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Hypoglycemia Prevents Increase in Lactic Acidosis During Reperfusion After Temporary Cerebral Ischemia in Rats

Dominique Sappey-Marinier[†], Laureano Chileuitt[‡], Michael W. Weiner^{§,¶}, Alan I. Faden^{††,‡‡}, and Philip R. Weinstein^{‡,*}

[†]Magnetic Resonance Unit, Veterans Administration Medical Center, San Francisco, CA 94121, USA

[§]Department of Medicine, School of Medicine, University of California, San Francisco, CA 94143, USA

[¶]Department of Radiology, School of Medicine, University of California. San Francisco, CA 94143, USA

^{††}Department of Neurology, School of Medicine, University of California. San Francisco, CA 94143, USA

^{‡‡}Department of Neurological Surgery, School of Medicine, University of California. San Francisco, CA 94143, USA

Abstract

Sequential ³¹P and ¹H MRS was used to measure cerebral phosphate metabolites, intracellular pH, and lactate in normoglycemic and hypoglycemic rats during 30 min of complete cerebral ischemia and 5.5 h of reperfusion. These results were correlated with brain levels of free fatty acids (FFAs), excitatory amino acids, cations, and water content at death. The lactate/*N*-acetyl aspartate ratio was not significantly different between groups before or during occlusion. During reperfusion, the ratio was higher in normoglycemic rats from 3 to 85 min ($p \leq 0.05$), and recovery time was faster in hypoglycemic rats (29 vs 45 min; $p = 0.04$), suggesting reduced lactate production and faster recovery of aerobic metabolism. During occlusion, significant but comparable decrease of intracellular pH occurred in each group. Intracellular pH was higher in hypoglycemic rats at 140 min and 260 min of reperfusion. Water content, Na and K⁺ concentrations, and FFA and excitatory amino acid levels were not significantly different between groups, but hypoglycemic rats had less depletion of levels of Mg²⁺ ($p = 0.011$). These results show that hypoglycemia has a limited but potentially beneficial effect on postischemic lactic acidosis.

INTRODUCTION

Reperfusion after cerebral ischemia may be associated with further tissue injury caused by secondary biochemical reactions. Hyperglycemia has been shown to increase ischemic lactic acidosis¹ and cellular damage experimentally^{2–6} and clinically,⁷ whereas hypoglycemic animals have less acidosis than those studied under normoglycemic and hyperglycemic conditions.^{8–10} However, potentially protective effects of insulin-induced hypoglycemia on energy metabolism and outcome are not consistently observed.^{1,11–18} For example, rats made

hypoglycemic by insulin injection before regional ischemia was induced by middle cerebral artery occlusion had smaller infarcts than normoglycemic controls,¹⁹ and chronically hyperglycemic diabetic rats had even larger infarct volumes. In other studies, after focal ischemia produced by middle cerebral artery occlusion, hyperglycemic rats had less acute ischemic neuronal damage than normoglycemic controls, which suggests that hypoglycemia might be deleterious.⁴ During global ischemia in rats, ATP depletion was greater under hypoglycemic than normoglycemic conditions.¹

In this study, we used *in vivo* ³¹P and ¹H MRS to study the time course of changes in energy metabolism during 30 min of temporary global ischemia and 5.5 h of reperfusion in rats with insulin-induced hypoglycemia. In order to assess the effect of ischemic lactic acidosis on delayed cytotoxic tissue injury, these findings were correlated with free fatty acid (FFA) and excitatory amino acid levels, cation concentrations, and water content at death after 5.5 h of reperfusion. Because electroencephalographic (EEG) activity is suppressed and glucose utilization is reduced when the serum glucose level is less than 20 mg/dL, moderate hypoglycemia (serum glucose 30–40 mg/dL) was chosen for study to avoid exacerbation by severe hypoglycemia of effects of ischemia on energy metabolism.¹ The goal of these experiments was to determine whether moderate hypoglycemia improves the metabolic (acute) and neurochemical (delayed) responses to brain ischemia.

MATERIALS AND METHODS

Fourteen Sprague–Dawley rats weighing 280–350 g were fasted for 24 h but had free access to water. Anesthesia was induced with 3% isoflurane; a tracheostomy was performed, and the rats were paralyzed by an i.p. injection of pancuronium (1 mg/kg). After intubation, the rats were mechanically ventilated with a Harvard rodent respirator (Model 683, South Natick, MA, USA). Anesthesia was maintained with 1% isoflurane in a mixture of 30% O₂/70% N₂O.

