Effect of global ischemia and reperfusion during ventricular fibrillation in myopathic human hearts

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Massé S, Farid T, Dorian P, Umapathy K, Nair K, Asta J, Ross H, Rao V, Sevaptsidis E, Nanthakumar K. Effect of global ischemia and reperfusion during ventricular fibrillation in myopathic human hearts. Am J Physiol Heart Circ Physiol 297: H1984–H1991, 2009. First published October 9, 2009; doi:10.1152/ajpheart.00101.2009.—The effect of lack of global coronary perfusion on myocardial activation rate, wavebreak, and its temporal progression during human ventricular fibrillation (VF) is not known. We tested the hypothesis that global myocardial ischemia decreases activation rate and spatiotemporal organization during VF in myopathic human hearts, while increasing wavebreak, and that a short duration of reperfusion can restore these spatiotemporal changes to baseline levels. The electrograms were acquired during VF in a human Langendorff model using global mapping consisting of two 112-electrode arrays placed on the epicardium and endocardium simultaneously. We found that global myocardial ischemia results in slowing of the global activation rate (combined endo and epi), from 4.89 ± 0.04 Hz to 3.60 ± 0.04 Hz, during the 200 s of global ischemia (no coronary flow) (P < 0.01) in eight myopathic hearts. Two minutes of reperfusion contributed to reversal of the slowing with activation rate value increasing close to VF onset (4.72 ± 0.04 Hz). In addition, during the period of ischemia, an activation rate gradient between the endocardium (3.76 ± 0.06 Hz) and epicardium (3.45 ± 0.06 Hz) was observed (P < 0.01). There was a concomitant difference in wavebreak index (that provides a normalized parameterization of phase singularities) between the epicardium (11.29 ± 2.7) and endocardium (3.25 ± 2.7) during the 200 s of ischemia (P = 0.02). The activation rate, gradient, and wavebreak changes were reversed by short duration (2 min) of reperfusion. Global myocardial ischemia of 3 min leads to complex spatiotemporal changes during VF in myopathic human hearts; these changes can be reversed by a short duration of reperfusion.

wavebreak incidence; phase singularity

DURING UNTREATED VENTRICULAR fibrillation (VF) in humans, profound changes have been recorded in surface electrical activity with evolution of time (24). In addition, the duration of VF, before resuscitation, has been recognized as a major determinant of survival (1). This had led to the practice of immediate rhythm analysis and defibrillation if a shockable rhythm is identified. Recently, it has been suggested that a period of coronary perfusion before rhythm analysis is preferable to immediate rhythm analysis in cardiac arrest patients and is being assessed in a randomized clinical trial (2). The lack of coronary perfusion (“no flow” time) leads to evolution of the VF process (6, 8, 12, 13, 17–19, 21, 22, 28, 32, 33, 38); however, the mechanisms that are operative during that time-frame at the endocardial and epicardial levels are yet to be defined in humans. The absence of coronary perfusion on excitability and complex wavebreak in human VF is not known and may have important mechanistic implications, since wavebreak is important in sustaining VF. Experimental models have evaluated this issue; recently, studies have been conducted in healthy animal hearts, using either an in vivo preparation or Langendorff preparation. Activation mapping (electrically and optically) was made, mainly from the epicardium, to make observations regarding organization and possible mechanisms that may be operational during global ischemia (3, 10, 14, 15, 36). However, in humans, it is ethically difficult to study the temporal evolution of VF, especially on the myocardial surface, and quantify the influence of reperfusion on this process. Using a human Langendorff model, we tested the hypotheses that global myocardial ischemia during VF in myopathic hearts decreases activation rate and regularity while increasing wavebreaks, and that a short duration of reperfusion can restore these parameters to baseline levels.

