Sodium-Hydrogen Exchange Inhibition During Ventricular Fibrillation
Beneficial Effects on Ischemic Contracture, Action Potential Duration, Reperfusion Arrhythmias, Myocardial Function, and Resuscitability

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Background—Inhibition of the sarcolemmal sodium-hydrogen exchanger isoform-1 (NHE-1) is emerging as a promising novel strategy for ameliorating myocardial injury associated with ischemia and reperfusion. We investigated whether NHE-1 inhibition (with cariporide) could minimize mechanical and electrical myocardial abnormalities that develop during ventricular fibrillation (VF) and improve outcome using a porcine model of closed-chest resuscitation.

Methods and Results—Two groups of 8 pigs each were subjected to 8 minutes of untreated VF and randomized to receive either a 3-mg/kg bolus of cariporide or 0.9% NaCl immediately before an 8-minute interval of conventional closed-chest resuscitation. Cariporide prevented progressive increases in left ventricular free-wall thickness (from 1.0 ± 0.2 to 1.5 ± 0.3 cm with NaCl, P < 0.001 versus 0.9 ± 0.1 to 1.1 ± 0.3 cm with cariporide, P = NS), maintained the coronary perfusion pressure above resuscitability thresholds (10 ± 8 versus 19 ± 3 mm Hg before attempting defibrillation, P < 0.05), and increased resuscitability (2 of 8 versus 8 of 8, P < 0.005). In 2 additional groups of 4 pigs each subjected to a briefer interval of untreated VF, cariporide ameliorated postresuscitation shortening of the action potential duration (APD) at 30%, 60%, and 90% repolarization (ie, APD60 at 2 minutes after resuscitation; 75 ± 29 versus 226 ± 16 ms, P < 0.05), minimized postresuscitation ventricular ectopic activity preventing recurrent VF, and lessened postresuscitation myocardial dysfunction.

Conclusions—NHE-1 inhibition may represent a highly potent novel strategy for resuscitation from VF that can ameliorate myocardial manifestations of ischemic injury and improve the effectiveness and outcome of closed-chest resuscitation. (Circulation. 2003;107:1804-1809.)

Key Words: action potentials ▪ cardiopulmonary resuscitation ▪ defibrillation ▪ ischemia ▪ myocardium

Strategies to improve resuscitability from ventricular fibrillation (VF) have traditionally centered on means to enhance the delivery of oxygen and energy substrates while limiting injury related to the resuscitation process itself. Yet, the possibility that improved resuscitability could result from targeting specific pathogenic mechanisms activated during ischemia and reperfusion has been largely unexplored.

We have identified activation of the sarcolemmal sodium-hydrogen exchanger isoform-1 (NHE-1) as a potentially important pathogenic target and demonstrated, in rat models of VF and resuscitation, that NHE-1 inhibition can ameliorate myocardial abnormalities relevant to cardiac resuscitation. In these models, NHE-1 inhibition reduced ischemic contracture during VF (improving the hemodynamic efficacy of chest compression), minimized postresuscitation ventricular ectopic activity (preventing recurrent VF), and lessened postresuscitation myocardial dysfunction. Contemporary to our studies, Wirth et al reported, in a swine model of regional coronary occlusion, similar antiarrhythmic effects of NHE-1 inhibition associated with preservation of the action potential duration (APD).

In the present studies, using a clinically more relevant porcine model of VF, we investigated the capability of NHE-1 inhibition to ameliorate the aforementioned myocardial abnormalities and to facilitate closed-chest resuscitation. The effects on ischemic contracture were investigated by use...
of transesophageal echocardiography (TEE). The effects on APD and ventricular ectopic activity were investigated by use of a monophasic action potential (MAP) recording/pacing catheter.4,5

Methods
These studies were approved by our Research and Development Committee and conducted according to institutional guidelines.