Temporary complete cerebral ischemia was produced by seven-vessel occlusion.^{20,21} A 3-cm skin incision was made in the midline anterior cervical region. Under a surgical microscope, the omohyoid muscle was transected bilaterally, the external carotid and pterygopalatine arteries were electrocoagulated with bipolar forceps. The basilar artery was exposed microsurgically through a 4-mm diameter, midline craniectomy drilled through the clivus and cauterized. Silastic tubing 0.47 mm in diameter (Dow Corning, Midland, MI, USA) was looped around each common carotid artery and sternomastoid muscle, and the ends were brought out through separate skin perforations to form a snare for remote-controlled arterial occlusion. Scalp, periosteum, and temporal muscles were reflected to eliminate fat and muscle signal from the MRS spectra. A femoral artery and vein were cannulated to monitor blood pressure, sample blood gases and glucose concentrations, and infuse drugs. Atropine (0.1 mg/kg) was administered to reduce tracheal secretions.

The mean arterial blood pressure was recorded continuously with a polygraph data recording system (Grass 79D, Grass Instrument Co., Quincy, MA, USA). Arterial blood glucose concentration was monitored with a glucometer (ACCU-CHECK IIm, Boehringer Mannheim Diagnostic, Rockville, MD, USA). Ventilation variables were adjusted to maintain physiologic blood gas levels. Rectal temperature was monitored and maintained at 37 °C with a water jacket connected to a temperature-regulated circulating water bath. Although it was not feasible to monitor brain or temporalis muscle temperature during these experiments, unpublished data obtained from thermistor probes during complete ischemia without MRS recording showed a consistent 3 °C drop in brain temperature with the rat in the magnet.

Seven rats to be made hypoglycemic received an i.v. injection of insulin (0.0042 units/kg) 45 min before the onset of ischemia; equal volumes of 0.9% saline were infused in the

normoglycemic group. Physiological saline solution was infused at 4 mL/kg/h during the experiments. Ischemia was induced by applying tension to the carotid artery snares. After 30 min, the snares were released to allow reperfusion. The restoration of blood flow was verified by microscopic inspection of vessels after 5.5 h of reperfusion.

Control ^1H and ^{31}P spectra were acquired and physiological variables were monitored for 30 min before and after insulin infusion (10 min per ^1H spectrum and 20 min per ^{31}P spectrum). Immediately before the carotid occlusion, ^1H control spectra were acquired for 10 min. During ischemia and 2 h of reperfusion, ^1H and ^{31}P spectra were alternated every 10 min (five ^1H spectra of 2 min each and one ^{31}P spectrum). For the last 3.5 h of reperfusion, ^1H and ^{31}P spectra were interleaved every 20 min. At the end of the reperfusion period, the brain was rapidly exposed by craniectomy, funnel frozen with liquid nitrogen, and stored at -70°C until specimen blocks containing mostly surface cortex with some underlying white matter were processed for neurochemical analysis.

Before the rat was put in the magnet, a two-turn, 10×14 mm elliptical surface coil was placed over the exposed skull. The coil was double-tuned to ^{31}P at 34.64 MHz and to ^1H at 85.57 MHz. The ^1H and ^{31}P spectra were obtained alternately at 2.0T with a 30-cm, horizontal-bore spectrometer (General Electric, Schenectady, NY, USA). The sensitive volume of the coil utilized is estimated to be 500 mm^3 and the adult rat brain measures approximately 300 mm^3 . Because complete global cerebral ischemia was produced in this model, the entire brain volume observed was subjected to circulatory arrest.

