Pyruvate stabilizes electrocardiographic and hemodynamic function in pigs recovering from cardiac arrest

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Abstract

Cardiac electromechanical dysfunction may compromise recovery of patients who are initially resuscitated from cardiac arrest, and effective treatments remain elusive. Pyruvate, a natural intermediary metabolite, energy substrate, and antioxidant, has been found to protect the heart from ischemia-reperfusion injury. This study tested the hypothesis that pyruvate-enriched resuscitation restores hemodynamic, metabolic, and electrolyte homeostasis following cardiac arrest. Forty-two Yorkshire swine underwent pacing-induced ventricular fibrillation and, after 6 min pre-intervention arrest, 4 min precordial compressions followed by trans-thoracic countershocks. After defibrillation and recovery of spontaneous circulation, the pigs were monitored for another 4 h. Sodium pyruvate or NaCl were infused i.v. ($0.1 \text{ mmol} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$) throughout precordial compressions and the first 60 min recovery. In 8 of the 24 NaCl-infused swine, the first countershock converted ventricular fibrillation to pulseless electrical activity unresponsive to subsequent countershocks, but only 1 of 18 pyruvate-treated swine developed pulseless electrical activity (relative risk 0.17; 95% confidence interval 0.13–0.22). Pyruvate treatment also lowered the dosage of vasoconstrictor phenylephrine required to maintain systemic arterial pressure at 15–60 min recovery, hastened clearance of excess glucose, elevated arterial bicarbonate, and raised arterial pH; these statistically significant effects persisted up to 3 h after sodium pyruvate infusion, while infusion-induced hypernatremia subsided. These results demonstrate that pyruvate-enriched resuscitation achieves electrocardiographic and hemodynamic stability in swine during the initial recovery from cardiac arrest. Such metabolically based treatment may offer an effective strategy to support cardiac electromechanical recovery immediately after cardiac arrest.

Keywords: acidemia, bicarbonate, cardiopulmonary resuscitation, phenylephrine, pulseless electrical activity, ventricular fibrillation

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Introduction

Although over 50 years have passed since the inception of closed chest cardiac massage, and despite many refinements of this approach in the intervening decades, cardiac arrest remains a leading cause of death and persistent disability in the developed world, and effective treatments are elusive.¹ Each year, about 360,000 Americans suffer out-of-hospital cardiac arrest.² Many victims are resuscitated, but later succumb to extensive ischemia-reperfusion injury to their internal organs.^{3–6} Only ~10% of emergency responder-treated out-of-hospital cardiac arrest victims survive to hospital discharge.²

A first priority following recovery of spontaneous circulation (ROSC) is to stabilize metabolic, electrolyte, and hemodynamic variables to prevent re-fibrillation or irreversible cardiogenic shock.⁷ In particular, measurements of blood pH, circulating concentrations of lactate, bicarbonate, sodium, potassium and glucose, and calculated base excess are crucial to evaluate the status of postarrest patients. Following cardiac arrest, blood pH falls sharply due to elevated lactate from anaerobic glycolysis and hypercapnia due to inadequate alveolar ventilation. Hypercapnia and respiratory acidosis can be ameliorated by mechanically ventilating patients, but resolving lactate-dependent metabolic acidosis requires careful monitoring of arterial

blood gas chemistry.⁷ Post-ROSC management of electrolytes, especially potassium, is essential to prevent electrical disturbances that could re-arrest the heart.⁸ Finally, unless hypoglycemia is the cause of cardiac arrest, hyperglycemia is typical in resuscitated patients and, if untreated, may contribute to long-term neurological impairment.^{7,9,10}

