SURGERY FOR CONGENITAL HEART DISEASE

COMPARISON OF NEUROLOGIC OUTCOME AFTER DEEP HYPOTHERMIC CIRCULATORY ARREST WITH ALPHA-STAT AND PH-STAT CARDIOPULMONARY BYPASS IN NEWBORN PIGS

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Objective: Deep hypothermic circulatory arrest for neonatal heart surgery poses the risk of brain damage. Several studies suggest that pH-stat management during cardiopulmonary bypass improves neurologic outcome compared with alpha-stat management. This study compared neurologic outcome in a survival piglet model of deep hypothermic circulatory arrest between alpha-stat and pH-stat cardiopulmonary bypass.

Methods: Piglets were randomly assigned to alpha-stat (n = 7) or pH-stat (n = 7) cardiopulmonary bypass, cooled to 19°C brain temperature, and subjected to 90 minutes of deep hypothermic circulatory arrest. After bypass rewarming/reperfusion, they survived 2 days. Neurologic outcome was assessed by neurologic performance (0-95, 0 = no deficit and 95 = brain death) and functional disability scores, as well as histopathology. Arterial pressure, blood gas, glucose, and brain temperature were recorded before, during, and after bypass.

Results: All physiologic data during cardiopulmonary bypass were similar between groups (pH-stat vs alpha-stat) except arterial pH (7.06 ± 0.03 vs 7.43 ± 0.09, $P < .001$) and arterial P_{CO_2} (98 ± 8 vs 36 ± 8 mm Hg, $P < .001$). No differences existed in duration of cardiopulmonary bypass or time to extubation. Performance was better in pH-stat versus alpha-stat management at 24 hours (2 ± 3 vs 29 ± 17, $P = 0.004$) and 48 hours (1 ± 2 vs 8 ± 9, $P = .1$). Also, functional disability was less severe with pH-stat management at 24 hours ($P = .002$) and 48 hours ($P = .053$). Neuronal cell damage was less severe with pH-stat versus alpha-stat in the neocortex (4% ± 2% vs 15% ± 7%, $P < .001$) and hippocampal CA1 region (11% ± 5% vs 33% ± 25%, $P = .04$), but not in the hippocampal CA3 region (3% ± 5% vs 16% ± 23%, $P = .18$) or dentate gyrus (1% ± 1% vs 3% ± 6%, $P = .63$).

Conclusions: pH-stat cardiopulmonary bypass management improves neurologic outcome with deep hypothermic circulatory arrest compared with alpha-stat bypass. The mechanism of protection is not related to hemodynamics, hematocrit, glucose, or brain temperature. (J Thorac Cardiovasc Surg 2001;121:336-43)

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Deep hypothermia with total circulatory arrest (DHCA) is often used during repair of complex cardiovascular defects in neonates. It offers excellent operating conditions to improve surgical results but poses a risk of ischemic brain damage. Outcome studies report seizures, psychomotor delay, and cognitive deficits in 5% to 40% of survivors after DHCA.[1,2] Magnetic resonance imaging shows a spectrum of lesions after DHCA, including periventricular leukomalacia, neocortical atrophy, and focal infarction in the neocortex and striatum.[1,3] In animal models of neonatal DHCA, brain damage occurs mainly in the neocortex and hippocampus and less often in the cerebellum and striatum, whereas other brain regions remain relatively free of damage.[4,5]

Several perioperative factors have been associated with DHCA brain damage in neonates, including blood pH management during deep hypothermic cardiopulmonary bypass (CPB) before and after DHCA. Two strategies are used for blood pH management. Alpha-stat management aims to keep arterial blood measured at 37°C with a pH of 7.40 and an arterial P_{CO_2} of 40 mm Hg, even though in vivo the hypothermic blood is alkalemic and hypocapnic. In pH-stat management the goal is to maintain in vivo hypothermic arterial blood at a pH of 7.40 and an arterial P_{CO_2} of 40 mm Hg; when measured at 37°C, the blood is acidemic and hypercapnic. Alpha-stat gas management preserves autoregulation[6] and optimizes cellular enzyme activity, whereas pH-stat management improves cerebral blood flow (CBF), cerebral oxygenation, and brain cooling efficiency during CPB.[7-9] Potential disadvantages for alpha-stat include less metabolic suppression[10,11] and for pH-stat, greater risk of microembolism[12] and free radical–mediated damage.[13]