Proton spectra were acquired using semi-selective pulses ($1\bar{I}$) in a phased-cycle, spin-echo sequence ($1\bar{I}-TE-2\bar{I}-TE\text{-acq.}$) to suppress the water signal.^{22,23} The TE of the sequence was set to 81.31 ms. The carrier frequency, which corresponds to the intensity maximum of the excitation profile, was continuously adjusted during the experiment and kept 30 Hz upfield of the chemical shift resonance of the lactate methyl group by reference to the *N*-acetyl aspartate (NAA) peak, set at 2.023 ppm. The evolution delay τ_1 of the binomial sequence ($1\tau-1-1$) was equal to 1.5 ms. The lactate resonance was edited with a second binomial DANTE (delays alternating with nutations for tailored excitation) sequence to invert the lactate C2 proton at 4.11 ppm.²⁴ The DANTE sequence, applied 68 ms after the 90° pulse of the spin-echo sequence, was composed of seven pulses separated by a delay τ_2 of 1.87 ms, giving a selectivity of 30 Hz and a flip-angle of 180° .²⁵ Five ^1H spectra with a repetition time of 1.5 s were accumulated for 2 min each as two alternated blocks of 16 scans with and without application of the DANTE sequence. The delay between scans was 1.5 s, and four dummy scans were performed. To obtain a lipid-free lactate signal, the two proton free-induction decays, accumulated with and without the DANTE sequence, were subtracted. Spectra were processed with convolution difference followed by Gaussian multiplication of the free induction decay to enhance resolution.

Phosphorus spectra were obtained with a one-pulse sequence and collected as 1024 transients using a repetition time of 0.33 s. In these partially saturated conditions, the pulse length giving the maximum signal was set to 9 μs , corresponding to a 30° flip angle. The spectra were processed with convolution difference followed by exponential multiplication of the free induction decay to increase the signal-to-noise ratio.

Brain lactate concentrations obtained from the edited spectra were expressed as a ratio of the peak integral of the lactate methyl group to that of NAA. The integral values of the NAA and lactate methyl peaks were obtained, respectively, from the control ^1H spectrum (without the DANTE sequence) and from the difference spectrum, with a curve-fitting routine (GEM-CAP, General Electric). As measured on phantoms, only half of the lactate methyl group is inverted.²⁶ Therefore, a coefficient of 1.5 was introduced in the calculation of the lactate/NAA ratio.

The lactate/NAA ratio was selected as the most reliable MRS parameter available to estimate changes of tissue lactate levels. Only a 20% decrease in baseline NAA peak areas could be detected during ischemia and no NAA alterations were seen during reperfusion. Creatine peak areas were evaluated as a possible standard, but the signal-to-noise ratios were unacceptably small and the baseline, ischemia, and reperfusion measurements were highly variable. With the 1D binomial sequence utilized, the excitation profile centered on NAA was very sharp while the creatinine resonance was of low intensity.

Phosphorus metabolite concentrations were obtained by fitting the P_i , phosphocreatine (PCr), and the β -ATP resonances with the curve-fitting routine. Intracellular pH (pH_i) was calculated as $6.79 + \log(\delta - 3.29) / (5.29 - \delta)$, where δ is the chemical shift difference between P_i and PCr peaks, given by the position of the corresponding curve-fitted line.²⁷

Biochemical analyses were performed on whole rat brains. Free amino acids were extracted, derivatized, separated, and quantitated by high performance liquid chromatography.²⁸⁻³⁰ The linearity of the detector response was checked with pure standards of each measured amino acid. Lipids were extracted from tissue specimens.^{28,30-33} Cholesterol and total FFAs were separated by thin layer chromatography.³⁴⁻³⁶ The linearity of the detector response was checked with known standards of fatty acid methyl esters. Water content was measured by the wet-dry weight technique. Na^+ , K^+ and Mg^{2+} concentrations were measured with an atomic absorption spectrophotometer (Perkin-Elmer, Norwalk, CT, USA).

The statistical significance of differences in the time course of changes in energy metabolite levels and pH_i was determined by repeated-measures analysis of variance (ANOVA) for comparison within groups and by multiple ANOVA and ANOVA with Bonferroni correction for comparison between groups. The unpaired *t*-test was used to determine the significance of differences in biochemical values. The calculations were performed with SYSTAT software (SYSTAT, Inc., Philadelphia, PA, USA) on an IBM PC and with StatView II (Abacus Concepts, Berkeley, CA, USA) and SuperAnova software (Abacus Concepts) on a Macintosh IIfx computer.

RESULTS

The physiological data and serum glucose levels are presented in Table 1. There were no significant differences in mean arterial pressure, serum pH, pO_2 , or pCO_2 between the hypoglycemic and normoglycemic groups. There were no changes in blood pressure or blood gases after insulin administration in the hypoglycemic group.

