A new role for cardioplegic buffering: Should acidosis or calcium accumulation be counteracted to salvage jeopardized hearts?

Manuel Castellá, MD^a
Gerald D. Buckberg, MD^a
Saleh Saleh, MD^b
Zhongtuo Tan, PhD^a
Louis J. Ignarro, PhD^c

See related editorial on page 1265.

Objectives: Thirty minutes of unprotected ischemia produced a jeopardized heart that was treated with a blood cardioplegic solution containing the natural erythrocyte and protein buffers. Cardioplegic pH was changed to 7.7 (buffered) or 7.2 (nonbuffered), and this was tested alone and after pretreatment with Na⁺-H⁺ exchange blockade (cariporide) to define their protective effects.

Methods: Twenty-four Yorkshire-Duroc pigs (27-34.5 kg) underwent 30 minutes of normothermic global ischemia, followed by 30 minutes of aortic clamping during protection with buffered (n = 12) or nonbuffered (n = 12) glutamate-aspartate-enriched blood cardioplegic solution. Twelve hearts (6 buffered and 6 nonbuffered) were pretreated with intravenous cariporide (5 mg/kg) 15 minutes before ischemia.

Results: Severe and comparable left ventricle dysfunction followed buffered or nonbuffered cardioplegia: Preload recruitable stroke work recovered to 56% ± 21% and 45% ± 20% of baseline levels; creatine kinase MB, conjugated dienes, and myeloperoxidase activity markedly increased; moderate myocardial edema occurred; and endothelin-1 increased 2-fold more than baseline values. Cariporide pretreatment caused a similar return of preload recruitable stroke work to 86% ± 9% and 90% ± 6% after buffered or nonbuffered cardioplegia (*P* < .05 vs nonpretreated groups), allowed only minor creatine kinase MB and conjugated diene changes, and reduced endothelin-1 release 3-fold compared with hearts without sodium-hydrogen exchange blockade.

Conclusions: The severe ischemia-reperfusion injury of 30 minutes of normothermic ischemia is not altered by an acidic or alkalotic pH cardioplegic solution. Correction of damage is achieved by adding Na⁺-H⁺ exchange blocker therapy before treatment with buffered and nonbuffered solutions; thus, sodium-hydrogen exchange inhibition plays a more vital role in recovery than pH management.

From the Department of Surgery, Divisions of Cardiothoracic Surgery,^a Nephrology,^b and Physiology,^c University of California, Los Angeles, School of Medicine, Los Angeles, Calif.

Received for publication May 2, 2002; revisions requested May 15, 2002; accepted for publication March 12, 2003.

Address for reprints: Gerald D. Buckberg, MD, Division of Cardiothoracic Surgery, 62-258 Center for the Health Sciences, Los Angeles, CA 90095-1701 (E-mail: gbuckberg@mednet.ucla.edu).

J Thorac Cardiovasc Surg 2003;126:1442-8

Copyright © 2003 by The American Association for Thoracic Surgery

0022-5223/2003 \$30.00 + 0

doi:10.1016/S0022-5223(03)00599-3

Acidosis follows ischemia and is conventionally the enemy of the surgeon who normally introduces a buffer to offset the cardiac results of H⁺ accumulation. Acidosis contributes to organ failure and death by slowing cell metabolism to delay enzyme reactions and impairs contraction by inhibiting the calcium-binding site on troponin-C.¹ Intracellular proton accumulation triggers the 2 major cellular buffer mechanisms to retain a normal pH: (1) the sodium-hydrogen

exchange (NHE) that exchanges intracellular H^+ for Na^+ and (2) the sodium-bicarbonate symporter that allows entrance of bicarbonate and sodium into the cell. Anaerobiosis limits the energy-dependent Na^+-K^+ pump, and sodium overload is compensated by the Na^+/Ca^{++} transporter, allowing calcium accumulation.

Reperfusion washes out the extracellular space to allow the full action of these transporters, especially when buffering causes rapid realkalinization. A promptly restored physiologic pH enhances intracellular calcium overload, contributing to rigor, hypercontracture, and death.² New approaches suggest that NHE system blockade will delay rapid realkalinization during reperfusion and avoid hypercontracture.^{3,4} This direct proton inhibition of calcium-induced hypercontraction has been known as the pH paradox.⁵

We studied whether pH was a central feature of protection against damage or was management interrelated with NHE exchangers that interface pH with potential damage from calcium entry. Our objectives were to define the following: (1) the effectiveness of buffering the cardioplegic reperfusion in extremely jeopardized hearts, (2) the contribution of preischemic NHE inhibition to the low and high pH cardioplegic reperfusate, (3) the importance of cardioplegic reperfusate pH during the management of severely injured jeopardized hearts compared with pH-independent calcium management, and (4) the possible interaction of pH management and NHE blockage on coronary endothelial damage after severe normothermic ischemic injury.

