Assessment of Myocardial Viability by Intracellular $^{23}$Na Magnetic Resonance Imaging

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**Background**—Because of rapid changes in myocardial intracellular Na\(^+\) (Na\(^+\)) during ischemia and reperfusion (R), $^{23}$Na magnetic resonance imaging (MRI) appears to be an ideal diagnostic modality for early detection of myocardial ischemia and viability. So far, cardiac $^{23}$Na MRI data are limited and mostly concerned with imaging of total Na\(^+\). For proper interpretation, imaging of both Na\(^+\)_i and extracellular Na\(^+\) is essential. In this study, we tested whether Na\(^+\)_i imaging can be used to assess viability after low-flow (LF) ischemia.

**Methods and Results**—Isolated rat hearts were subjected to LF (1%, 2%, or 3% of control coronary flow) and R. A shift reagent was used to separate Na\(^+\)_i and extracellular Na\(^+\) resonances. Acquisition-weighted $^{23}$Na chemical shift imaging (CSI) was alternated with $^{23}$Na MR spectroscopy. Already during control perfusion, Na\(^+\)_i could be clearly seen on the images. Na\(^+\)_i image intensity increased with increasing severity of ischemia. During R, Na\(^+\)_i image intensity remained highest in 1% LF hearts. Not only did we find very good correlations between Na\(^+\)_i image intensity at end-R and end-diastolic pressure ($R=0.85$, $P<0.001$) and recovery of the rate-pressure product ($R=−0.88$, $P<0.001$) at end-R, but most interestingly, also Na\(^+\)_i image intensity at end-LF was well correlated with end-diastolic pressure ($R=0.78$, $P<0.01$) and with recovery of the rate-pressure product ($R=−0.81$, $P<0.01$) at end-R. Furthermore, Na\(^+\)_i image intensity at end-LF was well correlated with creatine kinase release during R ($R=0.79$, $P<0.05$) as well as with infarct size ($R=0.77$, $P<0.05$).

**Conclusions**—These data indicate that $^{23}$Na CSI is a promising tool for the assessment of myocardial viability. (Circulation. 2004;110:3457-3464.)

**Key Words:** magnetic resonance imaging, myocardium, ischemia, sodium, reperfusion

Assessment of myocardial viability in patients with chronic coronary artery disease or acute and subacute myocardial infarction is clinically important for distinguishing stunned or hibernating myocardium from irreversibly injured myocardium. Patients may benefit from revascularization when viable tissue is present in the dysfunctional area of the myocardium.

Several clinical imaging modalities exist for assessment of viable myocardium, eg, $^{201}$Tl scintigraphy, positron emission tomography, stress echocardiography, and delayed contrast-enhanced magnetic resonance imaging (DCE-MRI), which have proven useful in the chronic situation. In the subacute situation, however, a reliable technique for the assessment of myocardial viability does not exist.

Previous research has shown that one of the best ways to determine myocardial viability is to evaluate sarcoplemmal function, in particular, Na\(^+\)_i,K\(^+\)-ATPase function. Intracellular sodium (Na\(^+\)_i) is a sensitive marker of myocardial ischemia and Na\(^+\)_i,K\(^+\)-ATPase function. During the very first minutes of myocardial ischemia, Na\(^+\)_i already starts to rise because of inhibition of the sarcolemmal Na\(^+\)_i,K\(^+\)-ATPase and continued or increased influx through the Na\(^+\)_i channel, via the Na\(^+\)_i-H\(^+\) exchanger, and possibly through other routes. This very early rise in Na\(^+\)_i makes it a very sensitive indicator of ischemia. Recovery of Na\(^+\)_i,K\(^+\)-ATPase function after an ischemic episode is essential for recovery of myocardial function. Reperfusion (R) of myocardium after a brief (20-minute) period of ischemia causes an immediate (within 5 seconds) reactivation of Na\(^+\)_i,K\(^+\)-ATPase function and a decrease in Na\(^+\)_i, which suggests that Na\(^+\)_i is also a very sensitive indicator of myocardial viability.