Although cerebral physiology differs between pH-stat and alpha-stat management, it remains uncertain whether neurologic outcome after DHCA differs. Nonsurvival neonatal animal models of DHCA have shown better cerebral physiologic and metabolic recovery with pH-stat than with alpha-stat management.[7,14] However, neurologic outcome cannot be truly judged in a nonsurvival model because cerebral physiologic and metabolic parameters are surrogate markers. A retrospective study of infants who had heart surgery reported worse developmental outcome with alpha-stat management.[15] A prospective study in a similar population showed earlier electroencephalographic recovery and shorter duration of mechanical ventilation with pH-stat management.[16] Despite results favoring pH-stat management, limitations with these studies have not resolved the controversy about optimal pH management. In the present study, we assessed whether pH-stat CPB improved neurologic outcome in a newborn pig model of DHCA by neurologic examinations and histopathologic evaluations.

Materials and methods

The Institutional Animal Care and Use Committee at the Joseph Stokes Research Institute at The Children's Hospital of Philadelphia approved this project. We studied 14 piglets, aged 5 to 10 days, which weighed 2.15 to 3.25 kg. Anesthesia was induced with an intramuscular injection of ketamine (33 mg/kg) and acepromazine (3.3 mg/kg) followed by inhalation of halothane (0.5%). After the trachea was intubated, the lungs were mechanically ventilated. The electrocardiogram (Hewlett-Packard Company, Andover, Mass) and end-expired carbon dioxide (Normocap, Datex-Ohmeda Division, Instrumentarium Corp, Helsinki, Finland) were monitored. Normothermia was maintained with a circulating warm water blanket and overhead heating lamps.

Aseptic technique was used for all surgical procedures. Catheters were inserted in a peripheral vein to administer drugs and in the femoral artery to monitor arterial pressures (23 PXL Statham transducer to a model 3800 recorder; Statham-Gould, Valley View, Ohio), blood gases and hemoglobin concentrations (I-STAT, Princeton, NJ), and glucose concentrations (Stan Bio Lab Inc, San Antonio, Tex).

Cefazolin (25 mg/kg) was administered intravenously. Through a 1-cm incision in the scalp, a 2-mm hole was made in the skull over the left coronal suture to insert a microthermistor (555, Yellow Springs Instrument Co, Yellow Springs, Ohio) in the epidural space to monitor cortical temperature. Thermistors (model 401; Yellow Springs Instrument Co) were used to monitor esophageal and rectal temperature.

Through a 4-cm incision in the right side of the neck, the external jugular vein and the common carotid artery were identified. Before vessel cannulation, pancuronium (0.2 mg/kg), fentanyl (25 μg/kg), and heparin (200 units/kg) were administered intravenously and halothane was discontinued. An 8F arterial cannula was advanced into the proximal ascending aorta and an 8F venous cannula was advanced into the right atrium. Approximately 10 mL/kg of blood was collected at the time of cannulation and stored at 4°C for transfusion after CPB.

The CPB circuit used a bubble oxygenator (Bio-2; Baxter Healthcare Corporation, Santa Ana, Calif) with an arterial filter (LPE 1440 extracorporeal filter; KOL Bio-Medical Instruments, Inc, Chantilly, Va) receiving oxygen at 1 L/min and a nonpulsatile roller pump (RS 7800; Minntech Renal Systems, Minneapolis, Minn) flowing at 100 mL $\cdot$ kg^{-1} $\cdot$ min^{-1}. For pH-stat, the oxygenator received 5% carbon dioxide and oxygen; for the alpha-stat group, the oxygenator received only oxygen. The pump prime (approximately 300-400 mL) contained pig whole blood (obtained from the sow), heparin 2000 units, calcium chloride 300 mg, sodium bicarbonate 5 mEq, pancuronium 1 mg, fentanyl 50 μg, dexamethasone 30 mg/kg, cefazolin 25 mg/kg, and furosemide 1 mg/kg. Plasma-Lyte A solution (Baxter Healthcare

