (IR-FLASH acquired 10–15 minutes after intravenous gadolinium infusion (Magnevist; Bayer Healthcare Pharmaceuticals Inc, Wayne, NJ), optimal TI to null remote myocardium, repetition time/TE=3.0/1.5 ms, flip angle=25°, and BW=936 Hz/pixel). To minimize off-resonance artifacts, volume-selective shim covering the entire heart was performed before start of data acquisition. Commonly scanned imaging parameters for all the scans were in-plane resolution=1.5×1.5 mm² and slice thickness=8 mm with no slice gap.

Image Analysis

CMR data from canines were analyzed in cmr.41 Infarcted, remote, and GZ myocardial territories were identified on LGE images as previously described for both patients44 and animals.45 Similarly, HIC+ and HIC− short-axis slices were visually identified on bSSFP images as described earlier for patients. First, remote regions identified on LGE images were copied on T1* maps, and T1* of remote myocardium was measured. Next, HIC+ and HIC− territories visually identified on bSSFP images that were positive for LGE were manually traced and copied onto T1* images (matched to slice positions and trigger time) to allow T1* values of both territories to be quantified. T1* values of the remote, HIC+, and HIC− regions were averaged across all animals. Infarct volume and GZ volume were measured from the LGE images using mean+5 SD41 and mean+2 to 3 SD42 criteria, respectively, relative to a reference ROI placed in the remote myocardium. Iron volume was measured from the T1*-weighted image acquired at TE=6.5 ms using the mean–2 SD criterion relative to the reference ROI.46 For a given canine, T1* value of the iron deposition detected within each slice of T1*-weighted image was measured, and whole-heart mean T1* value was obtained by averaging across all the slices. Index of iron burden for each canine was subsequently measured as the ratio of iron volume to the corresponding whole-heart mean T1* value (units of mL/s).

Animal Euthanasia and Histology

Animals were euthanized immediately after the CMR studies, and their hearts were excised. Each heart was manually sliced into 5-mm-thick slices along the LV short-axis, and ex vivo triphenyl tetrazolium chloride staining was performed. The ex vivo slices were carefully matched to the in vivo CMR images based on the location of the papillary muscles and infarct morphology and categorized as HIC+ and HIC−. Three ex vivo slices each from the HIC+ and HIC− territories were embedded in a paraffin block, sliced into 5-µm contiguous sections, and stained with Elastin-modified Masson’s Trichrome (for collagen deposition) and Perl’s stains (for iron deposition) using standard techniques. The sections were mounted on glass slides and scanned at 100-fold magnification using an ACIS II technology-based ChromaVision digital slide scanner (Clartente Inc, Aliso Viejo, CA).

Statistical Analysis

All statistical analyses were performed on R statistical software (version 2.15). Categorical variables are expressed as number (and percentage) of patients. Normality of continuous data was evaluated using Shapiro–Wilks test and quantile–quantile plots. Continuous variables with normal distribution are expressed as mean±SD, whereas those with non-normal distribution are expressed as median with interquartile range. Depending on the normality of the data, ANOVA or Kruskal–Wallis test was performed along with appropriate (Tukey [normal] or Wilcoxon signed-rank test [non-normal]) post hoc analysis to evaluate the differences in LVEF, scar size, GZ size, QRS interval, and QTc duration among the different groups: (1) HIC+/ICD+, (2) HIC+/ICD−, and (3) HIC−/ICD−. Bonferroni corrections were used to adjust the P value for multiple comparisons. Fisher exact test was used to compare groups for categorical variables. In addition, trend analysis (q-test for trend) was performed to observe any trends in the association of infarct size and GZ size with primary outcome. To do this, patients were grouped based on the infarct size and GZ size, and the relative frequency of primary outcome was measured across the different groups. Univariable logistic regression was used to identify the significant predictors of appropriate ICD therapy. Multivariable logistic regression was performed using only the significant univariable predictors. Odds ratios (ORs) were computed for the different predictors from the univariable and multivariable regressions. Receiver operating characteristic analysis was performed to determine the predictive capability of significant univariable and multivariable predictors. Statistical significance was set at P<0.05 for all analyses. Statistical tools used to assess findings in canines were similar to that used to examine patients data. To account for multiple measurements from the same canine, mixed-model ANOVA was used to compare the T1* values among remote myocardium, HIC+ scarred myocardium, and HIC− scarred myocardium. GZ volume measured from the HIC+ and HIC− canines were also compared using Student t test or Wilcoxon signed-rank test depending on the normality of the data. Linear regression analyses were performed to investigate the relationship between GZ volume and iron volume, GZ volume and T1* of iron, and GZ volume and index of iron burden. Statistical significance was set at P<0.05.
Results