$^{23}$Na MRI is a noninvasive methodology for measuring in vivo Na\(^+\), but so far, studies using this technique are scarce and mostly concerned with imaging of total Na\(^+\). After acute ischemia and R, increased total Na\(^+\) MRI intensity was shown to be associated with nonviable tissue. However, depending on the severity and duration of the ischemic insult, information on both Na\(^+\)_i and extracellular Na\(^+\) (Na\(^+\)_e) may be crucial. For instance, when coronary flow is entirely absent, the total-Na\(^+\) signal intensity may not change, whereas Na\(^+\)_i

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signal intensity may increase, or, in the case of extracellular edema, Na\(^{+}\), signal intensity may increase because of a larger extracellular space while Na\(^{-}\), signal intensity may be unaltered.

Recently, Weidensteiner et al\(^{10}\) showed that in ischemic myocardium, an increase in [Na\(^{+}\)], can be detected by a 3D chemical-shift imaging (CSI) technique. The time required to measure one 3D dataset was 30 minutes. We have adapted a 2D CSI technique to measure the spatial distribution of Na\(^{+}\), Na\(^{-}\), and total Na\(^{+}\) in <5 minutes, making relevant successive measurements feasible. Preliminary experiments with this technique showed that Na\(^{+}\), imaging is superior to total-Na\(^{+}\) imaging in identifying the affected zone after complete coronary occlusion.\(^{11}\) Increased Na\(^{+}\), signal intensity was already apparent from 0 to 15 minutes of left ascending coronary artery ligation of an isolated heart, whereas total Na\(^{+}\) signal intensity had not changed. Clinically, ischemia may represent a variety of residual flow conditions. Therefore, in the present study, cardiac images of isolated rat hearts were obtained during low-flow (LF) ischemia at various flow rates and subsequent R. The aims of this study were to explore the potential of Na\(^{+}\), imaging for the assessment of myocardial viability and to evaluate the added value of Na\(^{-}\), MRI over total Na\(^{+}\) MRI.

**Methods**

**Preparation of Isolated, Perfused Rat Hearts**

Male Wistar rats (300 to 350 g) were anesthetized with diethylether inhalation and treated with heparin (1000 U/kg IV). The heart was rapidly excised and placed in ice-cold Krebs-Henseleit (KH) buffer. The aorta was cannulated, the left ventricle was vented, and constant 76-mm Hg perfusion was initiated with KH at 37°C. The KH, containing (in mmol/L) 125 NaCl, 4.7 KCl, 1.0 MgCl\(_2\), 25 NaHCO\(_3\), 1.75 CaCl\(_2\), 0.5 EDTA, 10 d-glucose, and 0.5 pyruvate, was filtered before use through a 0.8-μm filter and was equilibrated with 95% O\(_2\) and 5% CO\(_2\), resulting in a pH of 7.4.

A balloon was inserted into the left ventricle and connected to a pressure transducer. Left ventricular end-diastolic pressure (LVEDP) was set to 7 mm Hg. Developed pressure was calculated as the difference between peak systolic pressure and LVEDP. All hearts were paced (5 Hz) with agar-wick electrodes connected to a stimulator. Hearts subjected to regional ischemia were instrumented with an apex cannula instead of an intraventricular balloon. The experimental protocol was in accord with the guidelines of the Committee for Animal Experiments of the University Medical Center, Utrecht, the Netherlands.

**MR Measurements**

Hearts in a 20-mm–diameter nuclear MR (NMR) tube were lowered into a 9.4-T, vertical-bore magnet, which was interfaced with a Bruker AVANCE 400 DRX spectrometer (Bruker Biospin) equipped with a 1000-mT/m microimaging gradient accessory.

**1H MRI of Isolated, Perfused Hearts**

The 20-mm–diameter birdcage coil was tuned to 400.15 MHz. Gated long- and short-axis \(^1\)H 2D gradient-echo images (matrix size, 256×256; field of view, 20×20 mm; slice thickness, 2.5 mm; echo time, 7 ms; repetition time, 200 ms) were acquired at the beginning and the end of the protocol to serve as anatomic references (see Figure 1). The short-axis slice was taken through the middle of the left ventricle according to the long-axis image.