Table I. *Neurologic performance scale*

Assessment	*Score*	*Maximum abnormal score*
Level of consciousness		25
Normal	0	
Clouded	5	
Stuporous	12	
Comatose	25	
Respiration		5
Normal	0	
Abnormal	5	
Cranial nerves		6
Vision absent	1	
Light reflex absent (right)	0.5	
Light reflex absent (left)	0.5	
Corneal reflex absent (right)	0.5	
Corneal reflex absent (left)	0.5	
Facial sensation absent	1	
Auditory absent	1	
Gag reflex absent	1	
Motor/sense function		14
Flexor response to pain in upper extremity absent (right)	1	
Flexor response to pain in upper extremity absent (left)	1	
Flexor response to pain in lower extremity absent (right)	1	
Flexor response to pain in lower extremity absent (left)	1	
Righting reflex absent	10	
Gait		25
Normal	0	
Minimal ataxia	5	
Moderate ataxia	10	
Able to stand	15	
Unable to stand	20	
No purposeful movement	25	
Behavior		20
Not drinking	10	
Not exploring	10	
Worst possible score		95

Testing of the oculocephalic reflex (1 point) and not grooming (4 point) were omitted from the original scale. This system is modified from Baker AJ, Zornow MH, Grafe MR, Scheller MS, Skilling SR, Smullin DH, et al. Hypothermia prevents ischemia-induced increases in hippocampal glycine concentrations in rabbits. Stroke 1991;22(5):666-73.

Corporation, Deerfield, Ill) was added to yield a hematocrit value of 20% to 25% during CPB.

Hypothermia was induced with core and surface cooling. CPB perfusate temperature was controlled by a water bath heater-cooler system (model 1141; VWR Scientific Products, West Chester, Pa). Arterial perfusate temperature was gradually decreased to keep perfusate temperature 5°C to 10°C less than body temperatures. Bags of ice were applied to the chest, abdomen, and head. Mechanical ventilation was suspended during CPB. No vasoactive drugs were administered during CPB.

After cooling to a brain temperature of 20°C, the pump was turned off. No arterial pressure and asystole on the electrocardiogram confirmed circulatory arrest. The venous CPB line remained open. After 90 minutes of circulatory arrest, CPB was resumed at 100 mL · kg^{-1} · min^{-1}. Rewarming was achieved by surface (external heating lamps and a circulating warm water blanket) and core techniques. Arterial perfusate temperature was gradually increased to a maximum of 38°C, keeping perfusate temperature 5°C to 10°C above body temperatures. At 15 minutes' reperfusion, the heart was defibrillated (50-100 joules) and mechanical ventilation resumed. After 20 minutes of reperfusion, the carbon dioxide in the pH-stat group was discontinued. Mannitol 0.5 gm/kg was administered at 28°C (cortical). CPB was discontinued when all temperatures were greater than 33°C.

After CPB, the cannulas were removed and protamine (4 mg/kg) was administered intravenously. Mean arterial blood pressure was maintained greater than 50 mm Hg by adjusting the rate of blood and crystalloid administration. No inotropic agents or vasoactive drugs were administered after CPB. Minute ventilation and supplemental oxygen were adjusted to maintain normocapnia (arterial P_{CO_2} 35-40 mm Hg) and normoxia (arterial P_{O_2} > 75 mm Hg). The epidural thermistor was removed, and all skin incisions were sutured closed. The autologous blood collected before CPB was transfused intravenously over 1 to 2 hours to replace clotting factors and platelets consumed during CPB, followed by intravenous 5% dextrose in lactated Ringer's solution at 4 mL · kg^{-1} · h^{-1}. Once the piglet had regained strength, protective airway reflexes, and regular breathing, the tracheal tube was removed. After close observation, the remaining catheters were removed and the animal was returned to the cage. Wounds were inspected daily and oral intake and weights were documented. If the piglet was unable to feed, bottle-feeding was instituted. Piglets unable to bottle-feed were administered lactated Ringer's solution intravenously to maintain hydration. The animals were allowed to survive for 48 hours.