METHODS

We studied eight Langendorff-perfused human hearts, which were explanted from patients with cardiomyopathy who underwent transplantation. Table 1 summarizes associated patient data. The investigation followed the principles outlined in the Declaration of Helsinki, and the research protocol was approved by the University Health Network Human Ethics Board. Immediately after being explanted, the hearts were preserved in a cardioplegic solution and transferred to our research laboratory. They were cannulated from the aorta and flushed thoroughly with Tyrode solution [composition: 6.9 g NaCl, 350 mg KCl, 370 mg CaCl₂(H₂O)₂, 140 mg MgSO₄, 2.09 g NaHCO₃, 160 mg of NaPO₄, and 1.09 g of glucose/l of solution]. The Tyrode was oxygenated with a pediatric oxygenator connected to a carbogen (95% O₂-5% CO₂) cylinder. The flow was maintained at 0.9 to 1.1 ml·g⁻¹·min⁻¹. During the experiments, Tyrode temperature was maintained at 38°C, and the hearts were placed in a heat-jacketed reservoir, also regulated at 38°C. The epicardial/endocardial temperature gradient was monitored and never varied by >0.2°C even during global ischemia, by submerging the heart in the circulating warm Tyrode. Multi-electrode mapping was performed on the entire epicardium using an 112-electrode sock array and on the left ventricular (LV) endocardium using another 112-electrode balloon array. Both unipolar and bipolar electrograms were recorded from each electrode. Heart rate was kept constant by pacing the anterolateral LV at 600 ms cycle length using bipolar hook electrodes. VF was induced using a 9-volt battery on the epicardium. As soon as a VF episode was
induced, perfusion was turned off to the heart for 200 s, mimicking global ischemia of long duration as may be seen in human cardiac arrest. At the end of this period, the perfusion system was turned back on to a flow rate similar to baseline for an additional 140 s (Fig. 1). The hearts were then defibrillated, and this protocol was repeated after a rest period of 15 min, giving a total of 16 VF episodes. Even though most investigators using Langendorff preparations subject their preparation to multiple inductions and defibrillations, for the purposes of this study, we subjected the hearts to only two inductions. Wu et al. (37), the only other group that has worked with myopathic explanted human hearts, found three inductions of VF (15 episodes of VF in 5 hearts) yielded stable fibrillation dynamics in these myopathic heart preparations as we did in the past (25).

An ischemia period of 200 s was chosen for this study after testing various ischemia durations on myopathic heart models, since we found that 200 s was the maximum ischemia that will allow for second induction and allow for completion of the protocol without edema interfering with the preparation. Longer periods would have been preferable to more accurately represent out-of-hospital cardiac arrest situations; however, the duration studied here has implications for witnessed arrests and in-hospital cardiac arrest and allows for proper completion of the protocol.

Mapping tools. The electrode arrays construction and mapping system has been described previously (29). Each endocardial electrode consists of two silver beads (2 mm diameter) separated by 2.1 mm center to center. Like the balloon electrode arrays, the sock is made of 112 bipolar electrodes organized in 14 rows of 8 electrodes each, mounted on an extensible mesh. Simultaneous unipolar and bipolar electrograms were recorded from all electrodes. The bipolar electrograms were evaluated to ensure appropriate contact. To represent these electrode arrays in two dimensions (2-D), polar maps are used, and have been described previously (20).

Data acquisition and analysis. The filter settings for unipolar signals were set to 0.5–200 Hz, and the sampling rate was 1,000 samples/s. We used Matlab 2007b (Mathworks, Natick, MA) for data analysis. After data acquisition, all electrograms were low-pass fil-