**23Na MR spectroscopy and CSI of Isolated, Perfused Hearts**

After \(^1\)H MRI, the \(^1\)H resonator was replaced by a quadrature-driven \(^23\)Na birdcage resonator, which was tuned to 105.85 MHz while the heart stayed in place. To separate Na\(^{+}\), and Na\(^{-}\), resonances, perfusion was switched to a KH that was modified by removal of EDTA and addition of the sodium salt of the shift reagent, thulium(III)\(1,4,7,10\)-tetraazacyclododecane-\(N\(^\bullet\)\),\(N\(^\bullet\)\),\(N\(^\bullet\)\),\(N\(^\bullet\)\)\(\text{N}\)\(\text{N}\)\(\text{N}\)\(\text{N}^\text{II}\)tetra(methyleneephosphonate) (TmDOTP\(^{-}\), 3.5 mmol/L). Na\(^{+}\) was kept constant at 150 mmol/L by adjusting the amount of NaCl. To correct for Ca\(^{2+}\) binding by the shift reagent, the total Ca\(^{2+}\) added to the perfusate was increased to 3.42 mmol/L, which resulted in a free [Ca\(^{2+}\)] of 0.85 mmol/L as measured by a Ca\(^{2+}\)-sensitive electrode.

\(^23\)Na MR spectra were acquired with a 5-kHz spectral width and 2048 data points. Typically, 128 free induction decays were accumulated after 90° pulses with a repetition time of 250 ms. Free induction decays were Fourier-transformed after gaussian multiplication. After polynomial baseline correction, the Na\(^{+}\), and reference Na\(^{-}\) peaks were integrated. All \(^23\)Na spectra were fully relaxed. Quantification was performed relative to the peak area of the reference resonance. The reference capillary contained a known amount of 250 mmol/L Na\(^{+}\) and 5 mmol/L TmDOTP\(^{-}\). NMR visibility of all \(^23\)Na signals was assumed to be 100%.

The 2D \(^{23}\)Na CSI dataset was acquired in 4 minutes, 18 seconds, with a repetition time of 27.4 ms (matrix size, 16×16; field of view, 20×20 mm; 256 complex data points in the spectral domain; spectral width, 5 kHz). The slice was selected with a 1000-μs sinc pulse and was centered on the axial \(^1\)H slice. The delay before acquisition was kept as short as possible and amounted to 307 μs. We used acquisition weighting according to a sine-ball function to improve the spatial response function and hence, minimize signal contamina-
tion between voxels.\(^{11}\) The number of acquisitions in the middle row of the symmetrical acquisition matrix was as follows: 0, 6, 24, 50, 78, 104, 122, 128, 122, 104, 78, 50, 24, 6, and 0 (total number of acquisitions, 9408). The acquisition weighting also resulted in a higher signal-to-noise ratio in the same amount of time because of more efficient k-space sampling. The nominal resolution was 1.25 mm in the x and y directions and 5 mm in the z direction (voxel size, 7.8 μL). Image resolution was improved by zero-filling in the spatial domain (resulting in a 64×64 matrix) and gaussian filtering in the spectroscopic domain. To calculate the average image pixel intensity of the Na\(^{+}\) standard, the buffer, and myocardial Na\(^{+}\), and Na\(^{-}\), appropriate regions were chosen on the \(^1\)H image and copied onto the respective \(^{23}\)Na images. Images with the highest resolution were used (20-mm–diameter birdcage coil) to perform peak quantification. Then, the same regions of interest were chosen on the \(^23\)Na images and peak areas were determined. 

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**Figure 1.** Schematic representation of protocol. S indicates stabilization; C, control perfusion; and SR, shift reagent. Low-flow perfusion occurred at 1%, 2%, or 3% of control coronary flow. All other abbreviations are as defined in text.
average pixel intensity of each of the different Na\(^+\) resonances were chosen. The intensities were expressed as percentage of the intensity of the buffer.

