Hyperglycemia Results in an Increase in Myocardial Interstitial Glucose and Glucose Uptake During Ischemia

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The purpose of this investigation was to assess the effects of hyperglycemia, in the absence of changes in plasma insulin and arterial free fatty acid (FFA) levels, on interstitial glucose levels and glucose uptake across the left ventricular wall during ischemia in domestic swine. Insulin secretion was suppressed with a continuous infusion of somatostatin. Arterial FFA levels remained stable due to the suppression of insulin. Microdialysis probes were used to estimate changes in interstitial glucose and lactate, and were placed in the subepicardium and the subendocardium of the left anterior descending ([LAD] ischemic) coronary artery perfusion bed and in the midmyocardium of the circumflex ([CFX] nonischemic) perfusion bed. The LAD coronary artery was cannulated and perfused with blood from the femoral artery through an extracorporal perfusion circuit. Ischemia was induced in the LAD perfusion bed by reducing the flow of the LAD perfusion pump by 60% for 50 minutes, and was followed by 30 minutes of reperfusion. Twenty minutes into the ischemic period, seven animals were given a bolus injection of 50% glucose (200 mg/kg) followed by a glucose infusion (10 mg/kg/min), resulting in an increase in arterial glucose levels from 5 to 13 mmol/L in the hyperglycemic group. Hyperglycemia resulted in a marked increase in dialysate glucose during ischemia and a greater than twofold increase in glucose extraction and uptake. Dialysate glucose correlated with plasma glucose in all three perfusion beds. In conclusion, hyperglycemia, in the absence of an increase in insulin and a decrease in arterial FFA, resulted in a doubling of glucose extraction, delivery, and uptake, which corresponded to the twofold elevation in interstitial glucose during ischemia.

GOODALE AND HACKEL[1] first reported a positive correlation ($r = .79$) between arterial glucose levels and myocardial glucose extraction in hyperglycemic dogs. A similar relationship was observed in acutely hyperglycemic humans.[2] However, it is not possible to determine the independent effects of hyperglycemia on myocardial glucose uptake from these studies, because hyperglycemia resulted in a reflexive increase in plasma insulin concentration and a resultant decrease in arterial free fatty acid (FFA) levels. Insulin and FFAs are independent regulators of myocardial glucose uptake both in vitro and in vivo.[3-8] Insulin stimulates glucose transporter translocation into the sarcolemmal membrane,[8] and also stimulates myocardial glucose uptake indirectly through inhibition of lipolysis in adipocytes, causing a reduction in plasma FFA levels and stimulation of cardiac pyruvate dehydrogenase activity.[3-7] When Barrett et al[3] made normal dogs acutely hyperglycemic in the absence of any major change in insulin or FFAs, there was not a significant increase in myocardial glucose uptake.

The effects of hyperglycemia per se on myocardial glucose uptake in ischemic myocardium is not well understood. Ischemia (~40% to 70% decrease in coronary blood flow) generally results in an increase in myocardial glucose uptake and glycogenolysis as compared with well-perfused conditions.[9-13] We recently observed that a 60% reduction in coronary blood flow results in a significant decrease in myocardial interstitial glucose levels measured by microdialysis.[9] This phenomenon is most likely due to the decrease in myocardial blood flow and glucose delivery during ischemia, suggesting that acute hyperglycemia and the resultant increase in glucose delivery would increase interstitial glucose levels and myocardial glucose uptake. This hypothesis has not been tested experimentally.

The purpose of this study was to assess the effects of hyperglycemia, in the absence of changes in insulin and FFAs, on myocardial interstitial glucose concentration and cellular glucose uptake during ischemia. We hypothesized that hyperglycemia per se would result in elevated interstitial glucose levels, leading to an increase in the arterial venous glucose difference and the rate of glucose uptake. A second aim of the study was to assess the effects of hyperglycemia on interstitial lactate levels during ischemia. Interstitial levels of glucose and lactate were approximated from the effluent emanating from microdialysis probes placed in the myocardium.

MATERIALS AND METHODS

Experiments were performed on 15 overnight-fasted domestic swine (euglycemic group, n = 8, 40.6 ± 2.5 kg; hyperglycemic group, n = 6, 39.4 ± 3.4 kg) in accordance with the Guide for the Care and Use of Laboratory Animals (National Institutes of Health publication no. 85-23) and the Institutional Animal Care and Use Committee at Syntex. The experimental preparation has been described in detail.[9] Animals were initially anesthetized with telazol (12 mg/kg intramuscularly) and medicated with atropine (.017 grains intramuscularly). The animals were ventilated to maintain arterial blood gases in the normal range (Po_2 > 100 mm Hg; Pco_2 = 35 to 45 mm Hg; pH 7.35 to 7.45) and anesthesia was maintained with isoflurane (0.75% to 1.5%). A transthoracotomy with bilateral rib resections was performed for wide exposure of the heart. The animal was then heparinized (20,000 U heparin intravenous bolus followed by 10,000 U/h). The anterior interventricular vein, which drains the perfusion territory of the left anterior descending (LAD) bed, was cannulated and the effluent was allowed to drain into the chest cavity. Blood from the chest cavity was continuously pumped into a reservoir, filtered through a 40-μm filter, and reinfused into the femoral vein. Blood pressure