### Table 1. Isolated human hearts clinical data

<table>
<thead>
<tr>
<th>No.</th>
<th>Sex (M/F)</th>
<th>Age, yr</th>
<th>Diagnosis</th>
<th>EF, %</th>
<th>LV Dimensions Diastolic/Systolic, mm</th>
<th>Medication</th>
<th>ICD (Y/N)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>M</td>
<td>25</td>
<td>IDCM</td>
<td>20–29</td>
<td>47/35</td>
<td>Asa, Altace, Ferrous gluconate, Pravastatin, Atacand, Plavix, Aspirin, Lasix, Aspirin, Clopidogrel, Enalapril, Amlodipine, Nitro patch, Niacin, Enalapril, Coreg, Asa, Lipitor, None</td>
<td>Coumadin, Ciprofloxacin</td>
</tr>
<tr>
<td>2</td>
<td>F</td>
<td>32</td>
<td>ICM</td>
<td>20–29</td>
<td>53/36</td>
<td>Metoprolol, Eltroxin, Aldactone, Coumadin, Digoxin, Amiodarone, Furosemide, Plavix, Amiodarone, Clonazepam, Lorasepam, Spironolactone</td>
<td>N</td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>56</td>
<td>IDCM</td>
<td>20–29</td>
<td>45/36</td>
<td>N</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>M</td>
<td>56</td>
<td>ICM</td>
<td>&lt;20</td>
<td>74/64</td>
<td>Plavix, Amiodarone, Clonazepam, Lorasepam, Spironolactone</td>
<td>Y</td>
</tr>
<tr>
<td>5</td>
<td>M</td>
<td>34</td>
<td>IDCM</td>
<td>&lt;20</td>
<td>59/50</td>
<td>Coumadin, Slow K, Prevacid, Metolazone, Spiriva, Advair, Salbutamol, Furosemide, Levotroxine, Sodium, Spironolactone, Ramipril, Venlafaxine, Sildenafil, citrate, Amiodarone, Coumadin</td>
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</tr>
<tr>
<td>6</td>
<td>F</td>
<td>48</td>
<td>ICM</td>
<td>&lt;20</td>
<td>60/48</td>
<td>N</td>
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</tr>
<tr>
<td>7</td>
<td>M</td>
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<td>20–39</td>
<td>64/59</td>
<td>N</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>M</td>
<td>59</td>
<td>ICM</td>
<td>&lt;20</td>
<td>57/54</td>
<td>Acetylsalicylic acid, Omeprazole, Magnesium, Amlodipine, Warfarin sodium, Potassium chloride, Atenolol, Spironolactone, Levothyroxine, Sodium, Spironolactone, Ramipril, Venlafaxine, Sildenafil, Amiodarone, Coumadin</td>
<td>N</td>
</tr>
</tbody>
</table>

M, male; F, female; IDCM, idiopathic dilated cardiomyopathy; ICM, ischemic cardiomyopathy; EF, ejection fraction; LV, left ventricle; Y, yes; N, no; ICD, implantable cardio-defibrillator. Patient 5 did not receive any medication before transplant because he was an acute case and he did not have time to get on medications.

Fig. 1. Ischemia-reperfusion protocol. After ventricular fibrillation (VF) was induced, perfusion was turned off to the heart for 200 s, mimicking global ischemia of long duration as may be seen in human cardiac arrest. At the end of this period, the perfusion system was turned back on to a flow rate similar to baseline for an additional 140 s (Fig. 1). The hearts were then defibrillated, and this protocol was repeated after a rest period of 15 min, giving a total of 16 VF episodes. Even though most investigators using Langendorff preparations subject their preparation to multiple inductions and defibrillations, for the purposes of this study, we subjected the hearts to only two inductions. Wu et al. (37), the only other group that has worked with myopathic explanted human preparations found three inductions of VF (15 episodes of VF in 5 hearts) yielded stable fibrillation dynamics in these myopathic human heart preparations as we did in the past (25).

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tered at 60 Hz using a 112th-order FIR equiripple filter and down-sampled to 125 Hz. For the analysis, 20-s electrograms from both endocardial and epicardial electrode arrays were recorded at time 0, 90, 180, 260, and 320 s for 16 VF episodes, for a total of 80 datasets. The electrograms located on the LV free wall were used for the analysis, for a total of 160 electrograms for each set.

**Dominant frequency mapping.** Previous publications have described the use of dominant frequency (DF) mapping to assess domains on the epicardium (39). Local activation rates (LAR) were calculated using average modified periodograms. For the analysis, a 2-D power spectral density (PSD) was estimated using the Welch averaged modified periodogram performed on the electrograms with a frequency resolution of 0.25 Hz. This was achieved by segmenting the 20-s recording data into five blocks of 4 s each (no overlapping). Hanning filter was used to reduce edge effect, and 1,000-point fast-Fourier transform was used to calculate the frequency spectrum. LAR for each electrogram was determined as the DF corresponding to the highest value of the power spectrum between 1.5 to 8 Hz.

**Spatial organization of VF wavebreak.** For estimating spatial organization (relationship between electrograms), we used wavebreak incidence (WBI) as described recently by Huizar et al. (15). In this technique, singularity point trajectories (SPT) are identified in phase movies, and each starting point of SPT is considered as the site of a new wavebreak. WBI is obtained by normalizing to the size of mapped area and the average number of excitation cycles. We used the technique described by Nash et al. (27) to compute phase maps for both endocardial and epicardial surfaces using unipolar electrograms. A 120 × 120 grid of interpolated electrograms was constructed by using triangle-based cubic interpolation of the detrended electrograms. This technique ensured that the interpolated surface always passed through all data points with no discontinuities in the first and zero derivatives (5). The results of this processing gave a series of 120 × 120 pixel frames, each representing 8 ms of data. Phase singularities (PS) were identified using a semiautomatic algorithm. Finally, the total number of PS was normalized to the average number of excitation cycles estimated as mean DF × t where t is the duration of the recording and mean D) the average DF from the 112 electrograms recorded on each surface.