### Protocol

Hearts were allowed to stabilize, and meanwhile, tuning and matching of the \(^1\)H resonator were optimized, as well as the static magnetic field. After \(^1\)H MRI, the \(^1\)H resonator was replaced by the \(^23\)Na birdcage resonator. The perfusate was switched to the TmDTPA

\[^{-}\text{containing KH, tuning and matching of the ^23Na resonator coil were performed, and homogeneity of the magnetic field was optimized. After 25 minutes of control perfusion, the constant-pressure perfusion was switched to constant-flow perfusion, and the flow was adjusted to 1\%, 2\%, or 3\% of coronary flow during the control period (\(n=4\) hearts per each LF group). After 60 minutes, R was started by switching the perfusion mode back to constant pressure for 45 minutes. \(^1\)H MRI, \(^23\)Na MR spectroscopy, and \(^23\)Na CSI measurements were carried out as depicted in Figure 1.

To investigate the spatial correlation between infarct and elevated Na\(^+\) image intensity, hearts (\(n=5\)) were subjected to regional ischemia for 40 minutes and R. To this end, a dual-perfusion cannula was used that allowed independent perfusion of the left and right coronary bed.\(^{13}\) During the stabilization period, the left side of the heart was perfused with a Gd-DTPA-BMA–containing perfusate (Omniscan, Amersham Health), and a T1-weighted \(^1\)H image was acquired to assess the area at risk. Total ischemia of only the left side of the heart was induced for 40 minutes (flow to the other side remained unaltered), followed by R. After 2 hours, the right side of the heart was perfused with methylene blue to determine the area at risk for histology. After that, the whole heart was perfused with triphenyl tetrazolium chloride (TTC) to stain the viable tissue, as described below.

### Assessment of Viability

To assess myocardial viability, recovery of the rate-pressure product (RPP=left ventricular developed pressure\times heart rate) and LVEDP were analyzed. In an additional group of experiments (1 or 2\% LF and R; \(n=4\) hearts per group), the hearts underwent the same protocol as before except that the reperfusion period was extended to 2 hours to allow reliable infarct size determination with TTC staining (Ytrehus et al\(^{14}\) and http://www.southalabama.edu/ishr/help/ttc/). After this period, hearts were perfused with 1\% TTC added to the perfusate for 10 minutes at a constant perfusion flow of ~2 mL/min. Hearts were then frozen and cut in five 2.5-mm-thick slices, which were immersed in 10\% formalin for 10 minutes. The slices were photographed, and infarct size was determined by planimetry and expressed as a percentage of total myocardial area. The effluent of these hearts was collected during the first 45 minutes of R and analyzed for creatine kinase (CK) activity at 25°C (Roche Diagnostics GmbH).

### Statistical Analysis

The data are summarized as the mean±SEM. A repeated-measures ANOVA compared measurements within each group. Differences between groups were analyzed by 1-way ANOVA. The test between any 2 means used Fisher’s least protected significant difference test. Differences were declared statistically significant when \(P<0.05\). Statistical computations were performed with SPSS software.

### Results

#### Hemodynamic Parameters

All groups of hearts displayed similar left ventricular pressures and thus, RPPs during control perfusion (Figure 2A). During LF ischemia, the time to onset of contracture and the time of maximal contracture were earliest in 1\% LF hearts (Figure 2B). Also, the amplitude of contracture was larger in the 1\% LF hearts than in the 2\% and 3\% LF hearts. In the 3\% LF group, contracture during ischemia occurred in only 1

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Figure 2. Relative RPPs (A) and LVEDPs (B) of isolated rat hearts during various degrees of LF perfusion and R. Open circles denote 1\%; filled squares, 2\%; and filled circles, 3\% of control perfusion from 0 to 60 minutes. All other abbreviations are as defined in text.
of LF ischemia, \([\text{Na}^+]\), had increased to 219±46% in 3% LF hearts (NS compared with the preischemic value), to 357±35% in 2% LF hearts (P<0.05 compared with the preischemic value), and to 396±18% in 1% LF hearts (P<0.05 compared with the preischemic value; Figure 4). In comparison, in our laboratory, the \([\text{Na}^+]\) increase after 20 and 30 minutes of total ischemia had been found to be \(\approx 200\%\) \(^{15}\) and 300%, \(^{6}\) respectively. Recovery of \([\text{Na}^+]\), during R was worst in 1% LF hearts: the average \([\text{Na}^+]\), remained highly elevated (321±41% at 40 minutes of R, P<0.05 compared with the preischemic value). In 2% and 3% LF hearts, \([\text{Na}^+]\), decreased during R to 218±12% (NS compared with end-LF value) and to 169±56% (P<0.05 compared with end-LF value), respectively, but remained elevated.