Animal Preparation
Male domestic pigs (30 to 42 kg; Oak Hill Genetics, Ewing, Ill) were sedated with ketamine (30 mg/kg IM) and anesthetized with pentobarbital (30 mg/kg IV for induction and 8 mg/kg IV every 30 minutes for maintenance). Ventilation was provided through an orotracheal tube with a volume-controlled ventilator (Bear 1000, Bear Medical Systems, Inc) set to deliver a tidal volume of 10 mL/kg, peak flow of 40 L/min, and FiO2 of 0.4. The respiratory rate was adjusted to maintain an end-expired PCO2 between 35 and 45 mm Hg. Rectal temperature was maintained between 36.5°C and 37.5°C with a servocontrolled water-circulated blanket. A lead II ECG was recorded through skin electrodes, and 2 self-adhesive conductive gel pads were positioned on the chest for electrical defibrillation.

In an initial series, a 5F pacing electrode was advanced through the right cephalic vein into the pulmonary artery for measuring cardiac output (Edward Critical Care Explorer, Baxter Healthcare Corp) along with right atrial and pulmonary artery pressures. A 7F high-fidelity Micro-Tip pressure-transducer pigtail-catheter (model SPC-474A, Millar Instruments) was advanced from the right carotid artery into the left ventricle for pressure measurements. A 5.0-MHz biplane TEE probe connected to an echocardiography system (128XP/10, Acuson) was advanced into the midesophagus for measuring left ventricular cavity size and wall thickness.

In a subsequent series, a 7F MAP recording/pacing contact electrode (EP Technology) was advanced under fluoroscopy from the right femoral artery into the left ventricular cavity, and its tip was positioned against the anterolateral ventricular wall endocardium. Stable endocardial contact was verified by MAP morphology.5 The orientation with maximal leftward (<30°) and anterior (<45°) deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic deflections, provided an adequate long-axis view of the left-side chambers. Although in many instances, chest compression precluded diastolic defe...
1. The aggregate data (Figure 2) demonstrated significant differences between groups from the second minute of chest compression on. During this interval, the end-of-relaxation left ventricular pressures remained largely unchanged in NaCl- and cariporide-treated pigs (i.e., $5\pm 3$ and $5\pm 3$ mm Hg at 2 minutes and $6\pm 2$ and $7\pm 3$ mm Hg at 6 minutes of chest compression, $P=NS$).

During the initial 3 minutes of chest compression, the coronary perfusion pressure increased to $\approx 18$ mm Hg in both groups (Figure 2). Thereafter, the coronary perfusion pressure declined in control pigs despite maximal force of compression but remained stable in cariporide-treated pigs, attaining an almost 2-fold difference immediately before defibrillation was attempted ($10\pm 8$ versus $19\pm 3$ mm Hg, $P<0.05$).

All 8 cariporide-treated pigs were defibrillated successfully, with return of spontaneous circulation after $4.6\pm 2.8$ electrical shocks ($438\pm 453$ J) and had minimal postresuscitation ventricular ectopic activity, with no episodes of recurrent VF (Table 1). In contrast, 4 of the 8 NaCl-treated pigs failed the initial resuscitation attempt: 2 had refractory VF and 2 developed pulseless electrical activity. Spontaneous circulation was restored in the remaining 4 pigs after the initial defibrillation attempts, requiring $6.4\pm 3.1$ electrical shocks ($744\pm 522$ J; $P=NS$ versus cariporide). These pigs, however, developed intense ventricular ectopic activity, with frequent episodes of recurrent VF requiring additional electrical shocks (Table 1). Spontaneous circulation was restored in 2 of these 4 pigs after $9.5\pm 0.7$ additional electrical shocks ($950\pm 71$ J), whereas 2 failed the postresuscitation defibrillation attempts. Thus, all 8 cariporide- but only 2 NaCl-treated pigs were resuscitated successfully and survived the postresuscitation interval ($P=0.005$).

After resuscitation, cariporide-treated pigs had myocardial wall thickness comparable to baseline but decreased left ventricular ejection fraction, cardiac index, and mean aortic pressure that partially reversed during the ensuing postresuscitation interval. Surviving control pigs had prominent myocardial wall thickness and worse myocardial dysfunction (Figure 2 and Table 2).