The time course of changes in serum glucose is shown in Fig. 1. There were no statistically significant differences between groups in serum glucose during the baseline period. Insulin reduced serum glucose from 112.5 ± 6.4 to 34.9 ± 4.6 mg/dL ($p = 0.0003$) 5 min before occlusion in the hypoglycemic group. Serum glucose remained significantly lower in the hypoglycemic group than in the normoglycemic group at each sampling period, but increased to near physiologic levels (85.4 ± 8.6 mg/dL) in the hypoglycemic group by 220 min of reperfusion. There were no statistically significant changes in serum glucose in the normoglycemic group during the experiment.

Neither the initial external carotid and basilar artery occlusions nor comparable levels of insulin-induced hypoglycemia without vascular occlusion altered baseline metabolite levels measured by ^{31}P (ATP, PCr and P_i) and 1H (lactate, NAA and creatine) MRS from the normal range in normoglycemic, nonischemic controls evaluated in preliminary experiments. There were no statistically significant differences in preischemic measurements in the lactate/NAA ratio of pH_i between groups.

Typical baseline phosphorus and proton spectra are shown in Fig. 2. Contamination from scalp and muscle signal was reduced by surgical retraction of those tissues. Contamination of proton spectra by skull lipids was reduced by the subtraction methods utilized. As shown on the ^1H spectra, careful reflection of scalp and muscle eliminated lipid contamination. The PCr signal observed is typical of a normal ^{31}P brain spectrum rather than a muscle spectrum.

The time course of changes in phosphorus metabolite levels is shown in Fig. 3(a). Within 5 min after occlusion, PCr and ATP rapidly fell to undetectable levels in both normoglycemic and hypoglycemic rats. Because of the time required for the ^{31}P measurement, it was not possible to distinguish the rate of depletion of PCr from that of ATP. P_i rapidly increased to approximately 2.5 times the preocclusion value in each group. There was no significant difference between groups in the extent or time course of changes in phosphorus metabolites during ischemia. Because the β -ATP peak was small initially and disappeared during ischemia, it was not feasible to evaluate changes in intracellular free Mg^{2+} as estimated by shift of this peak.³⁷

The time course of changes in pH_i is shown in Fig. 3(b). During occlusion, pH_i decreased from baseline levels in both the hypoglycemic group (6.47 ± 0.03 vs 7.07 ± 0.04 ; $p \leq 0.0001$) and the normoglycemic group (6.38 ± 0.04 vs 7.09 ± 0.06 ; $p \leq 0.0001$), but the differences between groups were not statistically significant ($p = 0.23$).

The time course of changes in the lactate/NAA ratio is shown in Fig. 4. During the first 10 min of ischemia, the lactate/NAA ratio increased rapidly and progressively in both groups. The ratio reached peak values during the last 10 min of the occlusion, with a trend toward higher values in the normoglycemic group (2.28 ± 0.27 vs 1.84 ± 0.35 ; $p = 0.17$) at the last occlusion time point. During occlusion, the lactate/NAA ratios did not correlate with glucose levels ($p = 0.26$, $r = 0.33$). The glucose values, which began returning toward normal levels early during reperfusion, varied in the hypoglycemic group (Fig. 1).

By 7 min after the onset of reperfusion, the lactate/NAA ratios in the normoglycemic group had increased by 20% from the final occlusion levels ($p = 0.04$); there was no significant change in the hypoglycemic group (Fig. 4). Significantly lower lactate/NAA ratios ($p \leq 0.05$) were observed in the hypoglycemia group for all measurement periods from 3 to 85 min of reperfusion and again at 240 min. The ratio recovered significantly faster in the hypoglycemic rats (29 vs 45 min of reperfusion, $p \leq 0.05$) and remained there until death in both groups. pH_i recovered significantly faster in the hypoglycemic rats (35 vs 75 min of reperfusion,

Statistically significant differences were observed in pH_i between groups at 140 ($p = 0.011$) and 260 min reperfusion ($p = 0.014$). The normoglycemic group was more acidic at both time points. However, these data must be interpreted with caution because serum glucose levels in the hypoglycemia group were returning to baseline at these time points.

The lactate/NAA ratios and serum glucose levels did not correlate significantly during reperfusion. However, there was a trend ($p = 0.06$, $r = 0.51$) toward less lactate production during reperfusion in hypoglycemic rats (Fig. 4). The phosphorus metabolite levels recovered rapidly and equally in both groups; there was a trend toward faster recovery in the hypoglycemic group [Fig. 3(a)].