Patient Studies

Baseline Characteristics

The present retrospective cohort study enrolled 94 patients with prior MI, where every patient was positive for LGE CMR. Their baseline characteristics are shown in Table 1. Basic risk factors and medical therapies at baseline, except median New York Heart Association functional class, did not differ between patients who did or did not have the HIC present in bSSFP images (Table 1). The baseline CMR values of the study population were as follows: mean LV mass index=93.0±23.2 g/m², LV end-diastolic volume index=118.9±35.3 mL/m², LV end-systolic volume index=83.3±33.6 mL/m³, mean LVEF=31.7±11%, and mean GZ=8.0±3.0%. LGE was observed in all patients included in the study with a mean scar volume (%LV) of 23.3±15.9. There was no difference in LVEF, LV end-diastolic volume, LV end-systolic volume, scar size, and GZ between the 2 study groups.

Clinical Follow-Up

All patients were followed up for occurrence of primary end point for a median of 698.5 (interquartile range, 689.5) days. Device interrogations occurred on months 1, 3, 6, and every 6

Table 1. Baseline Patient Characteristics (Based on the Presence of HIC on bSSFP CMR)

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>All Patients (n=94)</th>
<th>HIC− (n=48)</th>
<th>HIC+ (n=46)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y, mean±SD</td>
<td>62.6±10.5</td>
<td>63.0±11.6</td>
<td>62.2±9.2</td>
<td>0.71</td>
</tr>
<tr>
<td>Male sex (%)</td>
<td>90 (96)</td>
<td>46 (96)</td>
<td>44 (96)</td>
<td>0.94</td>
</tr>
<tr>
<td>Hypertension (%)</td>
<td>62 (66)</td>
<td>33 (69)</td>
<td>29 (63)</td>
<td>0.52</td>
</tr>
<tr>
<td>Diabetes mellitus (%)</td>
<td>38 (40)</td>
<td>21 (44)</td>
<td>17 (37)</td>
<td>0.29</td>
</tr>
<tr>
<td>Hyperlipidemia (%)</td>
<td>79 (84)</td>
<td>41 (85)</td>
<td>38 (82)</td>
<td>0.87</td>
</tr>
<tr>
<td>Smoking (%)</td>
<td>55 (59)</td>
<td>25 (52)</td>
<td>30 (65)</td>
<td>0.22</td>
</tr>
<tr>
<td>History of revascularization (%)</td>
<td>49 (52)</td>
<td>24 (50)</td>
<td>25 (54)</td>
<td>0.67</td>
</tr>
<tr>
<td>NYHA functional class, median (IQR)</td>
<td>2 (1)</td>
<td>3 (1)</td>
<td>2 (1)</td>
<td>0.01*</td>
</tr>
<tr>
<td>Systolic BP, mm Hg</td>
<td>124.1±19.5</td>
<td>122.6±21.3</td>
<td>125.7±17.4</td>
<td>0.49</td>
</tr>
<tr>
<td>Diastolic BP, mm Hg</td>
<td>72.7±11.3</td>
<td>71.5±11.9</td>
<td>73.9±10.7</td>
<td>0.35</td>
</tr>
<tr>
<td>History of ventricular arrhythmias (%)</td>
<td>13 (14)</td>
<td>6 (12)</td>
<td>7 (15)</td>
<td>0.71</td>
</tr>
<tr>
<td>ACE inhibitor (%)</td>
<td>67 (71)</td>
<td>33 (69)</td>
<td>34 (74)</td>
<td>0.92</td>
</tr>
<tr>
<td>Amiodarone (%)</td>
<td>7 (7)</td>
<td>3 (6)</td>
<td>4 (9)</td>
<td>0.73</td>
</tr>
<tr>
<td>ARB (%)</td>
<td>24 (25)</td>
<td>12 (25)</td>
<td>12 (16)</td>