\(^{23}\)Na Chemical Shift Imaging

Short-axis \(\text{Na}^+\), and \(\text{Na}^+\), CS images, obtained in 4 minutes, 18 seconds, of 3 hearts subjected to different degrees of ischemia are shown in Figure 5. \(\text{Na}^+\), was already visible during control perfusion. It is clearly shown that \(\text{Na}^+\), image intensity increased with increasing severity of ischemia. During R, \(\text{Na}^+\), image intensity in the 1% LF hearts remained high, whereas that of the 2% LF hearts decreased but remained elevated. In the 3% LF group, 2 hearts showed no apparent changes in \(\text{Na}^+\), image intensity during the entire protocol (Figure 5), whereas the other 2 showed an increase in \(\text{Na}^+\), image intensity during ischemia. However, of these 2 hearts, only 1 showed recovery of \(\text{Na}^+\), image intensity during R. Quantitative analysis of \(\text{Na}^+\) images was done at 3 time slots: control perfusion, after 55 to 60 minutes of LF (end-LF), and after 35 to 40 minutes of R (end-R). Average total (cardiac) \(\text{Na}^+\) image intensity did not show significant changes during the protocol in all groups (Figure 6A), and \(\text{Na}^+\), image intensity was significantly lower at the end of R compared with control perfusion in the 1% LF group only (Figure 6B). \(\text{Na}^+\), image intensity, however, increased to 222±70% in the 3% LF group, to 291±47% in the 2% LF group, and to 394±25% in the 1% LF group (Figure 6C), findings that are consistent with the \(^{23}\)Na MR spectroscopy data. During R, \(\text{Na}^+\), image intensity decreased in all groups but remained highest in the 1% LF hearts (309±34% versus 171±18% in 2% LF hearts and 136±61% in 3% LF hearts).

\(^{23}\)Na MR Spectroscopy/CSI and Viability

There was a very good correlation between \([\text{Na}^+]\), measured by MR spectroscopy and the intensity of \(\text{Na}^+\), on \(^{23}\)Na images at the 3 analyzed time points (control, end-LF, and end-R): \(R=0.91, P<0.01\) (n=36). To evaluate whether changes in \([\text{Na}^+]\), or \(^{23}\)Na image intensities were related to recovery of hemodynamic performance and viability, we looked at the values of parameters of individual hearts rather than averages of the different groups. \(\text{Na}^+\), image intensity at end-R was correlated very well with RPP at end-R (\(R=-0.85, P<0.001\), Figure 7A) and with LVEDP at end-R (\(R=0.88, P<0.001\), Figure 7B). More important, however, there was also a good correlation between \(\text{Na}^+\), image intensity at end-LF with
LVEDP and with RPP at end-R (Figure 7C and 7D, $R = 0.78$, $P < 0.01$; and $R = -0.81$, $P < 0.05$, respectively). This shows that $\text{Na}^+_{\text{i}}$ image intensity can predict the ability of myocardial tissue to recover after ischemia. A similar very good correlation was found when $^{23}\text{Na}$ MR spectroscopy data were analyzed: $[\text{Na}^+_{\text{i}}]$ at end-R was correlated with LVEDP and inversely with RPP at end-R ($R = 0.93$, $P < 0.001$; and $R = -0.91$, $P < 0.001$, respectively). Again, $[\text{Na}^+_{\text{i}}]$ at end-LF was correlated very well with LVEDP ($R = 0.85$, $P < 0.01$) at end-R and inversely with RPP ($R = -0.86$, $P < 0.01$) at end-R, emphasizing the predictive value of $\text{Na}^+_{\text{i}}$ in the assessment of myocardial viability.

Infarct size measured by TTC staining was significantly smaller in the 2% LF group compared with the 1% LF group (15.8% versus 48.12%, $P < 0.05$). The same was true for CK release during the first 45 minutes of R: 63 ± 18 versus 132 ± 25 IU/g dry wt ($P < 0.05$). Infarct size was correlated very well with CK release ($R = 0.95$, $P < 0.001$). Importantly, $\text{Na}^+_{\text{i}}$ image intensity at end-LF was well correlated with CK release during R ($R = 0.79$, $P < 0.05$) as well as with infarct size ($R = 0.77$, $P < 0.05$).