**Temporal organization of VF.** Different techniques have been used to quantify temporal electrogram organization using unipolar electrograms. The unipolar electrogram technique we used was first described by Zaitsev et al. (39) to measure the degree of local periodicity of action potentials obtained from optical mapping, which we adapted for unipolar recordings to create a frequency-based regularity index (FRI). The FRI is defined as the ratio of the power at the DF to the total power. For this study, the power at the DF was calculated by summing the power values at the highest peak and its adjacent values (fixed band of 1 Hz), and the total power was calculated as the sum over the range of 1.5–8 Hz. This technique provides an estimate of the amount of side components that are present in the electrograms, which are caused by conduction block, wave collision, nonstationarity, etc. For very organized and regular electrograms during VF, the PSD usually show a single sharp peak where most of the power is present (high FRI value), whereas PSD from irregular and disorganized electrograms exhibit either wide single peak or more usually multi-peak (multicomponent signals, low FRI). This major aim using this technique is to verify whether high-frequency sources correlate with a higher level of organization, which is what one would expect to see near a rotor core.

**Effects caused by cardiomyopathy.** As shown in Table 1, the hearts used in this study were from patients affected with two different types of diseases and had varying LV ejection fraction, both factors that may have influenced the evolution of VF during our protocol. To evaluate their effects, we added these factors (type of cardiomyopathy and LV ejection fraction) into our analysis. Ejection fraction was divided into the following two groups: >20% and ≤20% LV ejection fraction.

**Differential LV/RV response.** LV vs. right ventricle (RV) frequency gradient has been observed in animal models during VF (7). Although the focus of this study was the LV free wall, we tested the hypothesis that a similar gradient is present as well in myopathic human hearts by comparing the dominant frequencies measured from the 32 electrograms recorded on the RV side of the epicardial sock with the 80 electrograms measured from the LV.

**Statistical analysis.** The data were analyzed using Matlab 2007b with statistical toolbox (Mathworks). Values are shown as means ± 95% confidence intervals. A statistical model using a three-way ANOVA was employed to test the main effects of time, heart surface (epi vs. endo), electrode position, and associated two- and three-way interactions for the 80 datasets (16 VF episodes with 5 repeated measures). When the F statistic was significant, in case of multiple comparisons, Scheffé’s post hoc test was used to specify the differences, and the significance levels were adjusted accordingly. Finally, WBI analysis was performed on the first VF episode obtained from six out of the eight isolated hearts. Repeated-measures ANOVA were performed to study the influence of the different etiologies of the myopathic process, ejection fraction, and chamber specific influences on fibrillation dynamics. There were no missing values in our datasets.

**RESULTS**

**LAR evolution during ischemia and reperfusion.** Figure 2A shows the evolution of the average LAR over time; the statistical model revealed that duration of global ischemia has a significant effect (P < 0.01). The LAR decreased from 4.89 ± 0.08 to 3.60 ± 0.08 Hz during the 200 s of global ischemia (P < 0.01). During the 140 s of reperfusion, the activation rate increased close to baseline at 4.72 ± 0.08. This value was comparable to the measure just after the onset of the VF. ANOVA for LAR showed that there was no three-way interaction between electrode position, heart surface, and time. In our study where RV and LV activation rates were compared, ANOVA did not reveal a significant interaction between time and heart chamber (P = 0.199).