The ^1H and ^{31}P spectra were stable during the remaining 3.5–4 h of reperfusion after maximum recovery had occurred. There was no evidence of delayed recurrence of metabolic deterioration.

In three hypoglycemic rats that were excluded from the study because serum glucose levels were less than 20mg/dL (10, 12 and 18mg/dL), lactic acidosis persisted and PCr and ATP

failed to recover after ischemia. Although three of the seven hypoglycemic rats included in the study had serum glucose levels of 20–30 mg/dL before occlusion, energy metabolism did recover during reperfusion.

Although a trend toward reduction of arachidonic acid and other individual FFAs and excitatory amino acid levels was observed in the hypoglycemic group, there were no statistically significant differences between groups in cholesterol, total FFAs, or glutamate and other individual amino acid levels after 5.5 h of reperfusion (data not shown). Levels measured in both groups were substantially higher than those observed in nonischemic surgical controls in our laboratory.

There were no significant differences between normo- and hypoglycemic groups in the mean water content [$79.68 \pm 1.19\%$ vs $80.73 \pm 0.16\%$ (SD)], mean Na^+ concentration (255.4 ± 13.4 vs 249.4 ± 40.0 $\mu\text{mol/g}$ dry wt), or mean K^+ concentration (459.3 ± 41.5 vs 444.7 ± 63.1 $\mu\text{mol/g}$ dry wt) of brain. However, tissue Mg^{2+} levels were significantly higher in the hypoglycemic rats (32.00 ± 0.88 vs 28.83 ± 0.76 $\mu\text{mol/g}$ dry wt; $p = 0.011$).

DISCUSSION

The time course of the metabolic effects of temporary complete ischemia in hypoglycemic rats has not been described previously. The major finding of this study was that reperfusion-induced elevation of brain lactate was prevented by insulin administration sufficient to produce moderate hypoglycemia. Our unique observation of increased lactate levels during early reperfusion in normoglycemic but not in hypoglycemic rats is due to increased carbohydrate stores and glucose availability in the untreated group. However, this difference might well have been eliminated during the 30 min of ischemia. One might speculate that rapid restoration of brain blood flow and glucose levels triggers a relative hypermetabolism that exceeds oxygen delivery rates, stimulating anaerobic glycolysis.³⁸ Therefore, preventing or limiting an increase in postischemic lactate production by inducing hypoglycemia before reperfusion could reduce the extent of ischemic metabolic stress and subsequent cellular injury.³⁹ Incomplete recovery of cerebral blood flow and presumably also of O_2 delivery because of the no-reflow phenomenon, which has been shown to be more severe in normoglycemic than hypoglycemic rats, could also prevent normalization of perfusion and energy metabolism immediately after release of occlusion.⁴⁰

Because brain circulation is completely arrested and glucose delivery stops in this ischemia model,⁴¹ lactate levels rapidly reached a plateau in these experiments at levels lower than might be expected with an incomplete global ischemia model, in which some glucose delivery continues.^{1,42} Under nonischemic conditions, brain glucose levels and glycogen stores vary directly with serum glucose levels except during periods of fluctuating blood glucose induced by i.v. glucose infusion.^{43,44} The severity of lactic acidosis due to anaerobic glycolysis after the onset of cerebral ischemia is expected to vary with both the preischemic serum glucose level¹⁰ and the amount of intracellular glucose available, as well as with the completeness or severity of cerebral blood flow reduction during ischemia.⁴² However, since brain glucose and glycogen stores are depleted within 5 min after the onset of complete ischemia,⁴³ any possible benefit of reducing acidosis by pretreatment with insulin might have been difficult to detect during 30 min of complete ischemia, given the time resolution of the ^{31}P MRS methods used in this study. Previously reported observations that brain lactate was 50% lower in insulin-treated rats than in normoglycemic controls¹⁰ may be explained by differences in the methods used to produce ischemia.