Figure 8 shows short-axis $\text{Na}^+_{\text{i}}$ images of a heart subjected to acute regional ischemia (B–F) and R (G–J) and the corresponding $^1\text{H}$ gradient-echo images of the heart during control perfusion. Numbered arrows refer to glass reference capillary (1) and suction tubing (2). All other abbreviations are as defined in text.

Figure 6. Signal intensities on $^{23}\text{Na}$ images (relative to buffer $\text{Na}^+$ signal intensity) of isolated rat hearts during control (C) perfusion, at end of LF ischemia (endLF), and at end of R (endR). A, Total $\text{Na}^+$; B, $\text{Na}^+_{\text{e}}$; and C, $\text{Na}^+_{\text{i}}$ intensities. Open circles denote 1%; filled squares, 2%; and filled circles, 3% of control perfusion during LF. *$P < 0.05$ vs 2% LF group; †$P < 0.01$ vs 3% LF group; ‡$P < 0.05$ vs control value of same group; §$P < 0.05$ vs end-LF value of same group. All other abbreviations are as defined in text.
Discussion

In this study, the value of intracellular $^{23}$Na MRI for assessment of myocardial viability was explored in rat heart models of global LF ischemia and total regional ischemia. We found a very good correlation between Na$^+$ image intensity at end-R after LF ischemia and recovery of LVEDP and RPP at the same time. More important, however, there was also a good correlation between Na$^+$ image intensity at end-LF, with recovery of LVEDP and RPP at end-R. This shows that Na$^+$ image intensity can predict the ability of myocardial tissue to recover after ischemia. Furthermore, Na$^+$ image intensity at end-LF was well correlated with CK release during R as well as with infarct size. From regional ischemia experiments, a good spatial correlation between Na$^+$ image intensity and infarcted area was apparent. Total $^{23}$Na image intensities, however, did not change during ischemia and R, indicating that under these conditions, total $^{23}$Na MRI cannot be used to assess viability. This is the first study that shows the potential of intracellular $^{23}$Na MRI as a diagnostic modality for early detection of myocardial ischemia and viability.

$^{23}$Na MRI of an isolated rat heart was first reported by DeLayre et al.\textsuperscript{16} The heart could be visualized because of the lack of signal from the ventricular wall compared with signal from the ventricular cavities and buffer surrounding the heart. The technique has been improved since then, and total $^{23}$Na imaging has been performed on the hearts of rats,\textsuperscript{17} rabbits,\textsuperscript{9} and dogs,\textsuperscript{9,18,19} as well as humans.\textsuperscript{20,21} $^{23}$Na CSI with the aid of a shift reagent makes it possible to independently analyze the spatial distribution of Na$^+$, Na$^{++}$,
and total Na\(^{+}\) and therefore allows for a more precise understanding of pathophysiological changes in ion homeostasis during ischemia and reperfusion. In the regional ischemia experiments, Na\(^{+}\) imaging appeared superior to total Na\(^{+}\) imaging in identifying the affected zone after induction of ischemia, as we had also found before, after acute left coronary artery ligation of isolated rat hearts.\(^{11}\) The reason for this was that the occlusion was total, and total Na\(^{+}\) remained unchanged because of an apparently undeveloped collateral circulation, whereas Na\(^{+}\) influx into the intracellular compartment exceeded Na\(^{+}\) efflux as a result of ischemia. We anticipated that during LF ischemia, the situation might be totally different. Total Na\(^{+}\) intensity was expected to rise during LF ischemia, because [Na\(^{+}\)]\(_{i}\) will rise and [Na\(^{+}\)]\(_{e}\) will remain constant, in contrast to conditions of total occlusion, wherein [Na\(^{+}\)]\(_{i}\) image intensity will also rise, but at the expense of [Na\(^{+}\)]\(_{e}\) image intensity, resulting in no change of total Na\(^{+}\) image intensity. However, we observed no increase in total Na\(^{+}\) and a decrease in Na\(^{+}\) image intensities only in the group in which ischemia was most severe, indicating that in this group, the flow rate was insufficient to refresh the perfusate in the extracellular space. It is important to note that this decrease in total Na\(^{+}\) intensity was not beneficial, as might be falsely concluded if Na\(^{+}\) images would not have been available. The behavior of the Na\(^{+}\) image intensities during the protocol was as anticipated: the more severe the degree of ischemia, the larger the increase in [Na\(^{+}\)]\(_{i}\), and corresponding Na\(^{+}\) image intensity. This latter intensity appeared to be highly predictive for functional recovery during reperfusion. Ca\(^{2+}\) accumulation in the myocardial cell appears to play a central role in the damage that occurs during reperfusion of ischemic myocardium.\(^{22}\) One of the most important routes for Ca\(^{2+}\) influx during ischemia and in particular, during R is (reversed) Na\(^{+}\)-Ca\(^{2+}\) exchange, and therefore, increased [Na\(^{+}\)]\(_{i}\) may underlie a significant portion of this damage.\(^{23}\)