The intrinsic properties of the natural intermediary metabolite pyruvate make it an especially promising intervention for myocardial ischemic syndromes. Pyruvate is highly water soluble, avidly transported into cells and mitochondria by respective sarcolemmal and inner mitochondrial membrane monocarboxylate transporters, a readily oxidized, energy-yielding metabolic fuel, and an antioxidant capable of detoxifying a host of reactive oxygen and nitrogen intermediates.^{11,12} Thus, pyruvate augmented ATP phosphorylation potential, a measure of Gibbs free energy of ATP hydrolysis, and the redox state of the major cellular antioxidant glutathione in postischemic perfused guinea-pig hearts,^{13–15} myocardium of pigs undergoing cardiopulmonary bypass,^{16,17} and myocardium of goats receiving fluid resuscitation for hypovolemia.¹⁸ In a canine cardiac arrest model, intravenous administration of sodium pyruvate during cardiac arrest, open chest cardiac compression, and the first 25 min of ROSC improved postarrest cardiac electromechanical and metabolic recovery.¹⁹ Pyruvate enhanced left ventricular contractility and lusitropy to increase systemic arterial pressure during early ROSC and hastened electrocardiographic recovery evidenced by resolution of ST segment displacement when compared to NaCl-infused control experiments.¹⁹ An important limitation of this study is that open-chest cardiac massage and direct epicardial defibrillatory countershocks were employed, not conventional precordial compressions and transthoracic countershocks. Therefore, we tested the hypothesis that intravenous administration of sodium pyruvate during precordial compressions and the first 60 min ROSC restores hemodynamic, metabolic, and electrolyte homeostasis in a closed-chest porcine model of cardiac arrest.

Methods

Surgical preparation and instrumentation

All surgical and experimental procedures in pigs were conducted in accordance with the Guide to the Care and Use of Laboratory Animals²⁰ and the ARRIVE guidelines²¹ and were approved by the Institutional Animal Care and Use Committee of the University of North Texas Health Science Center (protocol #2012/13-29-A10). After an overnight fast, Yorkshire swine (30 ± 0.7 kg; 23 males, 19 females) were sedated with a telazol (6.7 mg/kg i.m.)/xylazine (1.3 mg/kg i.m.) cocktail and intubated, and a surgical plane of anesthesia was maintained by mechanical ventilation (tidal volume 12 – 15 ml·kg⁻¹; 12 – 14 cycles/min) with 1–3% isoflurane in 100% O₂. Epidermal electrodes were placed for standard limb lead II electrocardiography. The left femoral artery and vein were exposed by inguinal incision and blunt dissection. A 7-Fr polyurethane catheter was inserted into the femoral artery and advanced into the abdominal aorta to monitor systemic arterial blood pressure and

sample arterial blood. Another catheter was inserted into the femoral vein for infusing medications and 0.9% NaCl. The right external jugular vein was accessed via a 3-cm cervical incision, and a catheter was installed for insertion of a pacing wire and for infusion of resuscitative treatments. In a subset of nine experiments, a second catheter was inserted into the right external jugular vein and advanced into the jugular bulb. Simultaneous measurements of jugular bulb and aortic pressures permitted assessment of cerebral perfusion pressure, i.e., mean aortic pressure minus jugular bulb pressure, during chest compressions.

Cardiac arrest

A pacing wire was inserted into the jugular catheter and advanced into the right ventricle until the tip contacted the endocardium. To arrest the heart, a 0.5–1 s burst of 60 Hz electrical impulses was transmitted to the right ventricle via the pacing wire, causing ventricular fibrillation. Once ventricular fibrillation was confirmed by the characteristic electrocardiogram, monophasic decline in aortic pressure and absence of an arterial pulse,²² mechanical ventilation was suspended for the duration of cardiac arrest and cardiopulmonary resuscitation (CPR). Bicarbonate (0.3 – 0.4 mEq/kg) was administered i.v. at 5 min arrest to partially offset the acidemia that developed during cardiac arrest.^{23,24}

Cardiopulmonary resuscitation

After 6 min of cardiac arrest, mid-sternal chest compressions were applied manually, at a force sufficient to compress the chest by one-fourth of its antero-posterior diameter, followed by release and recoil, at a rate of 100 cycles/min, in accordance with current American Heart Association guidelines.²⁵ The person administering the compressions was blinded to intravenous treatments, arterial pressure, end-tidal PCO₂, and blood gas chemistry. At 7 min arrest (i.e., 1 min CPR), vasopressin (10 U) was injected into the right jugular vein to increase the efficacy of the chest compressions.²⁶ Vasopressin-enhanced CPR achieved a cerebral perfusion pressure of 69 ± 5 mmHg. Mechanical ventilation remained suspended throughout CPR, in accordance with extensive clinical evidence that assisted ventilation (rescue breathing) during CPR following witnessed cardiac arrest offers scant neurological or survival benefit.^{27–29}