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was monitored in the left ventricle using a 7F manometer-tipped catheter (Millar Instruments, Houston, TX). A cannula was sutured into the left atrial appendage for the injection of fluorescent microspheres. Blood from the femoral artery was withdrawn via a perfusion pump (Gilson Medical Electronics, Middleton, WI), passed through a 40-μm filter, and introduced into the LAD artery. The LAD perfusion pump rate was adjusted to achieve coronary venous oxygen saturation of approximately 40%. Ischemia was induced in the LAD perfusion bed by reducing the flow of the LAD perfusion pump to 40% of the control value. The perfusion system allowed us to control flow to the LAD bed independently of flow to the circumflex (CFX) bed, so that ischemia was induced in the LAD bed while flow to the CFX bed remained normal.

Microdialysis

Microdialysis probes (CMA/20; Bioanalytical Systems, West Lafayette, IN) were placed diagonally into the subepicardium and the subendocardium of the LAD (ischemic) perfusion bed and the midmyocardium of the CFX (nonischemic) region with the aid of an introducer needle. The probe in the nonischemic CFX perfusion bed was placed in the midmyocardial layer because the blood flow to this layer is stable and does not differ from either the adjacent subendocardial or subepicardial layers. Krebs-Henseleit buffer was perfused through each microdialysis probe at a rate of 2 μL/min with a syringe infusion pump.[9] The exchange area of interstitial fluid and the microdialysis probes has been estimated to be 3 mm.[14] The effluent from each probe (which we refer to as the dialysate) traveled through the outflow tubing and into capped microcentrifuge tubes. Before each experiment, the linearity in glucose recovery of each microdialysis probe was checked in vitro as described previously.[9]

Protocol

The protocol used in the present study is shown in Fig. 1. All animals were allowed to stabilize for a period of 50 minutes following insertion of the microdialysis probes. A somatostatin infusion (0.8 μg/kg/min; Bachem, Torrance, CA) was started into the femoral vein 10 minutes before the beginning of the protocol, to suppress insulin release and decrease plasma insulin to approximately 2 to 5 μU/mL from normal values of approximately 12 μU/mL.[9] Each experiment began with 30 minutes of aerobic flow to the LAD bed starting at time zero. At 30 minutes, ischemia was induced in the LAD bed by reducing the flow on the LAD perfusion pump to 40% of the aerobic flow. Thus, during the 50 minutes of ischemia only, the LAD bed was ischemic while perfusion of the CFX bed remained normal. At 50 minutes (20 minutes into the ischemic period), seven of 15 animals received a bolus injection of 50% glucose (200 mg/kg) in the femoral vein. This was followed by an infusion of 50% glucose at a rate of 10 mg/kg/min. At 80 minutes, flow to the LAD bed was increased again to aerobic flow for a period of 30 minutes in all animals. Heart rate, LAD mean pressure, left ventricular peak systolic and end diastolic pressures, and left ventricular peak dp/dt were recorded throughout the experiment. Microdialysis samples were collected from the LAD subepicardium and subendocardium and the CFX midmyocardium. Arterial and venous blood samples were simultaneously drawn from the femoral artery (upstream of the LAD perfusion pump) and the anterior interventricular vein every 10 minutes beginning at 5 minutes into the aerobic period. Fluorescent microspheres (15-μm diameter; Molecular Probes, Eugene, OR) were used to assess regional blood flow with injections made into the left atrium at times 20, 70, and 105 minutes over a 15-second period. A reference sample for calculation of blood flow was withdrawn from the femoral artery using a calibrated syringe withdrawal pump. At completion of the study, tissue samples for microsphere flow determinations were taken from the tissue surrounding each microdialysis probe in the LAD subepicardium, LAD subendocardium, and CFX midmyocardium, as well as samples from the LAD midmyocardium and the CFX subepicardial and subendocardial regions.