Thus, imaging of both Na\(^{+}\)\(_{i}\) and Na\(^{+}\)\(_{e}\) is essential for proper interpretation of total cardiac Na\(^{+}\). Furthermore, formation of edema can lead to increased total Na\(^{+}\) intensity (because of the extension of the extracellular space), whereas [Na\(^{+}\)]\(_{i}\) may not be increased, indicating that the cardiomyocytes are still viable.\(^{20}\) In other words, the discriminating power of the CSI technique is much higher (see the Table) compared with total \(^{23}\)Na and other imaging techniques.

### Methodological Considerations

In cardiac tissue, the concentration and NMR sensitivity of the \(^{23}\)Na nucleus are much lower than that of 1\(^{H}\), leading to a 20 000 to 30 000 times lower signal compared with that of 1\(^{H}\) in water. This disadvantage can partially be overcome by using larger voxel sizes, longer acquisition times, and faster pulse sequences. The latter can be used because of the shorter relaxation times of \(^{23}\)Na. Using a 16×16 matrix and acquisition weighting, we were able to acquire a full CSI dataset (256 phase-encoding steps) with a reasonable signal-to-noise ratio of the Na\(^{+}\) image in 4 minutes, 18 seconds. The disadvantage of the acquisition weighting is a slightly worse spatial resolution, but this is outweighed by the resulting improvement of the spatial response function, which prevents Gibbs ringing, and a timesaving effect.

The intensity of the (intracellular or extracellular) Na\(^{+}\) signal depends on (1) the Na\(^{+}\) content of the tissue, (2) MR visibility, and (3) the size of the intracellular and extracellular compartments. With regard to (1), myocardial ischemia results in an increase in [Na\(^{+}\)]\(_{i}\), because of continued or enhanced Na\(^{+}\) influx and reduced Na\(^{+}\) pumping.\(^{24}\) Suppose that during LF ischemia [Na\(^{+}\)]\(_{i}\) increases by 100% (from 10 to 20 mmol/L) and [Na\(^{+}\)]\(_{e}\) remains constant (150 mmol/L). Then, with relative sizes of the intracellular and extracellular space of 75% and 25%, respectively, Na\(^{+}\) image intensity doubles but total Na\(^{+}\) image intensity will increase by only \(\approx 17\%\). This shows that Na\(^{+}\) imaging is much more sensitive than total Na\(^{+}\) imaging and might explain why we did not find any significant changes in total Na\(^{+}\) intensity during our protocol.

With regard to (2), because of transverse (T2) relaxation during the 1000-\(\mu\)s pulse and the 307-\(\mu\)s postpulse delay in our CSI pulse sequence, signal loss occurs, which, based on our previous studies,\(^{15}\) we calculated to result in a reduction of MR visibility of \(\approx 11\%\). In the MR spectroscopy experiments, the loss was \(<1\%\), resulting in a visibility of \(>99\%\). However, the correlation coefficients calculated from the CSI data were on the same order as those calculated from the spectroscopy data, indicating that neither the signal loss resulting from the unavoidable delay in the pulse sequence nor the use of magnitude images for the determination of Na\(^{+}\) image intensity affected our results. Furthermore, the \(^{23}\)Na CSI sequence used on 4% agar solution with different Na\(^{+}\) concentrations (20, 50, 100, 150, and 200 mmol/L NaCl) showed an excellent correlation between Na\(^{+}\) concentration and \(^{23}\)Na image intensity \((R=0.996, P<0.001, \text{data not shown})\).