Resuscitative solutions were prepared within 30 min of use. Crystalline sodium pyruvate or NaCl of the highest commercially available purity (Sigma, St. Louis, MO) were dissolved in double-distilled H₂O to 2 M concentration, sterilized by passage through 0.22 µm membranes (Millipore Stericup® vacuum-driven filtration system) and stored on ice in sterile containers prior to infusion. To ensure that experiments were conducted in a blinded fashion, all resuscitative solutions were prepared in a separate room by an investigator not directly participating in the experiment who was unaware of each animal's condition and gender. Treatments were assigned by the sealed envelope system. Intravenous infusion (0.1 mmol·kg⁻¹·min⁻¹) of NaCl or sodium pyruvate was initiated at the onset of CPR and maintained until 60 min ROSC. Sodium pyruvate infusion

was calibrated to increase plasma pyruvate concentrations to 4–5 mM, which approximated the maximally effective concentration range for enhancing cardiac metabolism and performance defined in previous investigations.^{13,16} The investigators conducting the experiments and analyses remained blinded to the treatment until the study was completed. The sample sizes of the NaCl- and sodium pyruvate-treated groups were adjusted at the midpoint of the study to ensure similar numbers of survivors for statistical comparison of postarrest variables.

Seven sham control pigs (4 males, 3 females) were instrumented, anesthetized, and mechanically ventilated but not subjected to cardiac arrest, CPR, or defibrillation. The sham protocol was the same duration as the cardiac arrest-resuscitation protocol. These pigs received intravenous NaCl infusion for a period corresponding to that of treatment infusions in the cardiac arrest experiments.

Defibrillation and postresuscitation management

After 4 min CPR, transthoracic direct current countershocks were administered with external paddles (LifePack 12; Physio-Control) to restore cardiac rhythm. Up to three 6–7 J/kg countershocks were applied, followed by up to three 8–12 J/kg countershocks, at 30 s intervals with intervening CPR, until spontaneous cardiac rhythm was restored. These efforts were discontinued and the experiment terminated if cardioversion was not achieved within six countershocks (9 of 42 cardiac arrests).

Mechanical ventilation was resumed immediately after spontaneous cardiac rhythm was confirmed by a rapid increase in mean arterial pressure (MAP) and a discernible electrocardiographic rhythm. Next, lidocaine (20 mg in 1 ml 0.9% NaCl) was administered via the jugular catheter as a prophylactic measure to minimize recurrence of ventricular tachycardia or ventricular fibrillation and to hasten recovery of a normal QRS waveform complex. Mean aortic pressure ≥ 60 mmHg and a sustained spontaneous cardiac electrical rhythm were taken to indicate ROSC. A surgical plane of anesthesia was maintained and aortic pressure, end-tidal PCO_2 , and electrocardiogram were monitored throughout ROSC. Rate of ventilation was adjusted throughout the protocol to achieve an arterial PCO_2 of 35–45 mmHg. Beginning at approximately 15 min ROSC, the α -adrenergic vasoconstrictor phenylephrine was infused into the left femoral vein at a rate sufficient to maintain aortic pressure at 65–75 mmHg. The phenylephrine infusion was tapered and ultimately discontinued by approximately 60 min ROSC as cardiac function recovered. Upon completing the protocol, the pig was humanely euthanized by arresting the heart with electrical current applied via a pacing wire, in accordance with the recommendations of the American Veterinary Medical Association.³⁰

Analysis of arterial blood

Blood samples (5 ml) were withdrawn from the left femoral artery into heparinized sterile syringes 15 and 5 min before arrest, and at 5, 15, 30, 60, 120, 180, and 240 min ROSC. These samples were centrifuged at $10,000 \times g_{\text{max}}$ for 7.5 min

to sediment the formed elements, and then the plasma supernatant was flash-frozen in liquid nitrogen. Plasma samples were extracted as previously described,³¹ and pyruvate concentration was determined colorimetrically³² in a Shimadzu (Columbia, MD) model UV-1800 dual wavelength UV-vis spectrophotometer (337 nm measuring wavelength, 417 nm reference wavelength, $\epsilon = 5.65 \text{ M}^{-1} \cdot \text{cm}^{-1}$). Additional blood samples (1 mL) were collected for measurements of sodium, potassium, lactate, bicarbonate, and glucose concentrations, pH, base excess, and hematocrit in an Instrumentation Laboratory (Bedford, MA) Gem Premier 3000 blood gas analyzer.