Acidosis was assessed as an injury mechanism *in vivo* in previous ^{31}P MRS studies of global ischemia.^{8,45} In cats made hypoglycemic before ischemia, the observed reduction in pH_i was

attenuated both during ischemia and early during reperfusion. The observation, during occlusion, of a pH_i response to hypoglycemia in that study suggests that ischemia was incomplete because energy metabolism responded to manipulation of serum glucose levels.⁴⁰

The observation of significantly higher total tissue magnesium levels in the hypoglycemic group suggests that insulin administration may prevent an ischemia-induced decrease in Mg²⁺ similar to that observed in our previous MRS studies of intracellular Mg²⁺ after traumatic brain injury.³⁷ Normal rat brain values in that study³⁷ were higher (6.37 ± 0.11 $\mu\text{mol/g}$ wet wt) than in the normoglycemic ischemic rats in the present study (5.84 ± 0.12 $\mu\text{mol/g}$). Similarly, a 13% decline in total Mg²⁺ concentration and a 69% decrease in free Mg²⁺ was observed 1 h after fluid-percussion brain injury in our previous experiments.⁴⁶ However, total tissue Mg²⁺ levels measured in tissue that has not been cleared of intravascular blood by perfusion are difficult to interpret. Magnesium is an essential enzyme catalyst for such reactions as oxidative phosphorylation, glycolysis, and synthesis of protein and phospholipids. Reduction of intracellular free Mg²⁺ levels has been correlated in previous studies with direct measurement of total tissue magnesium levels and poor outcome after both brain and spinal cord trauma.^{46,47} Higher Mg²⁺ levels may reflect an improved bioenergetic state that facilitates adenosine diphosphate phosphorylation.⁴⁸ Since high-energy phosphorus metabolite levels were not decreased at the end of reperfusion in these studies, the Mg²⁺ depletion may have been above the threshold for metabolic impairment. The effect of insulin administration without ischemia on Mg²⁺ levels is unknown.

The lack of significant differences in FFA and excitatory amino acid levels between groups 6 h after the onset of ischemia indicates that persistent or delayed lipolytic or excitotoxic reactions as measured at that time point were either absent or not influenced by hypoglycemia during ischemia. Although interstitial levels of glutamate in the neurotransmitter pool are a small component of total tissue levels, which also include the metabolic pool, reduction of total tissue levels has been shown to affect the response to both global and focal ischemia.⁴⁹ Lack of increase in water content as a manifestation of edema in either group was also unexpected.⁶ It is possible that the 3°C drop in brain temperature observed during ischemia in pilot studies with this model could have a protective effect on edema, lipolysis, and excitotoxin release that masked any additional benefit of insulin treatment. Although previous MRS studies after 60 min of temporary global ischemia in normoglycemic rats did demonstrate elevated arachidonate and glutamate levels after 2 h of reperfusion,²⁸ we have no data after 5 h reperfusion or at earlier time points after 30 min of temporary ischemia. The present negative neurochemical observations could also be due to earlier resolution of expected postischemic elevations of arachidonic acid and to persistent decrease without recovery in glutamate levels in both the saline control and insulin-infused groups.^{28,50}

Because endotracheal ventilation is required to prevent hypoxia during prolonged survival in this model, anesthesia could not be reversed to allow neurological assessment during reperfusion. Similarly, histological comparisons that require 24 h survival to accurately assess ischemic neuronal damage were not feasible.

Energy metabolism failed to recover when serum glucose levels were less than 20mg/dL. Therefore, possible dose-dependent, deleterious effects of severe hypoglycemia on postischemic energy metabolism must be avoided if irreversible metabolic injury is to be prevented.^{1,11} The metabolic response to mild hypoglycemia (serum glucose of 70–90 mg/dL) was not evaluated in our study. Other reports indicate that after 20 min of temporary incomplete cerebral ischemia, the neurological grade and 24-h survival rates are higher in rats with serum glucose levels of 65–175 mg/dL than in those with more severe hypoglycemia or hyperglycemia.¹¹ We conclude from our MRS and neurochemical findings that it would be unlikely for a significant cerebral protective effect to be demonstrated for insulin-induced

hypoglycemia in the 20–40 mg/dL range during reperfusion after temporary complete ischemia with our model if histological or neurological outcome studies were to be performed.

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Abbreviations used

ANOVA, analysis of variance; DANTE, delays alternating with nutations for tailored excitation; EEG, electroencephalogram; FFA, free fatty acid; NAA, *N*-acetyl aspartate; PCr, phosphocreatine; pH_i, intracellular pH.

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Figure 1.

Time course of changes in serum glucose levels (mean ± SE) in seven normoglycemic and seven hypoglycemic rats. Insulin induced a significant decrease in glucose (112.5 ± 6.4 to 34.9 ± 4.6 mg/dL; $p = 0.0003$) 5 min before occlusion in the hypoglycemia group. Serum glucose increased nearly to physiologic levels (85.4 ± 8.6 mg/dL) in the hypoglycemic group by 220 min of reperfusion.