With regard to (3), when the sizes of intracellular and extracellular space change as a result of an intervention, \(^{23}\)Na image intensity will be affected. Either or both the intracellular and extracellular spaces may change as a result of edema and contracture. However, the size of the ventricles on the 1\(^{H}\) images before and after the protocol never differed by \(>10\%\). Apart from assuring that under all circumstances, the region selected for Na\(^{+}\) image intensity calculation was well within the outer contour of the left ventricle, no correction for swelling or contraction was applied.

### Clinical Implications

\(^{18}\)F-2-deoxyglucose positron emission tomography\(^{2}\) and \(^{201}\)Tl scintigraphy\(^{26}\) are being used clinically to assess myocardial viability, and, like \(^{23}\)Na MRI, test sarcolemmal function with a comparable spatial resolution. However, these techniques involve appreciable radiation exposure for the patient, in particular, in serial examinations. In addition, \(^{23}\)Na MRI has

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**Predicted Signal Intensities of \(^{23}\)Na MR Images in Infarcted and Jeopardized Myocardial Tissue Compared With Healthy Tissue**

<table>
<thead>
<tr>
<th>Condition</th>
<th>Total Na(^{+})</th>
<th>Na(^{+})(_{i}) (+SR)</th>
<th>Na(^{+})(_{e}) (+SR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chronic infarct (scar tissue)</td>
<td>↑↑</td>
<td>↑↑</td>
<td>0</td>
</tr>
<tr>
<td>(Semi)acute total occlusion</td>
<td>=</td>
<td>↑↓</td>
<td>↑</td>
</tr>
<tr>
<td>Partial occlusion</td>
<td>=↑↑</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Extracellular edema</td>
<td>↑</td>
<td>↑↑</td>
<td>↑</td>
</tr>
</tbody>
</table>

SR indicates shift reagent; ↑, increased; ↓, decreased; and =, unaltered. All other abbreviations are as defined in text.
the advantage that it can easily be combined with 1H MRI for evaluation of wall-motion abnormalities and cardiac function. DCE-MRI is rapidly becoming the new “gold standard” for assessment of myocardial viability in the chronic situation. However, it remains unclear how the difference in contrast is established. Furthermore, especially in the (sub)acute phase, signal enhancement due to edema in the peri-infarct zone can lead to overestimation of the affected area. In contrast to 23Na MRI, no information on the (patho)physiological state of the myocardium is obtained with DCE-MRI. The lower spatial resolution of 23Na MRI can be partially overcome by using 1H MR images as anatomic references.

So far, only total Na\(^+\) MRI has been applied in humans. Intracellular Na\(^+\) imaging requires a shift reagent suitable for use in humans, which is currently unavailable. With the development of magnets with higher magnetic field strengths, total acquisition time can be reduced, spatial resolution can be increased, or both. Extension of the technique to 3 spatial dimensions is highly desirable.

**Conclusions**

This is the first study that shows the feasibility of 23Na MR CSI for visualization of Na\(^+\), under (patho)physiological conditions. These data demonstrate that 23Na CSI at 9.4 T can be used to assess viability in isolated, perfused rat heart models of ischemia. The results of this study may contribute to the development of a new clinical diagnostic imaging modality for early detection of ischemia and viability, which may also help in evaluation of the efficacy of pharmacological therapies.

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**References**

9. Kim RJ, Lima JA, Chen EL, Reeder SB, Klocke FJ, Zerhouni EA, Judd RM. Fast 23Na magnetic resonance imaging of acute reperfused myo-
Assessment of Myocardial Viability by Intracellular $^{23}$Na Magnetic Resonance Imaging
Maurits A. Jansen, Jan G. Van Emous, Marcel G.J. Nederhoff and Cees J.A. Van Echteld

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