Series 2
The shorter duration of untreated VF allowed successful resuscitation in each instance. Control pigs ($n=4$) again displayed intense postresuscitation ventricular ectopic activity, with frequent episodes of recurrent VF (Table 1) coincident with marked shortening of the APD and changes in morphology to a more triangular shape. These abnormalities subsided within 10 minutes. In contrast, pigs treated with cariporide ($n=4$) had their APD preserved with morphology similar to baseline (Figures 3 and 4). In addition, cariporide-treated pigs had significantly less postresuscitation ventricular

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**Figure 1.** Left ventricular wall thickness measured at end of mechanical diastole (baseline) and end of “compression diastole” at 2 and 8 minutes of chest compression (CC) increased by approximately 50% in an NaCl-treated (top frames) but not in a cariporide-treated (bottom frames) pig. Calibration marks are spaced at 1-cm intervals.

**Figure 2.** Coronary perfusion pressure (CPP) and anterolateral left ventricular (LV) wall thickness at baseline (BL), during VF, and after resuscitation in pigs randomized to receive before chest compression (drug) either NaCl (closed symbols, $n=8$) or cariporide (open symbols, $n=8$). Only 2 successfully resuscitated NaCl-treated pigs provided postresuscitation data. Mean $\pm$ SEM. *$P<0.05$, †$P<0.001$ vs cariporide by 1-way ANOVA.
ectopic activity and significantly less postresuscitation myocardial
dysfunction (Table 1, Figure 5).

**Discussion**

NHE-1 inhibition during VF (1) attenuated the development of ischemic contracture; (2) reduced postresuscitation ventricular
teortic activity in association with preservation of APD, pre-
venting episodes of recurrent VF; and (3) ameliorated postresusc-
tion myocardial dysfunction. These effects enhanced the
hemodynamic effectiveness of closed-chest resuscitation and
facilitated successful resuscitation and survival.

**Ischemic Contracture**

Ischemic contracture refers to the progressive myocardial
wall thickening with reductions in ventricular cavity that
results from severe ischemia. Onset of ischemic contracture
is associated with decreases in ATP to levels < 10% of normal. Ischemic contracture of varying severity has been
reported during cardiac arrest to compromise resuscitabil-
ity in animal models and in human victims of cardiac
arrest. In our studies, ischemic contracture developed in control pigs
during VF, but only during the interval of low-flow coronary
perfusion promoted by chest compression. These observations,
along with previous reports in isolated rat heart models of VF1,2
and isolated cardiac myocytes,10 suggests that reperefusion and
reoxygenation could play an important precipitating role. Ische-
mic contracture occurred with essentially no changes in end-of-
chest-relaxation left ventricular pressures, confirming reductions
in myocardial compliance.

**TABLE 1. Ventricular Ectopic Activity During the Initial 5 Minutes After Resuscitation**

<table>
<thead>
<tr>
<th></th>
<th>Singlets, n</th>
<th>Bigeminy, n</th>
<th>Salvos, n</th>
<th>Episodes of VT, n</th>
<th>Episodes of VF, n</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Series 1 (8 minutes untreated VF)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NaCl (n=4)*</td>
<td>34.8±25.1</td>
<td>17.5±26.8</td>
<td>5.5±6.5</td>
<td>0.0±0.0</td>
<td>4.0±2.9</td>
</tr>
<tr>
<td>Cariporide (n=8)</td>
<td>6.3±9.1†</td>
<td>0.0±0.0†</td>
<td>1.3±3.5</td>
<td>0.1±0.4</td>
<td>0.0±0.0†</td>
</tr>
<tr>
<td><strong>Series 2 (6 minutes untreated VF)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NaCl (n=4)</td>
<td>41.5±23.6</td>
<td>12.5±8.2</td>
<td>6.3±6.7</td>
<td>2.5±2.4</td>
<td>2.5±2.1</td>
</tr>
<tr>
<td>Cariporide (n=4)</td>
<td>5.3±9.2†</td>
<td>0.0±0.0†</td>
<td>0.0±0.0†</td>
<td>0.0±0.0†</td>
<td></td>
</tr>
</tbody>
</table>

VT indicates ventricular tachycardia.
*Only pigs that restored spontaneous circulation after the initial resuscitation attempt were included in this analysis;
‡P<0.05 vs baseline by repeated-measures ANOVA and Dunnett’s multicomparison test.