Neurologic outcome consisted of behavioral and histopathologic examinations. At 24 and 48 hours after DHCA, a physician blinded to treatment group examined the animals and determined a neurologic performance score and a functional disability score. The *neurologic performance scale*[17] (Table I) consisted of a physical examination with points given for deficits. A normal examination score was 0 and the worst score was 95. The *functional disability score* was ranked from 1 to 5: *score 1* (no disability): able to run, explore the environment, and feed from the trough; *score 2* (mild disability): gait disturbances but able to ambulate, explore the environment, and feed from the trough; *score 3* (moderate disability): unable to walk and required bottle-feeding, but was alert and able to crawl; *score 4* (severe disability): not able to feed even with assistance and unable to crawl; *score 5:* death.

After the final observation, each animal was anesthetized with ketamine (33 mg/kg) and acepromazine (3.3 mg/kg) intramuscularly. The femoral vein was cannulated and the animal was killed (pentobarbital 100 mg/kg). A median ster-

Table II. *Physiologic data before, during, and after CPB in the alpha-stat and pH-stat CPB groups*

	Baseline		*Cooling CPB*		*Warming CPB*		*Two hours after CPB*	
	Alpha-stat	*pH-stat*	*Alpha-stat*	*pH-stat*	*Alpha-stat*	*pH-stat*	*Alpha-stat*	*pH-stat*
pH	7.44 ± 0.04	7.40 ± 0.08	7.43 ± 0.09	7.06 ± 0.03†	7.34 ± 0.11	7.27 ± 0.1	7.41 ± 0.07	7.40 ± 0.06
Arterial P_{CO_2} (mm Hg)	36 ± 5	44 ± 9	36 ± 8	99 ± 8†	27 ± 5	45 ± 14‡	36 ± 4	41 ± 4§
Arterial P_{O_2} (mm Hg)	316 ± 168	300 ± 189	800 ± 0	747 ± 112	522 ± 63	613 ± 92	146 ± 62	171 ± 119
MAP (mm Hg)	58 ± 10	63 ± 10	44 ± 11	44 ± 11	45 ± 12	46 ± 6	83 ± 11	95 ± 15
Hematocrit (%)	27 ± 3	22 ± 4*	25 ± 3	24 ± 3	26 ± 3	24 ± 2	31 ± 3	29 ± 3
T_{brain} (°C)	36 ± 1	36 ± 0.4	19 ± 0.5	19 ± 2	33 ± 2	31 ± 3	—	—
Glucose (mg/dL)	135 ± 37	121 ± 53	263 ± 51	217 ± 16	209 ± 30	207 ± 42	238 ± 70	200 ± 76

Mean ± SD; n = 7 each group. Arterial blood gases and pH are measured at 37°C. *Cooling CPB,* Deep hypothermic CPB before circulatory arrest; *warming CPB,* after circulatory arrest; P_{CO_2}, partial pressure of carbon dioxide; P_{O_2}, partial pressure of oxygen; *MAP,* mean arterial pressure; T_{brain}, cortical temperature.
*P = .05 versus alpha-stat.
†P < .001 versus alpha-stat.
‡P = .008 versus alpha-stat.
§P = .02 versus alpha-stat.

notomy was performed. The descending aorta was cross-clamped, the aortic root cannulated, and an outflow cannula placed in the right ventricle. One liter of 0.9% NaCl at 4°C and then 1 L of 4% paraformaldehyde were infused into the aorta to perfuse-fix the brain in situ. The brain was removed in toto and examined grossly. A cut was placed on the right ventral surface to mark the side of vessel ligation. Tissues were removed, soaked in 4% paraformaldehyde for 4 hours, and then soaked in 0.1-mol/L phosphate-buffered saline solution overnight at 4°C. The brain was sectioned coronally into 5-mm blocks, processed (Citadel 2000, Shandon-Lipshaw, Inc, Pittsburgh, Pa) in increasing concentrations of ethanol and xylene, and embedded in paraffin (model EG-1160, Leica histoembedder; Leica Microsystems, Inc, Wetzlar, Germany). Sections 8-μm thick from each block were cut with a microtome (Leica model RM-2155), mounted on poly-L-lysine–coated slides, and stained with hematoxylin and eosin.