Statistical methods

Hemodynamic function and plasma chemistry values in pyruvate-treated, NaCl-treated, and sham pigs were compared by two-factor (time, treatment) analyses of variance with repeated measures on time and Student-Newman-Keuls *post-hoc* analyses. Phenylephrine dosages and energy required for defibrillation were compared between pyruvate- and NaCl-treated pigs by Student's *t*-test. Results are reported as mean values $\pm$ SEM. *P* values < 0.05 were taken to indicate statistically significant treatment effects. The incidences of pulseless electrical activity (PEA) in pyruvate- vs. NaCl-treated pigs were compared by computing relative risk and 95% confidence intervals.

Results

CPR and pyruvate-enhanced cardioversion to productive rhythm

Before cardiac arrest, MAP was 85 ± 3 mmHg in the pigs destined to receive sodium pyruvate and 82 ± 2 mmHg in those which subsequently received NaCl ($P = 0.72$; Figure 1). MAP values achieved by precordial compressions did not differ significantly between the two treatments; at 4 min CPR, MAP was 78 ± 7 and 67 ± 5 mmHg in pyruvate- and NaCl-treated pigs, respectively ($P = 0.19$; Figure 1). In 8 of 24 NaCl-treated swine, the first countershock converted ventricular fibrillation to PEA which proved unresponsive to subsequent countershocks, but only 1 of 18 pyruvate-treated swine was converted to PEA (relative risk 0.17; 95% confidence interval 0.13, 0.22). Despite the decreased incidence of PEA, intravenous pyruvate did not produce a statistically significant effect on the number of countershocks required to achieve ROSC (NaCl 2.1 ± 0.4 ; pyruvate: 1.4 ± 0.2 ; $P = 0.18$) or the electrical energy required to restore spontaneous rhythm (NaCl $14.8 \pm 2.9 \text{ J} \cdot \text{kg}^{-1}$; pyruvate: $8.3 \pm 1.3 \text{ J} \cdot \text{kg}^{-1}$; $P = 0.20$).

Hemodynamic function: pyruvate lowered postarrest vasoconstrictor requirements

After defibrillation MAP abruptly yet temporarily increased in both CPR groups vs. sham pigs (Figure 2(a)). At 5 min ROSC, MAP in pyruvate-treated pigs (134 ± 8 mmHg) was higher ($P = 0.03$) than that of NaCl-infused pigs (112 ± 16 mmHg), but by 15 min, MAP had returned to baseline in both groups and remained stable thereafter ($P > 0.05$). Heart rate (HR; Figure 2(b)) was

Figure 1 Vasopressin enhances systemic arterial pressure produced by pre-cordial compressions. The peripheral vasoconstrictor vasopressin was administered at 1 min CPR (vertical dashed line). Mean values $\pm$ SEM from 24 NaCl- and 18 sodium pyruvate-infused pigs. BL: pre-arrest baseline; CPR: cardiopulmonary resuscitation

increased with NaCl ($138 \pm 13 \text{ min}^{-1}$) and pyruvate ($149 \pm 9 \text{ min}^{-1}$) infusion at 5 min ROSC ($P = 0.026$ and 0.016 , respectively, *vs.* sham), then gradually returned to baseline by 4 h ROSC. The product of HR-MAP (Figure 2(c)), a measure of myocardial contractile performance and ATP demand,³³ increased following defibrillation with both treatments *vs.* sham, yet to a greater extent with pyruvate *vs.* NaCl treatment ($P = 0.01$), and returned to baseline by 15 min ROSC. The pyruvate-treated pigs required less phenylephrine (Figure 2(d)) to maintain MAP at 65–75 mmHg ($0.04 \pm 0.01 \text{ mg} \cdot \text{kg}^{-1}$) than did the NaCl-treated pigs ($0.14 \pm 0.05 \text{ mg} \cdot \text{kg}^{-1}$; $P = 0.006$). Collectively, these results indicate that pyruvate improved contractile performance of the defibrillated heart during the critical first several minutes of recovery from cardiac arrest.