<td>0.30</td>
</tr>
<tr>
<td>ASA (%)</td>
<td>64 (68)</td>
<td>34 (71)</td>
<td>30 (65)</td>
<td>0.81</td>
</tr>
<tr>
<td>β-blocker (%)</td>
<td>75 (80)</td>
<td>37 (77)</td>
<td>38 (83)</td>
<td>0.39</td>
</tr>
<tr>
<td>Digoxin (%)</td>
<td>22 (23)</td>
<td>10 (21)</td>
<td>12 (26)</td>
<td>0.84</td>
</tr>
<tr>
<td>Diuretic (%)</td>
<td>55 (59)</td>
<td>23 (48)</td>
<td>32 (70)</td>
<td>0.89</td>
</tr>
<tr>
<td>Plavix (%)</td>
<td>10 (11)</td>
<td>4 (8)</td>
<td>6 (13)</td>
<td>0.43</td>
</tr>
<tr>
<td>Statin (%)</td>
<td>50 (53)</td>
<td>23 (48)</td>
<td>27 (59)</td>
<td>0.51</td>
</tr>
<tr>
<td>Heart rate, bpm (mean±SD)</td>
<td>70.5±13.7</td>
<td>68.7±11.7</td>
<td>72.5±15.5</td>
<td>0.20</td>
</tr>
<tr>
<td>QRS, ms (mean±SD)</td>
<td>129.8±29.6</td>
<td>131.4±30.9</td>
<td>127.6±28.5</td>
<td>0.53</td>
</tr>
<tr>
<td>QTc, ms (mean±SD)</td>
<td>456.4±39.9</td>
<td>459.1±41.7</td>
<td>453.8±38.9</td>
<td>0.52</td>
</tr>
<tr>
<td>LBBB (%)</td>
<td>32 (34)</td>
<td>14 (27)</td>
<td>18 (39)</td>
<td>0.31</td>
</tr>
<tr>
<td>LV mass index, g/m² (mean±SD)</td>
<td>93.0±23.2</td>
<td>92.6±23.2</td>
<td>93.5±23.7</td>
<td>0.85</td>
</tr>
<tr>
<td>LVEDV index, mL/m² (mean±SD)</td>
<td>118.9±35.3</td>
<td>116.0±33.1</td>
<td>121.0±37.3</td>
<td>0.47</td>
</tr>
<tr>
<td>LVESV index, mL/m² (mean±SD)</td>
<td>83.3±33.6</td>
<td>79.9±28.9</td>
<td>85.9±37.7</td>
<td>0.37</td>
</tr>
<tr>
<td>LVEF, % (mean±SD)</td>
<td>31.7±11</td>
<td>32.3±10.1</td>
<td>31.2±11.8</td>
<td>0.59</td>
</tr>
<tr>
<td>Scar volume, %LV (mean±SD)</td>
<td>23.3±15.9</td>
<td>22.0±16.6</td>
<td>24.2±14.9</td>
<td>0.47</td>
</tr>
<tr>
<td>Gray-zone (2–3 SD), % (mean±SD)</td>
<td>8.0±3.0</td>
<td>7.9±3.1</td>
<td>8.0±2.9</td>
<td>0.88</td>
</tr>
</tbody>
</table>

ACE, angiotensin-converting enzyme; ARB, angiotensin receptor blocker; ASA, acetylsalicylic acid; BP, blood pressure; bSSFP, balanced steady-state free precession; CMR, cardiac magnetic resonance imaging; EDV, end-diastolic volume; EF, ejection fraction; ESV, end-systolic volume; HIC, hypointense cores; IQR, interquartile range; LBBB, left bundle-branch block; LV, left ventricle; and NYHA, New York Heart Association.

*Significant (P<0.05).
months thereafter from the day of CMR imaging. End point criteria (appropriate ICD therapy, SCA, and SCD) were met in 19 patients with events occurring at a median of 249 (interquartile range, 540) days after ICD placement. Of these, 11 patients received an appropriate shock, 3 patients received appropriate antitachycardia pacing for sustained VT (1 of 3 with advanced heart failure), and 5 patients died because of SCD (1 of 5 with advanced heart failure). In addition, 3 patients had non-SCD caused by progressive heart failure and 4 patients with progressive heart failure died of noncardiac cause.