A neuropathologist blinded to the treatment group examined the slides. Previously, we found neurons in the neocortex and hippocampus to be vulnerable to death after DHCA, whereas neurons in other regions were infrequently damaged or not damaged.[5] Therefore, we examined only the neocortex and hippocampus of each animal. The damaged neurons were identified by a combination of nuclear and cytoplasmic morphology such as eosinophilic cytoplasm, shrunken cytoplasm, pyknotic nuclei, or karyorrhexic nuclei (fragmented with chromatin distributed irregularly throughout the cytoplasm). On a scale of 0 to 4, a histologic grade was applied to the neocortex and hippocampus: 0 = normal neuronal structure; 1 = rare clusters (<5) of damaged neurons; 2 = occasional clusters (5-15) of damaged neurons; 3 = frequent clusters (>15) of damaged neurons; and 4 = diffusely distributed damaged neurons (clusters run together).

To assess the density of damaged neurons, we estimated the percentage of damaged cells in affected areas of the neocortex and hippocampus. The number of damaged and normal appearing neurons was counted in the neocortex (from the surface of the gray matter down to the border with the white matter in the third and fourth gyrus lateral to midline) and in the hippocampus (along the CA1, CA3, and dentate gyrus regions).

Table III. *Other important variables between alpha-stat and pH-stat CPB animals*

	Alpha-stat	*pH-stat*
Total CPB time (min)	61 ± 7	62 ± 8
Cooling to arrest	21 ± 4	22 ± 8
Rewarming CPB	40 ± 3	40 ± 6
Time to extubation (min)	341 ± 70	329 ± 66
Weight (kg)		
Preoperative	2.87 ± 0.5	2.67 ± 0.4
Postoperative day 1	2.89 ± 0.6	2.74 ± 0.4
Postoperative day 2	2.92 ± 0.7	2.75 ± 0.5

Values are mean ± SD.

Experimental design. Piglets were randomly assigned to alpha-stat (n = 7) or pH-stat (n = 7) groups before surgical preparation. Arterial blood gases, pH, hematocrit, and plasma glucose were recorded before, during, and after discontinuing CPB at regular intervals. Continuously monitored variables included the electrocardiogram, heart rate, mean arterial blood pressure, end-expired carbon dioxide, and temperatures. For each animal, the duration of CPB and the time to extubation after circulatory arrest were recorded.

Statistical analyses. Descriptive statistics (mean ± standard deviation) were used to summarize the data for the alpha-stat and pH-stat groups. The primary outcome measures were neurologic performance, functional disability, and the histopathologic evaluation of the neocortex and hippocampus. Also, comparisons were made between the 2 groups using the physiologic data (pH, arterial P_{CO_2}, arterial P_{O_2}, arterial pressure, glucose, hematocrit, and temperature), CPB times, weights, and time to extubation. For normally distributed continuous data, a *t* test was used to compare the 2 groups. For heavily skewed data and ordered categoric data

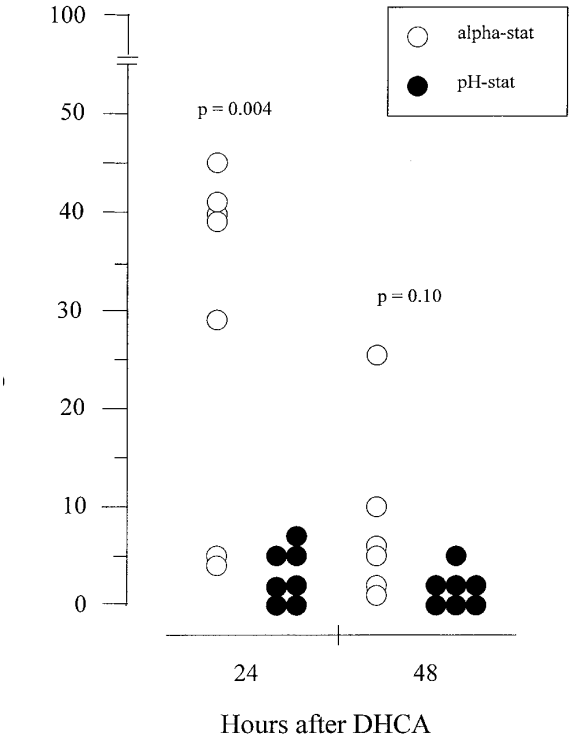

Fig 1. Neurologic performance score after DHCA with alpha-stat or pH-stat CPB on postoperative days 1 and 2. Score 0 represents no neurologic deficits and 95 represents brain death. One animal in the alpha-stat group died on postoperative day 1.