Pyruvate prevented postarrest arterial redox derangements and minimized hyperglycemia

Sodium pyruvate infusion achieved a stable arterial pyruvate concentration of 4–5 mM (Figure 3(a)). The excess pyruvate cleared within 60 min after infusion. Arterial lactate concentration (Figure 3(b)) was sharply increased ($P < 0.01$) at 5 min ROSC in both pyruvate-treated ($8.5 \pm 0.6 \text{ mM}$) and NaCl-treated ($7.3 \pm 0.3 \text{ mM}$) *vs.* sham pigs ($0.2 \pm 0.1 \text{ mM}$). Arterial lactate remained elevated until pyruvate infusion was discontinued, and then fell sharply, while lactate concentration gradually subsided over 4 h ROSC in NaCl-treated pigs (Figure 3(b)). In NaCl-treated pigs, the arterial lactate/pyruvate concentration ratio increased from 4 ± 1 pre-arrest to 24 ± 6 at 15 min ROSC ($P < 0.001$ *vs.* sham and pyruvate-treated pigs) and remained elevated until 4 h ROSC (Figure 3(c)).

Pyruvate infusion decreased the lactate/pyruvate ratio below that of NaCl-infused and sham pigs. After pyruvate infusion, the ratio increased to the already declining value of the NaCl-infused pigs.

Arterial glucose concentration increased sharply after cardiac arrest ($P < 0.001$ *vs.* sham) and remained elevated above the sham value, especially in the NaCl-infused pigs, until treatments were discontinued (Figure 3(d)). Glucose concentration remained higher in NaCl- *vs.* pyruvate-treated pigs at 15 min⁻¹ h ROSC ($P < 0.03$) before subsiding to the sham range by 4 h ROSC.

Transient hypernatremia and improved potassium management with pyruvate treatment

Arterial sodium concentration (Figure 4(a)) increased during the first 30–60 min of ROSC, i.e., during NaCl or sodium pyruvate infusion. A similar change was seen in NaCl-infused sham pigs. Arterial potassium concentration (Figure 4(b)) was elevated in both pyruvate- and NaCl-treated *vs.* sham pigs ($P < 0.001$) at 5 min ROSC, but by 15 min, pyruvate-treated pigs had lower potassium concentrations than NaCl-treated and sham pigs ($P = 0.002$), and this effect persisted until 4 h ROSC. An appreciable yet temporary increase in hematocrit (Figure 4(c)) was observed at 5–15 min ROSC in both NaCl- and pyruvate-treated pigs ($P < 0.001$ *vs.* baseline).

Pyruvate hastened resolution of postarrest arterial acidemia

Arterial blood pH (Figure 5(a)) fell by ca. 0.25 in both pyruvate- and NaCl-treated pigs at 5 min ROSC *vs.* the respective pre-arrest values. Arterial pH returned to baseline by 1 h ROSC in pyruvate-treated pigs, but recovered more gradually in NaCl-treated pigs. Arterial pH continued to increase after pyruvate infusion and plateaued at 2–3 h ROSC ($P = 0.003$ *vs.* NaCl-treated pigs). Interruption in pulmonary function during cardiac arrest produced marked hypercapnia in both pyruvate- and NaCl-treated pigs, which resolved in both groups by 2 h ROSC (Figure 5(b)). Arterial bicarbonate concentration (Figure 5(c)) fell in both pyruvate- and NaCl-treated pigs *vs.* the respective pre-arrest baselines ($P = 0.01$ and < 0.001) and contemporaneous sham values ($P = 0.009$ and 0.001). Arterial base excess (Figure 5(d)) paralleled arterial pH; thus, both pyruvate- and NaCl-treated pigs had base deficits *vs.* shams ($P < 0.001$) at 5–30 min ROSC. Pyruvate treatment restored arterial bicarbonate to baseline by 30 min ROSC, and bicarbonate concentration was persistently elevated throughout the 3 h postinfusion. In NaCl-treated pigs, arterial bicarbonate did not return to baseline and sham values until 3 h ROSC. Thus, cardiac arrest produced a combined respiratory/metabolic acidosis. Although the respiratory component subsided by 60 min ROSC, the metabolic acidosis resolved more gradually in the NaCl group. In contrast, pyruvate treatment hastened resolution of the metabolic acidosis and this effect persisted and even increased after pyruvate infusion was terminated.