**Activation rate gradient between epicardium and endocardium.** Figure 2B shows the gradient that develops during ischemia. The ANOVA showed that there was a significant difference of LAR measured on the endocardium between baseline and the end of ischemia (P < 0.01). Similar results were also seen between the end of ischemia and end of reperfusion. At the end of the ischemia period, a gradient of LAR was observed between epicardium and endocardium 3.76 ± 0.12 to 3.45 ± 0.12 Hz (P < 0.01), with endocardium activating faster than the epicardium. This effect was reversed during the reperfusion with LAR, equalizing between epicardium and endocardium (P = 0.4 at t = 260 s). We verified carefully that this was not related to temperature gradient by continually monitoring temperature on the epicardium and endocardium during the ischemia phase and reperfusion. ANOVA revealed an interaction between time and heart pathology (P < 0.01), revealing that there was a greater gradient developed on dilated hearts than on ischemic hearts during ischemia. The ANOVA also showed that the hearts with ejection fraction <20% had significantly greater DF gradient than hearts with ejection fraction >20% (P < 0.01) but only at the end of ischemia.

**Spatial organization of VF during ischemia-reperfusion.** WBI, an index that provides a normalized parameterization of PS, was evaluated during the protocol, and the results are shown in Fig. 3. On the epicardium and endocardium, the WBI

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at baseline was 6.46 ± 2.7 and 2.84 ± 2.7, respectively, and during the 200 s of ischemia the WBI on the epicardium was 11.29 ± 2.7 compared with the endocardium 3.25 ± 2.7 ($P = 0.02$). The increase on the epicardium was reversed close to the baseline during reperfusion (WBI = 7.48 ± 2.7, $P$ value from baseline = 0.63). There was no significant change of WBI observed on the endocardium (from 2.84 ± 2.7 at baseline to 2.80 ± 2.7 at end of reperfusion, $P = 0.97$). These differences are better illustrated by time-space plots. In short, time-space plots show in a single picture the evolution of electrical activity over time for a given region of the heart. Data from each 8-ms interpolated electrogram frame are projected into a line. All of the lines are successively stacked sequentially to form an image of time vs. space. The third dimension (intensity) is proportional to the amplitude of the electrogram. This representation is useful to identify areas of block or rotor cores that can meander with time. Two sets of time-space plots are shown on Fig. 4, $B$ (epicardial data) and $C$ (endocardial data). They are derived from corresponding locations on endocardium and epicardium for ease of comparison (shown as 2 blue curves drawn over three-dimensional phase maps in Fig. 4A). This shows that the endocardial activation becomes slower during ischemia with no increase in wavebreak. In contrast, epicardial activation becomes slower and shows more wavebreaks during the course of ischemia, demonstrating a differential effect on the epicardium.

**Temporal organization of VF during ischemia-reperfusion.**

FRI from unipolar electrograms was used to assess temporal organization. Figure 5 shows the evolution of the organization during ischemia and reperfusion. At baseline ($t = 0$), both endocardium and epicardium sites showed similar degrees of organization (0.54 ± 0.02 vs. 0.52 ± 0.02; $P = 0.19$). During an initial transient phase of 90 s during ischemia, there was increased organization on both surfaces, and this difference was significant on the endocardium ($P < 0.01$). At the end of the ischemia period, the organization levels for both surfaces...
were restored and were not different to the baseline level (0.55 \pm 0.02 and 0.52 \pm 0.02; \( P = 0.49 \) and \( P = 0.68 \)). After 60 s of reperfusion, both surfaces showed reversal of trend in level of organization compared with the respective values to what was seen at the onset of the VF, with overshoot (0.47 \pm 0.02 for epi; 0.49 \pm 0.02 for endo).

**DISCUSSION**

We have demonstrated in this study that, with the onset VF, the ensuing global myocardial ischemia leads to progressive spatiotemporal changes in electrical activation pattern on the human epicardium and endocardium within 3 min. These changes are probably the result of complex interplay between conduction velocity, excitability, refractoriness, and wavelength. The important finding of this study is that, during the period of ischemia, there was slowing of local myocardial activation rates and development of activation rate gradient between the endocardial and epicardial surfaces. There were concomitant differences between the epicardium and endocardium with regard to wavebreak, with greater occurrence of wavebreaks on the epicardium during ischemia. The activation rate, gradient, and wavebreak changes were reversed by a short duration of reperfusion. FRI during reperfusion had reversed its direction compared with ischemia toward the baseline value of FRI. It is important to note that Fig. 5 shows that, during reperfusion, FRI overshoots below baseline values. The overshoot of VF dynamics during reperfusion compared with baseline values seems to be a repeating phenomenon seen in the literature (3, 10). The mechanism of this overshoot is not known and not discussed elsewhere; it may relate to the recovery of excitability such that conduction velocity recovery precedes recovery from refractoriness and postrepolarization refractoriness such that VF wave fronts are many and faster but tend to block, leading to faster but disorganized-appearing VF dynamics.