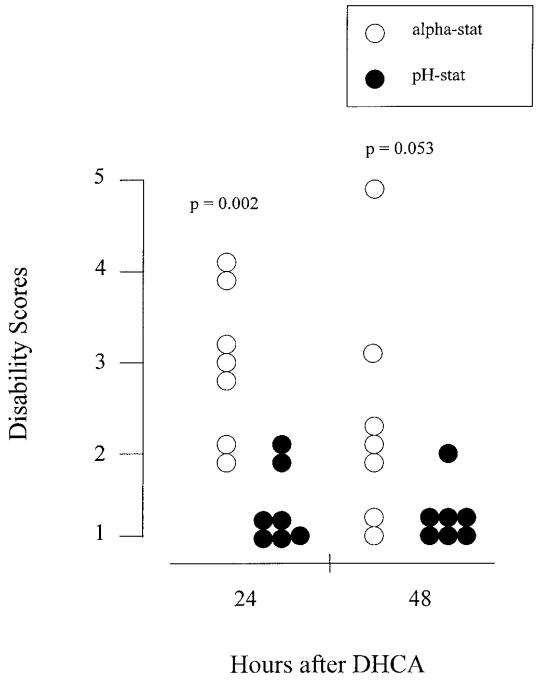

Fig 2. Disability scores after DHCA using alpha-stat or pH-stat CPB: *1* = no disability; *2* = mild disability; *3* = moderate disability; *4* = severe disability; and *5* = death.

(eg, the functional disability score and the histologic grade applied to the neocortex and hippocampus), comparisons between the alpha-stat and pH-stat groups were made with the Mann-Whitney *U* rank tests.

Results

Thirteen piglets survived according to the protocol. One piglet died the second night after DHCA. Table II displays physiologic data before, during, and after CPB for all animals. As intended, arterial pH was lower and arterial P_{CO_2} was higher in the pH-stat group during CPB ($P < .001$). After CPB was discontinued, arterial P_{CO_2} was still higher in the pH-stat group ($P = .02$). The pH-stat group had a lower hematocrit value before CPB ($P = .05$). Table III displays the pig weights, CPB durations, and times to extubation from the onset of CPB reperfusion. Postoperative weights were similar in both groups. CPB cooling, rewarming, and total duration were not different between the groups. Both groups were extubated at similar times.

Twenty-four hours after DHCA, performance ($P = .004$) and disability ($P = .002$) scores were better in the pH-stat group than in the alpha-stat group (Figs 1 and 2). In the pH-stat group, 5 of 7 animals (71%) had no evidence of neurologic disability and the remaining 2 had mild disabilities (mild ataxia). All animals were able to feed themselves, ambulate, and freely explore their surroundings. Forty-eight hours after DHCA, only 1 of 7 animals (14%) had a disability (mild ataxia); the others had fully recovered. In contrast, all piglets in the alpha-stat group had neurologic disability at 24 hours: 28.5% severe (2/7), 43% moderate (3/7), and 28.5% mild (2/7). Forty-eight hours after DHCA, 1 piglet had died of neurologic impairment, 1 remained moderately impaired, 3 had mild disability, and 2 had fully recovered. No animals in either group had seizure activity. One animal in the alpha-stat group had a hemiparesis.

All animals had histologic evidence of neuronal damage. In the alpha-stat group, 3 of 6 animals had areas of pseudolaminar necrosis and 1 had an infarction in the neocortex. No animals in the pH-stat group had areas of necrosis or infarction in the neocortex or hippocampus. The predominant type of neuronal cell death in both groups was apoptosis based on morphologic criteria (nuclear karyorrhexis [fragmented, rounded, dense chromatin] with minimal cytoplasmic change). Apoptotic cell death was less prevalent in the pH-stat group than in the alpha-stat group (Table IV). The pH-

stat group had fewer damaged neurons in the neocortex ($P < .001$) and the CA1 region of the hippocampus ($P = .04$), although the cellular density among the groups was the same. The pH-stat group had lower histologic scores in the neocortex ($P = .035$) and hippocampus ($P = .005$).