Figure 2 Pyruvate decreased vasoconstrictor requirements to maintain arterial pressure during postarrest recovery. (a) Mean arterial pressure (MAP), (b) heart rate (HR), (c) heart rate-mean arterial pressure product (RPP), and (d) total phenylephrine dosage to maintain MAP at 60–75 mm Hg during recovery. On this figure and Figures 3–5, the horizontal bar denotes the period of treatment infusion. Values are mean $\pm$ SEM from 12 NaCl-treated and 11 pyruvate-treated cardiac arrest and 7 sham experiments. * $P < 0.05$ vs. sham; † $P < 0.05$ vs. CPR + NaCl. ROSC: recovery of spontaneous circulation

Figure 3 Post-arrest lactatemia, plasma redox state, and glucose concentration. (a) Pyruvate concentration, (b) lactate concentration, (c) lactate/pyruvate concentration ratio, and (d) glucose concentration. Mean values $\pm$ SEM from 12 NaCl-treated and 11 pyruvate-treated cardiac arrest and 7 sham experiments. * $P < 0.05$ vs. sham; † $P < 0.05$ vs. CPR + NaCl. ROSC: recovery of spontaneous circulation

Discussion

This study tested the ability of intravenous pyruvate treatment to resolve cardiac electromechanical dysfunction and systemic electrolyte and acid-base derangements in a closed chest porcine model of cardiac arrest and CPR. Administration of sodium pyruvate during CPR and the first 60 min ROSC sharply lowered the conversion of ventricular fibrillation to defibrillation-refractory PEA and decreased phenylephrine dosages necessary to maintain hemodynamic stability during ROSC. Pyruvate hastened postarrest clearance of glucose, a prognostic indicator of survival in human cardiac arrest victims,⁷ and lowered the arterial lactate/pyruvate concentration ratio, an effect known to minimize reactive oxygen species formation and foster postischemic recovery of cardiac function.³⁴ By increasing arterial pH, pyruvate treatment hastened recovery from metabolic acidosis imposed by cardiac arrest. Finally, pyruvate lowered plasma K^+ concentration, which may avoid pro-arrhythmic hyperkalemia and re-arrest of the heart. Thus, intravenous pyruvate infusion during cardiac arrest-resuscitation hastened restitution of electrocardiographic, hemodynamic, and metabolic function in swine recovering from cardiac arrest.

Pulseless electrical activity

Pulseless electrical activity is a cardiac electrical rhythm that does not produce ventricular contraction or forward movement of blood, and additional countershocks often fail to restore effective cardiac contractions. It is estimated that approximately 60% of out-of-hospital resuscitation attempts result in the development of PEA.³⁵ Survival of cardiac arrest victims with PEA is much lower than those

presenting with ventricular fibrillation; indeed, only 2–5% of patients who present with PEA as their initial rhythm survive,^{36–38} while 32% of cardiac arrest victims presenting with ventricular fibrillation survive.² Even fewer patients in whom ventricular fibrillation converted to PEA following countershocks survive to hospital discharge.³⁹ In this study, none of the nine pigs that developed PEA after the first countershock could be restored to spontaneous circulation. On the other hand, pyruvate treatment sharply lowered the incidence of PEA *vs.* NaCl treatment.

A putative mechanism underlying development of PEA following cardiac arrest and attempted defibrillation is depletion of myocardial ATP and phosphocreatine.^{35,40} Ventricular fibrillation is energetically costly, and the likelihood of postcountershock PEA increases as myocardial ATP reserves are expended. Thus, the increased rate of cardioversion to a productive electrocardiographic rhythm in pyruvate- *vs.* NaCl-treated pigs may be attributed to pyruvate's previously reported enhancement of myocardial energy state during CPR.¹⁹ By increasing phosphocreatine phosphorylation potential and thereby fostering recovery of myocardial energy reserves, pyruvate would increase energy supply for cardiac electromechanical recovery.