Materials and Methods

All animals received humane care in compliance with the "Principles of Laboratory Animal Care," formulated by the Institute of Laboratory Animal Resources, and the "Guide for the Care and Use of Laboratory Animals" prepared by the Institute of Laboratory Animal Resources, National Research Council, and published by the National Academy Press, revised 1996.

Twenty-four Yorkshire-Duroc pigs (27-34.5 kg) were premedicated (ketamine 15 mg/kg and diazepam 0.5 mg/kg intramuscularly) and anesthetized (pentobarbital 30 mg/kg intravenously). Support with a volume-controlled ventilator (Servo 900C, Siemens-Elema, Solna, Sweden) was started after tracheostomy and endotracheal intubation. The femoral artery and vein were cannulated, and arterial blood gases were measured to keep PO_2 , PCO_2 , and pH values within the normal range. A pulmonary artery balloon-tipped catheter (model 132F5, Baxter Healthcare Corp, Irvine, Calif) measured cardiac output (thermodilution technique) before and after cardiopulmonary bypass (CPB).

After median sternotomy, an apical solid-state pressure transducer-tipped catheter (MPC-500, Millar Inc, Houston, Tex) monitored left ventricular (LV) pressure. LV dimensions were measured with endocardially placed 2-mm ultrasonic microtransducer crystals (Sonometrics, London, Ontario, Canada). Two pairs of crystals were oriented across the minor and major axes. LV volume was assessed by an ellipsoid-based formula. Pressure-volume

loops were recorded digitally (Sonometrics Corporation, London, Ontario, Canada).

After systemic heparinization (300 U/kg), extracorporeal circulation was achieved using a 12F aortic cannula and a dual-lumen 29F venous right atrial cannula with a membrane oxygenator (Affinity NT541, Medtronic, Inc, Minneapolis, Minn) and extracorporeal pump (Sarns, Ann Arbor, Mich). The circuit prime was 1000 mL Plasma-Lyte solution (Baxter Healthcare Corp, Deerfield, Ill), 700 mL stored porcine packed blood, and calcium chloride for normocalcemia (1.0-1.2 mmol/L). Oxygen tension was kept at 300 mm Hg and aortic pressure was kept at 50 to 70 mm Hg by adjusting the flow to keep approximately 70% mixed venous oxygen saturation at 35°C to 37°C. Potassium, calcium, and pH were kept at normal levels. Aortic pressure was measured during delivery of cardioplegic solution. The blood cardioplegic solution was hyperkalemic (20 mg KCl/L), hypocalcemic (0.2 mEq/L Ca^{2+}), and enriched with glutamate and aspartate.⁶ The coronary sinus was cannulated for blood sampling, and the LV was vented.

Experimental Protocol

All pigs underwent 30 minutes of normothermic aortic clamping before adding 30 more minutes of ischemia with cardioplegic protection with amino acid-enriched blood cardioplegic solution, including warm induction, cold maintenance, and a warm reperfusate. Pigs were divided into 4 groups.

Buffered cardioplegic reperfusion. In 6 pigs, reperfusion pH was 7.7 ± 0.06 during the added 30 minutes with use of 4:1 blood cardioplegic solution at 200 mL/L during warm induction (2 minutes), cold maintenance (2 minutes), each 15 minutes, and warm reperfusate (3 minutes) before unclamping.⁶

Nonbuffered cardioplegic reperfusion. In 6 pigs, reperfusion pH was 7.2 ± 0.1 , because buffer (tromethamine) was not added to the cardioplegic solution.

Cariporide and buffered cardioplegic reperfusion. In 6 pigs, 5 mg/kg HOE-642 was added to the extracorporeal circuit 15 minutes before normothermic ischemia (cariporide half-life in swine is 30 minutes), and a pH 7.7 blood cardioplegic solution was given during 30 added minutes of aortic clamping.

Cariporide and nonbuffered cardioplegic reperfusion. In 6 pigs, 5 mg/kg HOE-642 was added 15 minutes before normothermic ischemia. A pH 7.2 blood cardioplegia was used because tromethamine was not added.