**Hypointense MI Territories on bSSFP Images Versus Primary End Point**

Analysis of the CMR data showed that of the 19 patients having a primary end point 18 were classified as HIC+, whereas only 1 subject was classified as HIC− (Table 2). Figure 1 shows representative CMR findings in a patient with HIC+ experiencing the primary end point and a patient who was HIC− and did not experience the primary end point. In the group of patients in whom the primary end point was not met, there were 28 HIC+ and 47 HIC− patients.

Univariable regression analysis showed that only LVEF (β=−0.06; OR, 0.95; 95% confidence interval, 0.98–1.01; \(P=0.048\)) and the presence of HIC (β=3.41; OR, 30.21; 95% confidence interval, 3.82–238.77; \(P=0.001\)) were significant predictors of primary end point (Table 2). Multivariable regression analysis showed that both LVEF (β=−0.06; OR, 0.95; 95% confidence interval, 0.89–1.004; \(P=0.067\)) and the presence of HIC (β=3.50; OR, 33.29; 95% confidence interval, 4.16–265.6; \(P=0.001\)) were independent predictors of primary end point (Table 2). Receiver operating characteristic analysis showed that the addition of scar size to LVEF did not change the AUC, but the addition of HIC to LVEF and scar size increased the AUC from 0.68 to 0.87, demonstrating an additive prognostic value of HIC (Figure 2).

Univariable regression analysis showed that only LVEF (β=−0.06; OR, 0.95; 95% confidence interval, 0.98–1.01; \(P=0.048\)) and the presence of HIC (β=3.41; OR, 30.21; 95% confidence interval, 3.82–238.77; \(P=0.001\)) were significant predictors of primary end point (Table 2). Multivariable regression analysis showed that both LVEF (β=−0.06; OR, 0.95; 95% confidence interval, 0.89–1.004; \(P=0.067\)) and the presence of HIC (β=3.50; OR, 33.29; 95% confidence interval, 4.16–265.6; \(P=0.001\)) were independent predictors of primary end point (Table 2). Receiver operating characteristic analysis showed that the addition of scar size to LVEF did not change the AUC, but the addition of HIC to LVEF and scar size increased the AUC from 0.68 to 0.87, demonstrating an additive prognostic value of HIC (Figure 2).

Trend analysis showed a tendency for increasing frequency of primary outcomes with the increase in infarct size (Figure 3A) and size of GZ (Figure 3B), which is consistent with previous observations by others.\(^{32,35,43,44}\) Although there were no significant differences in the relative frequencies of primary outcomes among patient groups based on the infarct size (\(P=0.28\)), the relative frequencies of primary outcomes were significantly different among patient groups based on the GZ size (\(P<0.001\)).

**Canine Studies**

**CMR Findings**

Of 18 animals studied, all animals demonstrated visual LGE evidence for myocardial infarction and 9 were scored as HIC+. The relationship between the presence of HIC and primary outcomes was also analyzed. Table 2 shows the univariable and multivariable odds ratios for primary outcomes.

### Table 2. HIC vs Frequency of Primary Outcome

<table>
<thead>
<tr>
<th>Variable</th>
<th>With Primary Outcome (n=19)</th>
<th>Without Primary Outcome (n=75)</th>
<th>Univariable Odds Ratio (95% CI)</th>
<th>Multivariable Odds Ratio (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIC</td>
<td>HIC+ (n=18)</td>
<td>HIC− (n=1)</td>
<td>30.21 (3.82–238.77; (P=0.001^))</td>
<td>33.29 (4.16–265.6; (P=0.001^))</td>
</tr>
<tr>
<td>LVEF, %, mean±SD, range (min, max)</td>
<td>27.4±10.8 (15.1, 49.3)</td>
<td>21.3</td>
<td>0.95 (0.98–1.01; (P=0.048))</td>
<td>0.95 (0.89–1.004; (P=0.067))</td>
</tr>
<tr>
<td>Scar volume (%LV), mean±SD, range (min, max)</td>
<td>25.7±14.8 (1.9, 53.9)</td>
<td>22.0</td>
<td>1.01 (0.89–1.001; (P=0.728))</td>
<td>...</td>
</tr>
<tr>
<td>GZ (2–3 SD), %, mean±SD, range (min, max)</td>
<td>7.9±2.8 (2.6, 11.1)</td>
<td>12.1</td>
<td>1.03 (0.87–1.21; (P=0.74))</td>
<td>...</td>
</tr>
<tr>
<td>QRS, ms, mean±SD</td>
<td>126.9±28.7</td>
<td>156.0</td>
<td>1.00 (0.98–1.01; (P=0.695))</td>
<td>...</td>
</tr>
<tr>
<td>QTc, ms, mean±SD</td>
<td>455.1±33.9</td>
<td>422.0</td>
<td>1.00 (0.95–1.21; (P=0.790))</td>
<td>...</td>
</tr>
</tbody>
</table>