Postarrest hemodynamic recovery

Pyruvate treatment produced a temporarily higher MAP than did NaCl, which could possibly be ascribed to increased energy supply for cardiac work afforded by mitochondrial ATP production.^{13,41} In addition, pyruvate has been shown to potentiate the inotropic effects of catecholamines in postischemic myocardium,¹⁴ and this interaction may be particularly important after cardiac arrest when circulating catecholamine activity is likely elevated.⁴²

Figure 4 Arterial plasma electrolytes and hematocrit. (a) arterial Na^+ concentration, (b) arterial K^+ concentration, and (c) hematocrit. Mean values $\pm$ SEM from 12 NaCl-treated and 11 pyruvate-treated cardiac arrest and 7 sham experiments. * $P < 0.05$ vs. sham; † $P < 0.05$ vs. CPR + NaCl. ROSC: recovery of spontaneous circulation

Collectively, these actions of pyruvate lowered the requirement for vasoconstrictor phenylephrine to maintain MAP after cardiac arrest.

Plasma carbohydrates

Pyruvate infusion achieved circulating pyruvate concentrations of 4–5 mM, a range which was efficacious and

nontoxic in dogs recovering from cardiac arrest^{19,43} and in ischemically stunned guinea-pig hearts.^{13,14} Pyruvate is a readily oxidized mitochondrial fuel and is avidly transported across cellular and inner mitochondrial membranes by cotransport with H^+ (Figure 6).^{44,45} Accordingly, pyruvate quickly cleared from the circulation after infusion was discontinued. More rapid clearance of glucose during the first hour of recovery in pyruvate- vs. NaCl-infused pigs

Figure 5 Pyruvate hastened resolution of postarrest acidemia. (a) arterial pH, (b) PCO₂, (c) bicarbonate concentration, and (d) arterial base excess. Mean values $\pm$ SEM from 12 NaCl-treated and 11 pyruvate-treated cardiac arrest and 7 sham experiments. * P < 0.05 vs. sham; † P < 0.05 vs. CPR + NaCl. ROSC: recovery of spontaneous circulation

Figure 6 Proposed mechanisms of pyruvate-enhanced electrolyte homeostasis and acidemia resolution. Plasmalemmal monocarboxylate transporters (MCT) co-import pyruvate and H⁺ which lowers extracellular H⁺ concentration. The resultant transmembrane H⁺ gradient augments K⁺ import by the H⁺/K⁺ antiporter. Pyruvate oxidation in the TCA cycle supports oxidative phosphorylation (OxPhos), generating ATP to power K⁺ import by Na⁺/K⁺ ATPase

suggests enhanced recovery of glucose metabolism with pyruvate treatment, especially since the enhanced glucose clearance occurred despite infusion of a competing metabolic fuel, pyruvate.¹⁰

Elevated arterial lactate is considered clinically adverse,⁷ but that produced by pyruvate treatment via the lactate dehydrogenase equilibrium is an exception. Pyruvate infusion increased lactate concentration above that observed in the NaCl-infused pigs, a natural consequence of the chemical equilibrium effected by lactate dehydrogenase. After pyruvate infusion, clearance of lactate from the circulation paralleled the rapid decline in circulating pyruvate.

The elevated lactate concentration after cardiac arrest in the NaCl group exemplifies ischemia-induced lactic acidosis. Elevated arterial lactate/pyruvate concentration ratios are associated with increased formation of reactive oxygen intermediates.³⁴ In contrast, pyruvate infusion sharply lowered arterial lactate/pyruvate ratio in the face of postarrest metabolic stress.

Postarrest hypernatremia

Infusion of 2 M sodium, either as pyruvate or chloride salt, produced hypernatremia which gradually subsided over the 3 h following infusion. The treatments imposed a sodium load totaling 6 mmol sodium·kg⁻¹. If the ca. 30 kg pigs of this study contain ca. 20 L total body water, the 200 mmol infused Na⁺ load would produce ca. 10 mEq/L increase in plasma Na⁺ after osmotic equilibration, which approximates the transitory hypernatremia which plateaued at 30–60 min infusion. Importantly, this hypernatremia subsided within 3 h after sodium pyruvate or NaCl administration; thus, sodium pyruvate treatment did not impose persistent hypernatremia.