Measurements. CPB was continued 30 minutes after unclamping the aorta. Global LV function before and 30 minutes after CPB was assessed by pressure-volume analysis. LV pressure and volume were recorded during transient inferior vena cava occlusions to obtain a series of evenly declining pressure-volume loops. Global stroke work and end-diastolic volume were calculated with a video-graphics program (Sonometrics). Preload recruitable stroke work (PRSW) for each series was identified as the relation between stroke work and end-diastolic volume, and quantified by a slope (M_w) and x-intercept (L_w). The slope ($erg \cdot cm^{-3} \cdot 10^3$) provides a reliable measure of intrinsic myocardial performance independent of loading, geometry, and heart rate.^{7,8} Postbypass LV performance was the percentage of recovery from prebypass values.

Figure 1. Recovery of preload recruitable stroke work, expressed as percentage of preischemic control values (* $P < .05$ vs non-cariporide groups).

Coronary Sinus Blood Analysis

Samples were taken 5 minutes after initiating CBP (baseline) and just before ending CPB (reperfusion), whereas the coronary flow rate was controlled at 100 mL/min into the crossclamped aorta to fix delivery.

Conjugated diene (CD) levels to mark oxidant-mediated lipid peroxidation were determined spectrophotometrically after chloroform-methanol 2:1 (vol/vol) extraction. Concentration was expressed as absorbance at a wavelength of 240 nm per 0.5 mL plasma. Creatine kinase (CK) MB (units/liter) was determined by an ultraviolet-spectrophotometric method (Sigma Chemical Co, St. Louis, Mo). Nitric oxide (NO) (micromoles/liter) was determined as its spontaneous oxidation products, nitrite and nitrate, which were converted to NO and quantitated by a chemiluminescence assay using a nitrogen oxides analyzer (DASIBI Environmental Corp, model 2108, Glendale, Calif). Endothelin (ET)-1 (picograms/milliliter) was determined after sample purification (Ethyl C2 Amprep minicolumns, Amersham Pharmacia Biotech, Piscataway, NJ) by an enzyme immunometric assay (ACETM EIA kit, Cayman Chemical Company, Ann Arbor, Mich).

Myocardial Biopsy

At the end of the experiment, pigs were killed by bolus injections of pentobarbital 5 mg followed by 15 mL cold hyperkalemic blood (KCl, 30 mEq/L). Transmural LV myocardium samples (0.5 g from anterior free wall) were immediately frozen in liquid nitrogen until analyzed for neutrophil-specific myeloperoxidase activity (units/gram). Tissue water content was measured by weight loss before and after incineration.

Statistical Analysis

Statistical analysis within and between groups was performed with multiple analysis of variance followed by application of the Stu-

dent t test with Bonferroni's correction for multiplicity. All data are expressed as mean $\pm$ SD.

Results

LV Performance

There was no operative mortality. Reperfusion with blood cardioplegia, with or without buffering, allowed $56\% \pm 21\%$ and $45\% \pm 20\%$ PRSW recovery (from 58.1 ± 8.2 to 37.9 ± 15.2 , and from 51.6 ± 7.5 to 23.3 ± 10.9 erg $\cdot$ cm $^{-3} \cdot 10^3$), respectively (Figure 1). There was no difference in recovery related to pH management. Conversely, NHE inhibition markedly improved recovery of PRSW to $86\% \pm 9\%$ and $90\% \pm 6\%$ (from 56.1 ± 7.0 to 48.7 ± 10.3 , and from 55.6 ± 6.9 to 49.8 ± 7.1 erg $\cdot$ cm $^{-3} \cdot 10^3$) ($P < .05$ vs nonpretreated groups) with buffered and nonbuffered cardioplegic reperfusates.

Myocardial Damage

CK-MB without cariporide pretreatment increased similarly with buffered (72 ± 15 U/L) and nonbuffered (69 ± 16 U/L) reperfusates (Figure 2). Conversely, cariporide pretreatment similarly decreased CK-MB in buffered (42 ± 4 U/L) and nonbuffered (30 ± 5 U/L) cardioplegic reperfusates ($P < .05$).

High CD levels occurred in the non-pretreated groups (Figure 3) and in the buffered and nonbuffered cardioplegic groups without cariporide. Conversely, cariporide pretreatment similarly reduced CD values in buffered and nonbuffered groups (1.31 ± 0.11 and 1.19 ± 0.09 A/mL, respectively).