CI indicates confidence interval; GZ, gray-zone; HIC+, hypointense core present; HIC−, hypointense core absent; LVEF, left ventricular ejection fraction; and OR, odds ratio.

*Significant (\(P<0.05\)).

**Figure 1.** Presence of hypointense core (HIC) within myocardial infarction (MI) territories on balanced steady-state free precession (bSSFP) CMR. Representative bSSFP and late-gadolinium-enhancement (LGE) images from 2 patients receiving implantable cardioverter-defibrillator therapy; 1 who met the primary end point (Event+) and 1 who did not meet the primary end point (Event−). For the Event+ patient, blue arrows denote the MI region on LGE imaging and the HIC region on bSSFP imaging. In the Event− patient, no HIC was observed by bSSFP within the MI region, indicated by the red arrow on LGE imaging.
and the remainder were scored as HIC−. For each animal, 10 to 14 spatially matched cine, $T_2^*$ and LGE image slices were available for analysis. A total of 94 slices (of 232) were positive for myocardial infarction on the basis of LGE imaging. Of these, 32 were classified as HIC+ and 62 as HIC− on bSSFP images. $T_2^*$ mapping confirmed the visual presence of distinct hypointense regions on all HIC+ slices, whereas HIC− slices failed to show this finding.

Representative LGE, bSSFP, and $T_2^*$-weighted images acquired 12 weeks post MI are shown from HIC+ and HIC− canines in Figures 4A, 4B, and 4C and 5A, 5B, and 5C, respectively. Figure 4A and 4B together show visually evident HIC+ regions within the region of MI. Spatial concordance of HIC+ in bSSFP images (Figure 4B) with $T_2^*$-weighted images (Figure 4C), a validated approach for detecting iron deposits, was visually evident, suggesting that the HIC+ regions in bSSFP images are likely from iron. In contrast, note that in Figure 5, although an MI is evident (Figure 5A), there are no hypointense regions within the corresponding MI territories in bSSFP (Figure 5B) and $T_2^*$ (Figure 5C) images.

Individual and mean cohort $T_2^*$ values for remote, HIC+, and HIC− tissue are shown in Figure 6. The mean $T_2^*$ of HIC+ territories (21±5 ms) were significantly lower than those of remote myocardium (32±3 ms; $P<0.001$) and HIC− territories (30±4 ms; $P<0.001$). The mild difference in $T_2^*$ ($≈2$ ms) observed between remote tissue and HIC− territories was found to be statistically significant ($P=0.04$).

Mean GZ volume measured from HIC+ canines was significantly higher than that of the HIC− canines (0.94±0.58 versus 0.49±0.14; $P=0.02$). Significant linear relationships were found between GZ volume and iron volume ($R^2=0.41$; $P<0.001$; Figure 7A), GZ volume and $T_2^*$ of iron ($R^2=0.18$; $P<0.001$; Figure 7B), and GZ volume and index of iron burden ($R^2=0.39$; $P<0.001$; Figure 7C).