In our studies, ischemic contracture developed in control pigs
during VF, but only during the interval of low-flow coronary
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mic contracture occurred with essentially no changes in end-of-
chest-relaxation left ventricular pressures, confirming reductions
in myocardial compliance.

**TABLE 2. Myocardial Function (Series 1)**

<table>
<thead>
<tr>
<th>Baseline (−10 min)</th>
<th>Minutes After Resuscitation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>5</td>
</tr>
<tr>
<td>LV end-diastolic area, cm²</td>
<td>9.3±2.8</td>
</tr>
<tr>
<td>NaCl (n=8)*</td>
<td></td>
</tr>
<tr>
<td>Cariporide (n=8)</td>
<td>9.1±2.0</td>
</tr>
<tr>
<td>LV end-systolic area, cm²</td>
<td>3.7±1.1</td>
</tr>
<tr>
<td>NaCl (n=8)</td>
<td></td>
</tr>
<tr>
<td>Cariporide (n=8)</td>
<td>3.7±0.7</td>
</tr>
<tr>
<td>LV ejection fraction, %</td>
<td>60±6</td>
</tr>
<tr>
<td>NaCl (n=8)</td>
<td></td>
</tr>
<tr>
<td>Cariporide (n=8)</td>
<td>58±7</td>
</tr>
<tr>
<td>AoP (mean), mm Hg</td>
<td>107±15</td>
</tr>
<tr>
<td>NaCl (n=8)</td>
<td></td>
</tr>
<tr>
<td>Cariporide (n=8)</td>
<td>126±8</td>
</tr>
<tr>
<td>PAOP, mm Hg</td>
<td>5±1</td>
</tr>
<tr>
<td>NaCl (n=8)</td>
<td></td>
</tr>
<tr>
<td>Cariporide (n=8)</td>
<td>5±1</td>
</tr>
<tr>
<td>CI, L· min⁻¹·m⁻²⁻</td>
<td>6.3±1.1</td>
</tr>
<tr>
<td>NaCl (n=8)</td>
<td></td>
</tr>
<tr>
<td>Cariporide (n=8)</td>
<td>7.2±1.2</td>
</tr>
</tbody>
</table>

LV indicates left ventricular; AoP, aortic pressure; PAOP, pulmonary artery occlusive pressure; and CI, cardiac index.
*Only 2 successfully resuscitated NaCl-treated pigs provided postresuscitation data.
†P<0.05 vs baseline by repeated-measures ANOVA and Dunnett’s multicomparison test.
‡P<0.05, §P<0.01 vs NaCl by 1-way ANOVA.
NHE-1 inhibition using the potent and selective inhibitor cariporide prevented ischemic contracture and enabled chest compression to maintain a coronary perfusion pressure above resuscitability thresholds. In a limited number of experiments in which the left ventricle was well visualized during chest compression, cariporide attenuated reductions in left ventricular cavity size. Thus, preservation of end-of-chest-relaxation ventricular volumes with enhanced forward blood flow probably explained these favorable effects of NHE-1 inhibition. In our second series, statistically insignificant higher coronary perfusion pressures also favored cariporide (16 ± 2 mm Hg before defibrillation was attempted). The longer duration of untreated VF most likely explained greater ease of maintaining coronary perfusion pressures above resuscitability thresholds in control pigs.

### Postresuscitation Ventricular Arrhythmias and APD

Electrical instability with recurrent VF commonly occurs after cardiac resuscitation and may contribute to the nearly 30% incidence of early postresuscitation deaths. Ventricular arrhythmias probably share mechanisms common to those present during reperfusion after coronary occlusion. In this setting, prominent repolarization abnormalities occur that are characterized by shortening of the APD and change to a more triangular shape, decreased amplitude, APD alternans, and afterdepolarizations. APD shortening is partly explained by opening of sarcolemmal K$_{ATP}$ channels. However, recent evidence also implicates NHE-1 activation.