Hematocrit

Cardiac arrest provoked an appreciable, albeit transitory, increase in hematocrit of approximately 0.06–0.08. Hyperadrenergic conditions provoke contraction of the porcine spleen, increasing the amount of circulating erythrocytes to augment oxygen delivery to tissues.⁴⁶ In this study, cardiac arrest-resuscitation also temporarily raised the hematocrit. The subsequent decline in hematocrit during ROSC likely reflected hemodilution due to osmotic expansion of extracellular fluid volume secondary to sodium infusion, and also occurred during NaCl infusion in the

nonarrested sham pigs. Pyruvate did not modulate the increase in hematocrit induced by cardiac arrest.

Acid-base responses to pyruvate infusion

Elevated pH and base excess in pyruvate- vs. NaCl-treated pigs may be explained by pyruvate-mediated electrolyte shifts (Figure 6). Pyruvate enters cells via high-capacity monocarboxylate transporters^{44,45} which co-transport pyruvate with hydrogen ions. By lowering the extracellular H^+ concentration and generating a more favorable H^+ gradient for the exchange of extracellular K^+ for intracellular H^+ via the H^+/K^+ antiporter, pyruvate uptake could guard against development of postarrest hyperkalemia. The pyruvate-driven shift of H^+ into cells would shift the extracellular carbonic anhydrase equilibrium in favor of H^+ and HCO_3^- formation, thereby increasing extracellular HCO_3^- concentration. Moreover, pyruvate has been shown to preserve function of the Na^+ , K^+ ATPase, which, by transporting K^+ into cells contributes to the decreases in extracellular K^+ concentration (Figure 6).⁴⁷ Catecholamines, which are elevated after cardiac arrest,⁴² activate K^+ uptake in skeletal myocytes⁴⁸ and myocardium^{49,50} via Na^+ , K^+ ATPase activation. Pyruvate potentiated β -adrenergic inotropic effects in postischemic myocardium.^{14,15} A similar potentiation may have contributed to the decreased serum K^+ concentration during postarrest pyruvate treatment, an effect that subsided after pyruvate clearance. Such reductions in circulating potassium would ameliorate pro-arrhythmic hyperkalemia that may re-arrest the heart.⁸ On the other hand, serum K^+ concentrations below 3 mmol/L are associated with QT prolongation and risk of cardiac standstill.⁵¹ Although reversible, this hypokalemic effect may impose an upper limit on the therapeutic pyruvate dosage in cardiac arrest patients.

Limitations

The experimental model differed in several important respects from clinical cardiac arrest. Experiments were conducted in healthy, juvenile swine free of the chronic diseases that afflict cardiac arrest patients. These juvenile swine had compliant chest walls that facilitated delivery of effective chest compressions. Chest compressions were begun at 6 min cardiac arrest, and defibrillation attempted at 10 min arrest to ensure a high survival rate, whereas the durations of cardiac arrest and CPR in human victims often are more prolonged and are associated with much higher mortality. Cardiac arrest was induced in anesthetized animals, whereas most human cardiac arrest victims are not anesthetized when they are stricken. Although cerebral perfusion pressures achieved by precordial compressions were monitored in a subset of nine animals, the effects of pyruvate vs. NaCl treatments on cerebral perfusion pressure were not compared. Mongan *et al.*⁵² demonstrated that i.v. sodium pyruvate infusions in pigs, at rates similar to those of the present study, altered neither systemic arterial pressure nor cerebral perfusion pressure vs. equimolar NaCl infusions. In the present study, there was no treatment effect on systemic arterial pressure during the precordial compressions. Male and female pigs were studied, since

both genders are susceptible to cardiac arrest, but the potential for confounding effects of endogenous hormones on experimental variables must be recognized.

Authors' Contributions: Study conception: AHOY, RTM; experimental design: BHC, AGW, BH, AHOY, RTM; surgical preparation: BHC, AQN, AGW, BH; preparing experimental treatments: RTM; conducting experimental protocols: BHC, AQN, RAH, AGW, BH; collecting, extraction and analysis of plasma samples: RAH; data reduction, analysis and statistics: BHC, AQN, RAH, AGW; preparation of figures: BHC, RAH, AHOY, RTM; wrote manuscript: BHC, RTM; edited manuscript: BHC, AQN, BH, AHOY, RTM.

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