**Fig. 4.** Time-space plots and phase maps from epicardium and endocardium during ischemia-reperfusion. A: left anterior view of the same model, showing only the electrodes with phase data. The blue lines show corresponding endo and epi areas that are used to generate respective time-space plots shown in B and C. B: epicardial phase maps and time-space plots during ischemia-reperfusion show a decrease in the frequency of wavefronts and increased fractionation of the time-space plots with ischemia that are reversed by reperfusion. RV: right ventricle. C: endocardial phase maps and time-space plots during ischemia-reperfusion also show a decrease in the frequency of wavefronts that are reversed by reperfusion. However, this change is not as pronounced, and less fractionation is seen on the time-space plots. Phase singularities are marked with white dots on both sets of phase maps. Note the absence of phase singularities on the endocardial phase maps during baseline and ischemia. There is further organization transiently during ischemia that is reversed by reperfusion. Please see text for more details.

**Fig. 5.** Evolution of organization during ischemia and reperfusion. The frequency-based regularity index shows an initial increase in organization during the first half of the ischemia period seen on the endocardium, which is reversed at the end of ischemia. A smaller increase of organization was also found on the epicardium, but was not significant.
Differential VF activation between endoepicardium in humans during ischemia. There are baseline differences in WBI and number of PS between endocardium and epicardium. Although the specific mechanisms have not been evaluated, we and others have previously seen a greater degree of organization of human VF on the endocardium compared with the epicardium (20, 26, 31). The strength of the simultaneous epicardial and endocardial mapping we deployed in this study has allowed us to demonstrate that, in humans, the activation rate decreases and that an endocardial-to-epicardial activation rate gradient develops during the evolution of VF. This gradient has also been observed in other species during VF in ischemia (9, 35, 36). One of the possible explanations for this gradient may lie in the differential expression K channels that ultimately determine K$^+$ concentration (7). ATP-dependent K$^+$ (K$_{ATP}$) channels are critically dependent on high-energy phosphates and global ischemia. More importantly, they have been characterized to be differentially expressed between the epicardium and endocardium (23). Miyoshi et al. (23) also found epi-endo differences in K$_{ATP}$ channel in a dog model and showed that ischemia-induced extracellular K rise was suppressed by glibenclamide on the epicardium while the rise on endocardium remained unaffected. We believe that this differential expression of K$_{ATP}$ channel density between epicardium and endocardium may also serve as an alternate explanation for the rate and organization differences between epicardium and endocardium surface seen in this study. This hypothesis needs further investigation and will be the subject of a future study, where pharmacological agents will be used to selectively open and close the K$_{ATP}$ channels. This, we hope, will allow us to modulate the rate gradient between epicardium and endocardium and its possible effects on human VF.

Caldwell et al. (7) found that there was significant intercompartamental DF heterogeneity during ischemia, and they postulated that the differential expression of inward-rectifier potassium current may be responsible for these gradients in VF dynamics. Unlike Caldwell et al., we did not find gradient of activation rates between LV and RV epicardial electrograms. In our study where RV and LV activation rates where compared, ANOVA did not reveal a significant interaction between time and heart chamber ($P = 0.199$).

The other possible explanation for this gradient may lie in the differential distribution of subendocardial Purkinje fibers that reside on the endocardium and the fact that they are resistant to ischemia compared with working myocardin (9). It is plausible that the midmyocardial, subepicardial junction leads to greater wavebreak on the basis of ischemic regions in the intramural zones (16). In our study, the endocardium during VF had a greater degree of spatiotemporal regularity and less wavebreak. Although there was spatiotemporal disorganization with 200 s of ischemia, there was an initial increase of organization seen on the endocardium in the early stage of ischemia. A similar transient initial increase of organization has also been reported by Huang et al. (14) and Huizza et al. (15), but on the epicardium. Using recordings made on the epicardium of isolated pig hearts, Huizza et al. (15) found that the recurrence of propagation direction increased during early ischemia (between 1 and 2 min), although in our case we observed this phenomenon only on the endocardium, and this may relate to the fact that, in ungulates, the Purkinje fiber distribution is different from humans. Wu et al. (36) also observed a gradient between endocardium and epicardium on the LV of a rabbit heart during ischemia, which was also reverted during reperfusion.