Table IV. *Neuronal density and damage in the neocortex and hippocampus of animals subjected to alpha-stat and pH-stat CPB*

	Alpha-stat	*pH-stat*
Cells/mm2		
Neocortex	14 ± 3	16 ± 5
Hippocampus CA1	22 ± 5	21 ± 2
Hippocampus CA3	18 ± 10	22 ± 4
Hippocampus dentate gyrus	19 ± 2	20 ± 3
Percent damaged neurons		
Neocortex	15 ± 7	4 ± 2*
Hippocampus CA1	33 ± 25	11 ± 6†
Hippocampus CA3	16 ± 23	3 ± 5
Hippocampus denate gyrus	3 ± 6	1 ± 1
Histologic scores		
Neocortex	2 ± 0.6	1.1 ± 0.4‡
Hippocampus	2.7 ± 0.5	0.9 ± 0.9§

Values are mean ± SD.
*$P < .001$ versus alpha-stat.
†$P = .04$ versus alpha-stat.
‡$P = .035$ versus alpha-stat.
§$P = .005$ versus alpha-stat.

Discussion

These results demonstrate better neurologic outcome after DHCA with the pH-stat strategy during CPB than with the alpha-stat strategy. Both behavioral and pathologic assessments revealed improved neurologic outcome. On the basis of behavioral assessments, the pH-stat group had less disability and greater neurologic performance postoperatively. On pathologic assessment, the pH-stat group had less neuronal damage in the neocortex and the hippocampus, the only regions consistently damaged in this DHCA model.

Previous studies reported that pH-stat CPB provides better recovery of cerebral physiology after DHCA than does alpha-stat CPB.[7-9,14,18] In the neonatal pig DHCA model, the pH-stat strategy resulted in greater CBF during CPB, less brain water content after CPB, and faster recovery of cerebral high-energy phosphates, cytochrome aa3 oxidation, and intracellular pH homeostasis during reperfusion than the alpha-stat strategy.[7,8,14] The pH-stat strategy also increased cerebral metabolic recovery and CBF compared with the alpha-stat strategy in a piglet DHCA model with aortopulmonary collaterals.[18] However, these studies were nonsurvival models in which surrogate markers of neurologic outcome were used, and conclusions of improved neurologic outcome could only be suggested.

Clinical studies comparing outcome between pH-stat and alpha-stat management have not been conclusive. In a randomized controlled clinical trial of neonates with complex congenital heart disease undergoing cardiac surgery,[16] there was lower postoperative morbidity, shorter recovery time to first electroencephalographic activity, and a tendency toward fewer seizures with the use of the pH-stat strategy. In a subset of infants with transposition of the great arteries undergoing the arterial switch procedures, those treated with pH-stat CPB had significantly fewer episodes of postoperative acidosis and hypotension, shorter durations of mechanical ventilation, and shorter stays in the intensive care unit than those treated with alpha-stat CPB. However, no statistically significant differences in acute neurologic outcome were observed, although this study was underpowered to detect a difference. In a retrospective review of infants undergoing cardiac surgery from 1983 to 1988, alpha-stat CPB management before circulatory arrest was associated with a worse developmental outcome than pH-stat management.[15] Because the comparisons between alpha-stat and pH-stat were historical, the difference in outcome may have arisen from other factors. Prospective studies in adult cardiac surgery found either no difference[19] or worse neuropsychologic outcome[20,21] after the use of the pH-stat technique. The differences between pediatric and adult outcomes may be attributed to the use of moderate hypothermia and lack of circulatory arrest in adult operations and/or different mechanisms of brain damage between infants and adults.

Other work in newborn animal models has found hypercapnia to be neuroprotective against normothermic ischemia. Hypercapnic acidosis by itself does not interfere with brain cellular energy production[22] or neurologic recovery,[23] nor does it lead to neuronal damage.[24] In 7-day postnatal rats exposed to unilateral carotid artery ligation and systemic hypoxia, Vannucci and coworkers[25] found that those treated with 6% carbon dioxide (average arterial P_{CO_2} = 54 mm Hg) demonstrated either no damage or mild atrophy on histopathologic examination at 30 days, whereas control animals (average arterial P_{CO_2} = 26 mm Hg) had more cerebral atrophy and cystic lesions. In a subsequent study,[26] immature rats subjected to hypoxic-ischemic conditions and hypercapnia had better preservation of CBF, higher brain glucose concentrations,

and lower brain tissue lactate concentrations, indicating better oxidative metabolism and cerebral physiologic recovery.