Figure 2.

(a) Normal baseline proton brain spectrum (lower right) and stacked ^1H edited spectra obtained sequentially in a normoglycemic rat during 30 min complete global cerebral ischemia and 5.5 h reperfusion are presented. Note increase in size of lactate peak during occlusion with additional increase followed by gradual recovery during reperfusion. (b) Sequential ^{31}P spectra from the same rat demonstrate temporary PCr and ATP depletion and P_i increase during 30 min of global ischemia.

Figure 3.

(a) Time course of changes in MRS peak areas for ATP, PCr and P_i (mean $\pm$ SD) during temporary ischemia in normoglycemic and hypoglycemic rats. All data in parts per million normalized to preischemic values of 1.0. There were no significant differences between the groups.

Figure 3.

(b) Time course of changes in intracellular pH (pH_i) (mean ± SD) measured by ³¹P MRS in normoglycemic and hypoglycemic rats. The difference between groups during ischemia is not statistically significant. However, pH_i recovered to preischemic levels more rapidly (35 vs 75 min of reperfusion; $p < 0.05$) in hypoglycemic than in normoglycemic rats. pH was slightly but significantly higher only at 140 and 260 min of reperfusion ($p = 0.011$ and 0.014 , respectively) in hypoglycemic rats than in normoglycemic rats.

Figure 4.

Time course of lactate/NAA ratios (mean $\pm$ SE) in seven normoglycemic and seven hypoglycemic rats. The lactate/NAA ratio was significantly lower ($p \leq 0.05$) in the hypoglycemia group from 3 to 85 min of reperfusion, and again at 240 min reperfusion.

Table 1

Physiological results in normo- and hypoglycemic rats subjected to 30 min of complete cerebral ischemia followed by 5.5 h of reperfusion (mean $\pm$ SE)

Time of measurement	Blood pressure (mmHg)	pCO ₂ (mmHg)	pO ₂ (mmHg)	pH	Serum glucose (mg/dL)
Normoglycemic (<i>n</i> = 7)					
Surgery	90 $\pm$ 7	26 $\pm$ 6	155 $\pm$ 14	7.34 $\pm$ 0.04	111 $\pm$ 7
Baseline	89 $\pm$ 3	26 $\pm$ 3	153 $\pm$ 9	7.33 $\pm$ 0.02	122 $\pm$ 16
Ischemia	61 $\pm$ 4	25 $\pm$ 3	149 $\pm$ 9	7.28 $\pm$ 0.03	118 $\pm$ 7
Reperfusion					
30 min	84 $\pm$ 3	27 $\pm$ 3	156 $\pm$ 14	7.27 $\pm$ 0.05	104 $\pm$ 5
2 h	77 $\pm$ 3	25 $\pm$ 2	152 $\pm$ 14	7.29 $\pm$ 0.02	105 $\pm$ 5
4 h	79 $\pm$ 5	24 $\pm$ 2	158 $\pm$ 12	7.27 $\pm$ 0.03	108 $\pm$ 9
Hypoglycemic (<i>n</i> = 7)					
Surgery	83 $\pm$ 2	22 $\pm$ 3	191 $\pm$ 16	7.32 $\pm$ 0.02	115 $\pm$ 8
1st baseline	81 $\pm$ 2	26 $\pm$ 2	169 $\pm$ 6	7.29 $\pm$ 0.01	110 $\pm$ 5
2nd baseline	81 $\pm$ 4	26 $\pm$ 2	153 $\pm$ 5	7.29 $\pm$ 0.01	35 $\pm$ 5
Ischemia	60 $\pm$ 5	27 $\pm$ 2	175 $\pm$ 14	7.27 $\pm$ 0.03	43 $\pm$ 7
Reperfusion					
30 min	81 $\pm$ 4	25 $\pm$ 1	173 $\pm$ 14	7.26 $\pm$ 0.04	68 $\pm$ 10
2 h	80 $\pm$ 3	24 $\pm$ 2	170 $\pm$ 9	7.28 $\pm$ 0.03	79 $\pm$ 9
4 h	91 $\pm$ 3	22 $\pm$ 2	163 $\pm$ 9	7.29 $\pm$ 0.03	85 $\pm$ 8