Figure 2. Levels of CK-MB in units per liter from coronary sinus plasma, at baseline and after 30 minutes of reperfusion (* $P < .05$ vs non-cariporide groups).

Figure 3. Levels of conjugated dienes in coronary plasma expressed as absorbance (A) per 0.5 mL, at baseline and after 30 minutes of reperfusion (* $P < .05$ vs non-cariporide groups).

Myeloperoxidase activity averaged 0.029 ± 0.004 U/g after unprotected ischemia and either buffered or nonbuffered (0.030 ± 0.004 U/g) blood cardioplegic reperfusion. Cariporide pretreatment reduced myeloperoxidase activity to 0.014 ± 0.004 U/g and 0.013 ± 0.002 , without difference between buffered and unbuffered reperfusion groups ($P < .05$ pretreated vs nonpretreated groups).

Sixty minutes of CPB without ischemia slightly increased water content from $79.5\% \pm 0.5\%$ to $80.4\% \pm 0.1\%$. Significant water gain followed unprotected ischemia and controlled cardioplegic reperfusion without ca-

riporide (81.5% and 81.6% in buffered and nonbuffered cardioplegic reperfusion groups) (Figure 4). Cariporide pretreatment reduced water content to 80.2% and 80.1% in buffered and nonbuffered groups, respectively, levels similar to those in management of nonischemic hearts with blood cardioplegia.

Endothelial Response

A 2-fold increase in ET-1 (1.25 ± 0.07 pg/mL) followed buffered (2.05 ± 0.38 pg/mL) and nonbuffered (1.98 ± 0.36 pg/mL) cardioplegic administration. Cariporide pre-

Figure 4. Left ventricular anterior free wall myocardial water content expressed as percentage of weight, taken at the end of the reperfusion period (* $P < .05$ vs non-cariporide groups).

treatment decreased ET-1 in buffered and unbuffered groups (1.18 ± 0.10 and 1.13 ± 0.06 pg/mL) to levels comparable with controls without ischemia ($P < .05$ vs noncariporide groups). Baseline NO levels showed major variability between groups, with an F less than 0.05 that limited subsequent statistical analysis.

Discussion

We studied ways to protect jeopardized hearts that must undergo an added interval of aortic clamping for surgical repair. Our results show that cardioplegic solution pH management during simulated surgical repair does not influence contractile dysfunction and abnormal biochemical and endothelial markers of structural damage. These effects contrast with the beneficial protection of alkalotic solutions during cardioplegic arrest in nonjeopardized hearts.^{9,10} We hypothesize a mechanism related to a time-dependent calcium accumulation after severe unprotected ischemia that is so profound that pH management during cardioplegic reperfusion plays only a marginal role in improving myocyte and endothelial recovery.

Our data do not question the role of buffering in less jeopardized hearts requiring cardioplegic protection. Buffering remains a central surgical theme, because alkalotic buffering during cardioplegic arrest slows the evolution of acidosis and delays NHE activation. Repetitive cardioplegic

deliveries maintain arrest and prolong the interval before ischemic calcium overload becomes a threat for contractile dysfunction. This process differs from the deep acidosis after prolonged unprotected ischemia (ie, ~30 minutes) that more severely activates NHE and produces intracellular sodium and calcium overload, myocardial edema, and calcium-induced damage.^{2,3,11-13}

In this jeopardized heart study, controlled alkalotic hypocalcemic cardioplegic reperfusion limited sudden hypercontracture, cytoskeleton loss, and sarcolemmal disruption, but allowed only partial recovery of function. We ascribe the persistent dysfunction to NHE activation from ischemia-induced acidosis resulting in calcium accumulation. The NHE inhibition-related improvement was independent of the pH of the blood cardioplegic perfusate, because improvement followed either acidotic (pH 7.2) or alkalotic (pH 7.7) methods of pH management (ie, with buffered vs nonbuffered solutions). These findings indicate the operative factor may be calcium loading during the ischemia-reperfusion interval, but intramyocardial ionic measurements were not performed to confirm this concept.