**Histological Findings**

Ex vivo triphenyl tetrazolium chloride staining confirmed the presence of myocardial infarction in both HIC+ (Figure 4D) and HIC− (Figure 5D) canines. A brown discoloration consistent with chronic iron deposition was observed within the core of MI territories among HIC+ canines, but not among HIC− canines. EMT staining showed extensive collagen deposition within the MI territories in both HIC+ (Figure 4E) and HIC− slices (Figure 5E). Perl staining confirmed that significant chronic iron deposition was only present within the infarcted territories of HIC+ slices (Figure 4F), but not of HIC− slices (Figure 5F). EMT and Perl staining of remote myocardium showed no abnormal pathologies in either canine group (Figures 4G and 4H and 5G and 5H).

**Discussion**

SCA, typically culminating from mVAs, is the leading cause of death in the United States. More than 40% of patients dying of SCA have CMIs. Although the ICD is a highly effective tool for the primary prevention of SCA, current guideline recommendations (LVEF<35%) inadequately identify those patients at greatest risk, resulting in an undesirably high number needed to treat to save 1 life.12,13,15–19 Accordingly, improved methods for the prediction of mVAs are in critical need.

Multiple studies examining mechanisms of arrhythmogenesis in CMI over the past decade have identified fibrosis as the primary structural substrate associated with mVA.45 It is thought that the presence of collagen fibers acts as a barrier against electrophysiological propagation, facilitating a re-entry circuit. However, not all patients with CMI develop mVA, despite the presence of intramyocardial collagen. Although scar size has emerged as an important parameter associated with mVA, the exact association of scar size and mVA remains...
unclear. Klem et al.\(^3\) showed that although there is a sharp increase in SCD incidence (from 10% to 45%) as scar volume increases from 0% to 5% to 10%, the incidence does not appear to be influenced by scar volume beyond this range. In contrast, Kwon et al.\(^4\) showed that scar size >30% is a key risk factor for events, and Yan et al.\(^3\) showed that every 10% increase in scar size increases the risk of developing SCD by 50%. These observations suggest that other factors in addition to scar size, such as tissue composition, may play an important role. To date, however, only a few studies have examined tissue composition of CMI beyond that of fibrosis.

Several mechanisms by which heart function may decline and predispose patients to mVA in acute and chronic post-MI settings have been investigated. Among these, it has long been suspected that iron composites within the acute MI region are proarrhythmic and have the potential to promote early adverse remodeling and mVAs. This notion inspired the use of chelation therapies to decrease oxidative stress from iron within acute MI territories with the goal of decreasing acute injury and marginalizing electric anomalies\(^6\); however, results have been mixed. Notably, all studies to date examining this therapy were confined to the acute setting (ie, not >1–2 days after an event) or did not randomize subjects (animals or patients) to chelation therapy on the basis of imaging evidence for iron deposition. Indeed, until recently, the presence of iron, its influence on adverse remodeling, and any electric implications during the chronic phase of MI (ie, weeks to months after an event) had not been explored.\(^2\)\(^{6,7}\)

CMR has been instrumental in demonstrating that intramyocardial hemorrhage is frequently present in reperfused acute MI. However, until recently, the fate of extravasated red blood cells within the infarct territory was not known. Recent CMR studies have shown that hemorrhagic infarction can lead to chronic iron deposition with a characteristic subendocardial location identified using \(T_2^*\) CMR. In addition to...
showing its actual presence in the chronic setting, it was also shown that such infarcts with iron deposition are disposed to worse remodeling, are subjected to prolonged iron-driven inflammation, and exhibit altered electrophysiological parameters (QTc, presence of isolated late potentials, etc.) compared with infarcts without iron deposition.26,27 Most notably, in situ forensic pathology studies have consistently noted, visually evident, hypointense T2-weighted CMR signals, consistent with iron deposition among SCA victims with CMI.28,29 These findings suggest that hemorrhagic acute MI may have a significant influence on the development of mVA in the chronic setting.

We performed a retrospective observational study of CMI patients undergoing ICD therapy to assess the incremental predictive value of HIC, a marker of iron deposition using an iron-sensitive CMR sequence (bSSFP), over previously explored indices (LVEF, scar volume, GZ, QTc, and QRS). In univariable analysis using logistic regression, we found only LVEF and HIC to be significant predictors, whereas all other parameters did not reach statistical significance. Using multivariable logistic regression analysis, only LVEF and HIC reached statistical significance. In particular, receiver operating characteristic analysis showed that when HIC was added to LVEF, there was a marked increase in the area under the curve compared with LVEF alone, demonstrating improved predictive accuracy. Moreover, using an established canine model of chronic reperfused MI, we confirmed theoretical predictions that iron deposition will appear hypointense on bSSFP-based imaging and that these HIC+ regions have a marked decrease in T2* by quantitative mapping (a CMR approach validated for myocardial iron deposition). In addition, our histological observations confirmed that HIC+ regions within MI territories are indeed related to local iron deposition.