The pH-stat strategy during CPB may protect the immature brain from hypoxic-ischemic damage by several mechanisms. Through increased carbon dioxide, pH-stat increases CBF and cerebral oxygen delivery and may prevent cerebral hypoperfusion during CPB.[7,8,14,27] The pH-stat strategy improves brain-cooling efficiency during CPB in that all regions cool more rapidly and homogeneously.[9] Improved cerebral hypothermia confers better brain protection. The pH-stat strategy also increases CBF during reperfusion after DHCA and therefore may improve brain resuscitation.[7,8] The pH-stat strategy increases cortical oxygen supply before arrest and decreases the rate of cortical oxygen consumption, thereby slowing cerebral deoxygenation during DHCA.[14] Theoretically, pH-stat CPB enhances oxygen delivery during hypothermia, as hypothermia shifts the oxygen-hemoglobin dissociation curve to the left while carbon dioxide shifts the curve to the right. Hypercapnic acidosis suppresses neuronal activity to minimize metabolic demand during DHCA.[27] Hypercapnia may be protective by reducing brain tissue glutamate concentrations[28] and/or brain tissue lactate[29] during ischemia. Hypercapnia decreases lactate production by inhibiting pyruvate dehydrogenase. In vitro studies show that acidosis blunts excitatory amino acid neurotoxicity by inhibiting the *N*-methyl-D-aspartate receptor,[30-32] thereby reducing calcium influx into the cell.[33,34] Of these potential mechanisms, cerebral hypothermia did not contribute to our results because the majority of neuronal damage occurred in the superficial gray matter, closest to where the brain temperature was being monitored. This was true for both groups. Furthermore, in our DHCA model, temperature gradients in the rest of the brain were less than 1°C for both pH-stat and alpha-stat,[9] so that it is unlikely that cerebral hypothermia differences played a role in the hippocampal neuroprotection with pH-stat CPB.

The pattern of brain damage in neonates after DHCA differs from that in children and adults. In adult models of normothermic ischemia, neurons in the hippocampus, cerebellum, striatum, thalamus, amygdala, and neocortex become damaged.[35,36] In non-neonatal DHCA models, the predominant lesion is selective neuronal necrosis and infarction in the cerebellum, striatum, and neocortex.[37] In our neonatal DHCA model, the predominant lesion is neuronal apoptosis in the hippocampus and the gray matter of the cerebral cortex; necrosis and infarction are less frequently observed. In the pH-stat group, the neuropathologic lesions were qualitatively similar to the alpha-stat group, although far fewer. One of the concerns of pH-stat CPB management is the risk for cerebral emboli through increased CBF. Our findings did not support this concern.[5]

The limitations of our study include the closed-chest CPB model, use of a bubble oxygenator, and length of DHCA. The closed-chest CPB model requires ligation of the right carotid artery. Despite carotid artery ligation, the circle of Willis maintains blood flow to both cerebral hemispheres in the piglet.[38] Previous work has shown ligation of one carotid artery does not affect the pattern of neuronal damage in this DHCA model.[5] In clinical practice, the membrane oxygenator has largely replaced bubble oxygenators. Although bubble oxygenators can increase gaseous microemboli, emboli do not appear to be the source of neuronal death in our model.[5] However, if emboli were to contribute to neurologic injury in this model, then pH-stat management would protect against this mechanism of injury, as well as DHCA global ischemia. Although the period of DHCA is usually shorter in clinical practice, we selected 90 minutes for DHCA because this duration of circulatory arrest consistently damages the brain in alpha-stat treatment, making neuroprotective studies possible.

As a surgical technique, DHCA offers many advantages during the repair of complex cardiovascular defects in neonates but poses a risk of ischemic brain damage. Our results demonstrate that pH-stat CPB confers neurologic protection during DHCA. The mechanism of protection is not related to hemodynamics, hematocrit, glucose, or brain temperature.

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