Potential mechanisms to limit calcium loading include maintaining arrest during early reperfusion, pretreating with NHE inhibition, and limiting calcium loading with specific blockade of Ca^{2+} at cellular membrane and sarcolemmal reticulum levels. Only cardioplegia and NHE blockade were

combined in our experiments. Cariporide administration before ischemia may have prevented ischemic injury, as we showed previously.¹⁴ Cariporide's 30-minute half-life allowed some supplementation of the cardioplegic solution, because marked improvement extended the functional benefits of these acid and alkaline solutions without cariporide. A third alternative, not tested in this study, is that cardioplegic cariporide supplementation prevented reperfusion injury. We recently found that reserving cariporide supplementation to only controlled reperfusion¹⁵ mirrored the preoperative finding in this study. Clearly, inhibition of Na^+ H^+ exchange has a broad application, because cariporide can act as both a pretreatment¹⁴ in jeopardized hearts (as an intravenous cardioplegic solution component because of its 30-minute half-life in this report) and a direct cardioplegic additive during controlled reperfusion.¹⁵

Although pH management with either alkalotic or acidotic cardioplegic solutions did not retard ET-1 release without cariporide, NHE blockage limited ET-1 release in both the buffered and nonbuffered studies. The NO effects were variable, but an endothelial benefit was indicated because NHE blockade reduced leukocyte vascular adherence. Coronary vasoactivity was not assessed to confirm this endothelial benefit.

Therapeutic Potential

The clinical role of cariporide in modifying ischemia-reperfusion damage is not yet established. Experimental pretreatment with NHE blockers reduces infarct size,^{2,3} improves recovery of LV dysfunction after infarction,³ reduces apoptosis,¹⁶ preserves adenosine triphosphate,¹⁷ and reduces arrhythmias.¹³ Unfortunately, initial clinical trials with cariporide did not prevent death or myocardial infarction in high-risk nonsurgical patients,¹⁸ but they showed some benefit in heart surgery patients with jeopardized muscle.³ Comparisons of effectiveness were limited by the wide spectrum of methods of intraoperative protection. This report evaluates the importance of buffering during NHE inhibition in a single but well-tested cardioplegic strategy.⁶

Buffering remains a central and therapeutic factor during aortic clamping in jeopardized hearts, but the experimental model plays an important role in defining the interaction between pH management and NHE inhibition. The absence of a difference between high pH and low pH solutions in non-cariporide studies should not lead to the conclusion that buffering is not important. A nonbicarbonate buffer, such as Tris-hydroxymethylaminomethane, can avoid subsequent carbon dioxide production and sodium loading, consequences described with the use of bicarbonate.⁴ The critical factor is not related to pH per se, but to how breakdown of a bicarbonate buffer can accentuate acidosis and stimulate resultant Na^+ H^+ exchange and subsequent calcium loading.

Study Limitations

This study did not include tissue pH or ion flux measurements over time, because repetitive biopsy measurements of Na^+ , Ca^{2+} , or water content could have affected the clinical variables measured. Water content was measured at the end of reperfusion and may be closely related to Na^+ load.

We studied a stunned myocardium model because global LV function may improve if more than 30 minutes of support is provided or inotropic interventions started. Thirty minutes of normothermic ischemia initiated a critical lesion that was almost completely avoided by cariporide pretreatment. We did not add the extra 30 minutes of aortic clamping without cardioplegic protection because such treatment is essential in jeopardized hearts needing repair. This "all or nothing" effect with cardioplegia may hide differences between the buffered and unbuffered groups, because the injury in the pretreated animals was minor. However, this design limitation was offset in other recent experiments in which cariporide was added only during controlled reperfusion and produced similar results.¹⁵

Conclusion

The use of acidic or alkalotic pH management during cardioplegic reperfusion does not change functional, biochemical, or endothelial recovery in severely jeopardized hearts. The prevention of calcium overload by NHE inhibition pretreatment corrects myocardial dysfunction after normothermic ischemia, and pH management of the cardioplegic solution does not affect this benefit. The absence of a salutary effect of adding a cardioplegic buffer should not exclude buffering of the cardioplegic solution. The prevention of intracellular calcium overload by NHE blockade may be a critical supplement to the cardioplegic perfusate for enhancing myocyte and endothelial recovery in severely jeopardized hearts.