In this study, we also examined the predictive utility of previously explored indices on the primary outcome. Among these, only LVEF was predictive. Total scar volume, expressed as a continuous variable, showed a trend toward greater volume in those with events, but this was not significant. However, the incidence of primary end point was ≈7-fold higher in patients with a scar volume >10% than in those with scar volume <5%. This is consistent with the previous observations that the risk of developing mVA seems to be low in patients with small infarcts, and significantly elevated among those with large infarcts. On the basis of our canine studies, we speculate that the source of HIC+ in our patient population was because of iron deposition related to a prior hemorrhagic acute MI. It is reasonable to expect that HIC+ infarcts are less likely to occur in the setting of small infarcts and may provide a rational explanation for an observed threshold phenomenon relating infarct size to future arrhythmic risk.

Although we showed that the hypointense regions in CMI are likely related to iron deposition, several other sources can mimic hypointensities on bSSFP imaging of the myocardium. First, the well-known banding artifacts can be a source of these artifacts. However, such bands tend to be continuous across adjacent organs and are also known to impart flow artifacts. When such banding artifacts were identified to be present in this study, data from those imaging slices were not included in our analysis. Another source of hypointensity in bSSFP images is motion artifact, which is typically observed only in imaging frames when the cardiac motion is severe. Because our analysis conditioned that the hypointense regions be present throughout the cardiac cycle (ie, in all frames), we were able to limit motion artifacts from being incorrectly identified as HIC+. Third, although it is possible that calcification may also contribute to hypointensity within the infarct core, iron deposits are more readily visible as hypointense regions because the magnetic susceptibility shift arising from calcium deposits is at least 2 orders of magnitude weaker than with iron.47 However, the evidence...
in the literature for arrhythmogenic potential of calcium deposits within MI seems negative. Finally, the presence of fat may also appear as hypointense lines (India-ink artifact) or hypointense lines enclosing hyperintense territories in bSSFP images. On the basis of our definition that HIC+ regions need to have at least 8 contiguous nearest-neighbor pixels, we believe that we were able to minimize fatty infarcts from being counted as HIC+. These efforts enabled us to ensure that potential confounders of HIC+ were kept to a minimum, but further studies are needed to fully elucidate the substrate in patients.

**Limitations**

This study has several limitations. First, we conducted a retrospective observational study from a single-center with a limited sample size. Thus, the findings from this study need to be confirmed in a larger, multicenter setting with adequate power to examine all relevant indices on the primary outcome. Second, we examined the presence of HIC using bSSFP imaging in a clinical cohort and performed ex vivo validation in a canine model of chronic reperfusion MI. The latter was used to demonstrate that it is iron deposition being identified using HIC scoring criteria. The inherent limitation of this design is that validation is based on an independent, albeit well-established animal model of chronic MI. Histological confirmation of iron deposition was not available in the patient cohort because of its retrospective study design and lack of postmortem tissue for iron-sensitive staining. Furthermore, although every effort was made to remove potential confounders in the image analysis, the scoring of HICs in clinical cohorts may be improved using $T_2$ mapping sequences. Given the retrospective nature of this study, $T_2$ acquisitions were not performed as part of the CMR examination and were therefore not available for analysis. However, the ability to categorize HIC using routine bSSFP imaging offers greater generalizability to routine clinical practice. Third, we recruited only MI patients who were considered eligible for ICD therapy (ie, LVEF<35%). Of great interest is the prognostic utility of this technique among those currently not meeting eligibility criteria for ICD therapy. Accordingly, further studies are needed to examine the relationship between post MI iron and mVA in patients with LVEF>35%. Fourth, the scan parameters used for LGE and bSSFP imaging in patients differed slightly from those used in canines because of practical limitations (eg, heart rate variations, imaging field of view, size of hearts, etc.). Nonetheless, because LGE and bSSFP acquisition parameters between patient and canines studies were relatively similar, we do not expect to see any differences in scar size or marked differences in HIC visualization that would confound the interpretations of the study. Fifth, inter- and intraobserver variability were not evaluated for detecting HICs on bSSFP images because the images were evaluated in consensus by 2 blinded reviewers similar to other early investigations. Furthermore, this study was not designed to assess the long-term changes in imaging biomarkers because CMR was confined to a single time point before ICD implantation. It would be of interest for the design of future studies to consider incorporating multiple imaging time points to assess the temporal changes in the imaging biomarkers of interest. Finally, we recognize that our findings are associative and do not provide a causal relationship between iron deposition and arrhythmia formation. Given that inflammation has been associated with frequent cause of life-threatening ventricular arrhythmias and SCD, we conjecture that a possible mechanism by which iron deposits could influence the arrhythmogenesis is via prolonged iron-driven inflammation in the chronic phase of myocardial infarction. Additional studies are required to elucidate the proarrhythmic effects of cytokines and inflammatory mediators within iron-laden chronic infarctions.