Prominent APD shortening occurred in control pigs during the interval of maximal postresuscitation ventricular ectopic activity. These abnormalities were similar but quantitatively more prominent than those reported by Wirth and coworkers after regional ischemia in pigs. NHE-1 inhibition was remarkably effective in limiting APD shortening, reducing postresuscitation ventricular dysrhythmias, and preventing recurrent VF and arrhythmic deaths.

### Postresuscitation Myocardial Dysfunction

Reversible diastolic and systolic dysfunction of varying severity occurs after cardiac resuscitation and may contribute to the

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**Figure 3.** Representative endocardial MAP recordings at baseline (BL) and initial 15 minutes postresuscitation in an NaCl-treated and a cariporide-treated pig. Cariporide prevented changes in MAP duration and morphology and secured a more stable rhythm.

**Figure 4.** APD measured at 30%, 60%, and 90% repolarization along with cycle length at baseline (-10 minutes) and after resuscitation in pigs randomized immediately before chest compression to NaCl (closed symbols, n=4) or cariporide (open symbols, n=4). Mean ± SEM. *P<0.05, †P<0.01, ‡P<0.001 vs NaCl by 1-way ANOVA.

**Figure 5.** Postresuscitation (PR) left ventricular function in pigs randomized to NaCl (closed bars, n=4) or cariporide (open bars, n=4) in series 2. Mean ± SEM. *P<0.05 vs baseline (BL) by repeated-measures ANOVA; †P<0.05 vs cariporide by 1-way ANOVA.
Mechanisms of Protective Action

Excellent reviews have been published on the pathogenic mechanisms of NHE-1 activation and the protective actions of NHE-1 inhibition. Yet, the application of these concepts to the cardiac arrest setting is novel. The intense intracellular acidosis that develops during VF after cessation of coronary blood flow is believed to activate the sarcolemmal NHE-1, leading to a proton-driven sarcolemmal Na\(^+\) influx with progressive cytosolic Na\(^+\) accumulation as the Na\(^+\)-K\(^+\) pump fails to extrude Na\(^+\) during ischemia. Cytosolic Na\(^+\) overload becomes a “substrate” for ischemia and reperfusion injury. During closed-chest resuscitation, the coronary blood flow rarely exceeds 20% of normal, failing to reverse ischemia but allowing normoacidic blood to perfuse the coronary circuit. Unremitting ischemia with a large transsarcolemmal proton gradient creates optimal conditions for NHE-1 to remain active throughout chest compression and probably the early minutes after return of spontaneous circulation.

NHE-1 inhibition reduces sarcolemmal Na\(^+\) entry and the detrimental downstream effects on cell physiology. Less cytosolic Na\(^+\) overload is thought to spare ATP use by the Na\(^+\)-K\(^+\) pump, retarding its intracellular depletion and the consequent ischemic contracture. Reduced cytosolic Na\(^+\) overload may also attenuate mitochondrial injury by limiting Na\(^+\)-induced membrane depolarization, mitochondrial swelling, cytochrome c release and by preserving oxidative phosphorylation. Less cytosolic Na\(^+\) overload could also attenuate Na\(^+\)-induced reverse-mode operation of the Na\(^+\)-Ca\(^2+\) exchanger and the ensuing outward (repolarizing) current, presumably responsible for APD shortening. Less cytosolic Ca\(^2+\) overload could also favor postresuscitation electrical stability and myocardial function.

Clinical Implications

The effects of NHE-1 inhibition reported here are similar to those previously reported in isolated heart and intact rat models of VF. In addition, comparable myocardial protective actions have been reported in settings of regional and global myocardial ischemia in nonfibrillating hearts from different species. Sarcolemmal NHE-1 is expressed in human myocardium, and clinical trials have demonstrated myocardial benefits derived from NHE-1 inhibition in subsets of patients undergoing emergent coronary revascularization. Thus, effects similar to those reported here in pigs may also apply to humans and facilitate closed-chest resuscitation from VF. Clinical studies on NHE-1 inhibition during cardiac resuscitation are eagerly awaited.

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References


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