References

1. Karmazyn M, Moffat MP. Role of Na^+ H^+ exchange in cardiac physiology and pathophysiology: meditation of myocardial reperfusion injury by the pH paradox. *Cardiovasc Res.* 1993;27:915-24.
2. Garcia-Dorado D, Gonzalez MA, Barrabes JA, Ruiz-Meana M, Solares J, Lidon RM, et al. Prevention of ischemic rigor contracture during coronary occlusion by inhibition of Na^+ H^+ exchange. *Cardiovasc Res.* 1997;35:80-9.
3. Ruiz-Meana M, Garcia-Dorado D, Julia M, Inserte J, Siegmund B, Ladilov Y, et al. Protective effect of HOE 642, a selective blocker of Na^+ H^+ exchange, against the development of rigor contracture in rat ventricular myocytes. *Experimental Physiol.* 2000;85:17-25.
4. Schaefer C, Ladilov YV, Siegmund B, Piper HM. Importance of bicarbonate transport for protection of cardiomyocytes against reoxygenation injury. *Am J Physiol Heart Circ Physiol.* 2000;278:H1457-63.
5. Currin RT, Gores GJ, Thurman RG, Lemasters JJ. Protection by acidotic pH against anoxic cell killing in perfused rat liver: evidence for a pH paradox. *FASEB J.* 1991;5:207-10.
6. Buckberg GD. Antegrade/retrograde blood cardioplegia to ensure cardioplegic distribution: operative techniques and objectives. *J Card Surg.* 1989;4:216-38.
7. Glower DD, Spratt JA, Snow ND, Kabas JS, Davis JW, Olsen CO, et

- al. Linearity of the Frank-Starling relationship in the intact heart: the concept of preload recruitable stroke work. *Circulation*. 1985;71:994-1009.
8. Glower DD, Spratt JA, Kabas JS, Davis JW, Rankin JS. Quantification of regional myocardial dysfunction after acute ischemic injury. *Am J Physiol*. 1987;255:H85-93.
 9. Stinner B, Krohn E, Gebhart MM, Bretschneider HJ. Intracellular sodium activity and Bretschneider's cardioplegia: continuous measurement by ion-selective microelectrodes at initial equilibration. *Basic Res Cardiol*. 1989;84:197-207.
 10. Buckberg GD. Development of blood cardioplegia and retrograde techniques: the experimenter/observer complex. *J Card Surg*. 1998;13:163-70.
 11. Avkiran M. Rational basis for use of sodium-hydrogen exchange inhibitors in myocardial ischemia. *Am J Cardiol*. 1999;83:10G-8G.
 12. Scholz W, Albus U, Counillon L, Goegelein H, Lang HJ, Linz W, et al. Protective effects of cariporide, a selective sodium-hydrogen exchange subtype 1 inhibitor, on cardiac ischemia and reperfusion. *Cardiovasc Res*. 1995;29:260-8.
 13. Jayachandran JV, Zipes DP, Weksler J, Olgin JE. Role of the Na^+/H^+ exchanger in short-term atrial electrophysiological remodeling. *Circulation*. 2000;101:1861-6.
 14. Castellá M, Buckberg GD, Tan Z, Ignarro LJ. Myocyte and endothelial effects of preconditioning the jeopardized heart by inhibiting Na^+/H^+ exchange. *J Thorac Cardiovasc Surg*. 2002;124:1113-21.
 15. Castellá M, Buckberg GD, Tan Z, Ignarro LJ. Blood cardioplegic protection in profoundly damaged hearts: role of Na^+/H^+ exchange inhibition during pretreatment or during controlled reperfusion supplementation. *Ann Thorac Surg*. 2003;75:1238-45.
 16. Chakrabarti S, Hoque AN, Karmazyn M. A rapid ischemia induced apoptosis in isolated rat hearts and its attenuation by the Na^+/H^+ exchange inhibitor HOE 642 (cariporide). *J Mol Cell Cardiol*. 1997;29:3169-74.
 17. Mathur S, Karmazyn M. Interaction between anesthetics and the Na^+/H^+ exchange inhibitor HOE 642 (cariporide) in ischemic and reperfused rat hearts. *Anesthesiology*. 1997;29:3169-74.
 18. Théroux P, Chaitman BR, Danchin N, Erhardt L, Meinertz T, Schroeder JS, et al. Inhibition of the sodium-hydrogen exchanger with cariporide to prevent myocardial infarction in high-risk ischemic situations. Main results of the GUARDIAN trial. *Circulation*. 2000;102:3032-8.

Availability of Journal back issues

As a service to our subscribers, copies of back issues of ***The Journal of Thoracic and Cardiovascular Surgery*** for the preceding 5 years are maintained and are available for purchase from Mosby until inventory is depleted. Please write to Mosby, Subscription Customer Service, 6277 Sea Harbor Dr, Orlando, FL 32877, or call 800-654-2452 or 407-345-4000 for information on availability of particular issues and prices.