**Conclusions**

This is a hypothesis-generating study, which provides sentinel evidence that HICs on iron-sensitive CMR may be a powerful predictor of mVA in patients with CMI. Further prospective studies sufficiently powered to evaluate the incremental utility of HIC versus contemporary risk prediction markers are warranted.

**Sources of Funding**

This research was supported in part by grant NA6488 from the Heart and Stroke Foundation, the Canada Foundation of Innovation Leaders Opportunity Fund, and the Ontario Research Fund. Imaging in Cardiovascular Therapeutics grant to Dr White and in part by NIH R01 HL091989 to Dr Dharamkumar.

**Disclosures**

None.

**References**


10 Cokic et al Iron-Sensitive CMR for Risk of VT/VF


CLINICAL PERSPECTIVE

Sudden cardiac arrest, typically culminating from malignant ventricular arrhythmias (mVA), is the leading cause of death in the United States. It is estimated that ≥50% of the sudden cardiac arrest victims have a chronic myocardial infarction. Currently, the substrate mediating the development of mVAs in patients with chronic myocardial infarction is unclear, limiting the accuracy in identifying the best candidates for life-saving prophylactic implantable cardioverter-defibrillator (ICD) therapy. Current guidelines recommend implantable cardioverter-defibrillators for patients with severely decreased left ventricular ejection fraction (left ventricular ejection fraction, <35%); however, only 20% to 30% of these patients benefit from it; the remaining 70% to 80% of this patient group never develop mVAs. To date, many risk predictors have been explored, including, infarct size, type of infarction, infarct transmurality, border zones, surviving bundles, and surface ECG parameters. Although these have demonstrated value for the prediction of mVA over conventional left ventricular ejection fraction–based stratification, it is also apparent that arrhythmia substrate is complex and is not adequately characterized by the markers explored to date. In this study, we provide sentinel evidence that hypointense cores on iron-sensitive cardiac magnetic resonance images may be used to markedly increase the power in predicting mVA based on left ventricular ejection fraction alone in patients with chronic myocardial infarction.
Iron-Sensitive Cardiac Magnetic Resonance Imaging for Prediction of Ventricular Arrhythmia Risk in Patients With Chronic Myocardial Infarction: Early Evidence
Ivan Cokic, Avinash Kali, Hsin-Jung Yang, Raymond Yee, Richard Tang, Mourad Tighiouart, Xunzhang Wang, Warren S. Jackman, Sumeet S. Chugh, James A. White and Rohan Dharmakumar

_Circ Cardiovasc Imaging_. 2015;8:
doi: 10.1161/CIRCIMAGING.115.003642
_Circulation: Cardiovascular Imaging_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2015 American Heart Association, Inc. All rights reserved.
Print ISSN: 1941-9651. Online ISSN: 1942-0080

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circimaging.ahajournals.org/content/8/8/e003642

**Permissions:** Requests for permissions to reproduce figures, tables, or portions of articles originally published in *Circulation: Cardiovascular Imaging* 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: Cardiovascular Imaging* is online at: http://circimaging.ahajournals.org/subscriptions/