Temperature differences are known to have significant effect on activation rate. We monitored the temperature gradient carefully during the state of no flow during our experiments. Given the fact that the Langendorff preparation was submerged in warmed Tyrode and that there were no significant differences in temperature, we hypothesize that gradient changes may be caused by one of many factors discussed above.

Potential implications for cardiopulmonary resuscitation. Although our study provides mechanistic insight into VF dynamics during global ischemia, the translation value of this study lies in its extrapolation to resuscitation science. The findings of our study provide the intriguing possibility of relating the human perfused heart VF recordings from the epicardium and endocardium to clinical studies of electrocardiogram (ECG) during VF arrest. For example, it is plausible that VF recorded using body surface ECGs is mostly affected by epicardial activation pattern, but the response to shock is determined by both epicardial and endocardial activation. This could potentially explain why shocking cardiac arrest victims based on surface ECG has limitations in predicting outcome. It has been recognized that resuscitation outcomes following successful defibrillation may be poor on the basis that the ischemic heart leads to asystole (pulseless electrical activity) or the heart may be too ischemic to provide meaningful mechanical activity.

Our current paradigm of treatment for witnessed VF arrests suggests that at least for patients treated early after VF onset, attempts should be made to deliver defibrillation without delay. The Seattle Group observed that 90 s of chest compression before shock compared with immediate analysis (not randomized) was associated with better outcome, especially for response intervals >4 min (11). Wik et al. (34), testing the specific hypothesis of whether cardiopulmonary resuscitation (CPR) should precede shocking, found that early and late was equivalent for patients with ambulance response times shorter than 5 min. However, patients for whom response time was >5 min had better survival with upfront CPR than patients with immediate rhythm analysis.

The Resuscitation Outcomes Consortium (ROC) investigators are conducting a large clinical trial that will test the two strategies: 1) early rhythm analysis vs. 2) a period of 2–3 min CPR after arrival of emergency medical service personnel (2). Our study was not meant to simulate out-of-hospital cardiac arrest but test the physiological principles of global ischemia and reperfusion. We found that perfusion restores higher VF activation rates similar to that found early after VF onset, providing the physiological rationale for the paradigm of upfront CPR preanalysis in humans. This is especially important, since it has been shown that the probability of success depends on the median VF frequency and frequency is a reliable noninvasive variable that can be used to predict defibrillation outcome (30). However, the improvements in outcomes of this strategy of CPR to restore activation rates in out-of-hospital arrest patients will need to be evaluated in ongoing clinical trials such as the ROC-Primed study (2).
VENTRICULAR FIBRILLATION IN MYOPATHIC HUMAN HEARTS

Limitation

The hearts were Langendorff perfused in a denervated environment. It is unknown if these factors influence activation patterns, although we did observe VF rates being lower than what normally is seen clinically. We explain this difference by the absence of autonomic regulation. During chest compression in humans under CPR situations, reperfusion is incomplete and may not reflect the degree of anterograde flow established in our model. In addition, we did not evaluate defibrillation efficacy or postdefibrillation mechanical activity as investigated by Berg et al. (4) in an animal model testing the reperfusion first followed by defibrillation strategy. Determining proper defibrillation thresholds will require multiple shocks and multiple inductions with the concern of change in fibrillation dynamics during the insults; thus, defibrillation threshold was not evaluated in our study. Also, for this reason, we limited the inductions to two per heart. Our study was not meant to simulate out-of-hospital cardiac arrest but to test the physiological concepts of ischemia reperfusion on human VF. Finally, caution should be exercised in extrapolating the model used in this study of myopathic human hearts treated with a variety of medication to the larger population of patients at risk of VF. Our study involved global fibrillation dynamics during ischemia and reperfusion in a whole heart; global histological analysis was not available for the hearts studied to determine the interaction of fibrillation with our findings.

In conclusion, global myocardial ischemia of 3 min during human VF decreases LAR and leads to an increased gradient in activation rate between the endocardium and epicardium with a concomitant increase in wavebreak on the epicardium but not on the endocardium. These spatiotemporal changes are reversed by a short duration of reperfusion.

GRANTS

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DISCLOSURES

No conflict of interest.

REFERENCES