Analytical Methods

Dialysate and blood glucose and lactate levels were measured using commercially available enzymatic methods (Sigma, St Louis, MO) as previously described.[2,9,16] Blood samples for glucose and lactate analysis were immediately deproteinized in ice-cold 6% perchloric acid (1:2 vol/vol), weighed, centrifuged, and analyzed in quadruplicate for glucose and triplicate for lactate. Arterial plasma samples were analyzed in duplicate for glucose. Plasma FFAs were assayed in duplicate using an enzymatic spectrophotometric method.[9] Regional myocardial blood flow was assessed as described previously.[9,17]

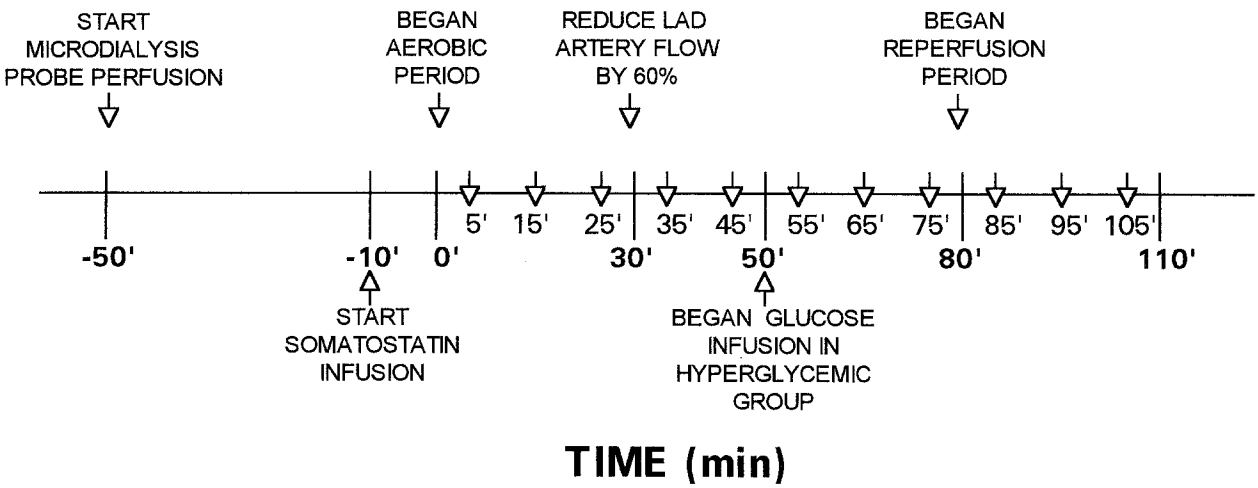

Fig 1. Time line for the study protocol. (▽) Time of withdrawal of arteriovenous samples from the femoral artery and anterior interventricular vein.

Calculations

Regional myocardial blood flow was calculated using the reference withdrawal method as previously described.[9,17] Glucose delivery to the LAD perfusion bed was calculated as the product of arterial glucose and LAD pump flow. Glucose uptake was calculated as the product of the arteriovenous glucose difference and LAD pump flow. FFA uptake was calculated as the product of the arteriovenous FFA difference and LAD pump flow.

Statistics

Overall analyses were made using a two-way repeated-measures ANOVA with terms for group, period, and their interaction. Pairwise comparisons within group and group within period were made using Fisher's least-significant difference test on the adjusted means. Correlation coefficients were calculated by linear regression analysis. Slopes for the line of best fit for dialysate glucose concentration plotted as a function of plasma glucose concentration were compared among the CFX midmyocardium, LAD subepicardium, and LAD subendocardium using an analysis of covariance. All results are presented as the adjusted mean ± SE. All comparisons with P less than .05 were deemed significant.

RESULTS

Hemodynamics

Hemodynamic measurements for both the euglycemic and hyperglycemic groups are presented in Table 1. In both groups, ischemia resulted in a decrease in left ventricular peak dp/dt, LAD bed MVO_2, and LAD mean arterial pressure as compared with the aerobic periods. Glucose infusion did not have any significant effect on hemodynamic parameters.

Myocardial Blood Flow

Table 2 summarizes the results of myocardial blood flow measurements. Due to technical problems and the fact that one animal in each group died upon reperfusion, blood flow was measured in only five of seven hyperglycemic animals during the aerobic and ischemic periods, and in four hyperglycemic and five euglycemic animals during reperfusion. Myocardial blood flow in the ischemic LAD midmyocardium and subendocardium was significantly less during the ischemic period compared with both the aerobic and the reperfusion periods in both groups. Furthermore, during the ischemic period there was a transmural gradient in blood flow in both groups, with the most severe reduction occurring in the subendocardium. Blood flow to the CFX perfusion bed remained unchanged throughout ischemia.

Dialysate Glucose

Euglycemic group. LAD subepicardial dialysate glucose was not significantly different between any of the four periods in the euglycemic group, despite a 60% reduction in flow during the 30- to 80-minute ischemic period (Fig 2A). LAD subendocardial glucose in the euglycemic group was significantly higher during the aerobic period compared with the 30- to 50-minute and 50- to 80-minute ischemic periods (Fig 2B). Dialysate glucose in the CFX midmyocardium (nonischemic bed) did not change throughout all four periods in the euglycemic group (Fig 3).

Hyperglycemic group. The hyperglycemic group received a bolus injection of glucose (200 mg/kg) at 50 minutes, followed by a continuous infusion (10 mg/kg/min) into the femoral vein. Within the hyperglycemic group, dialysate glucose was significantly higher during reperfusion in all three regions compared with the aerobic period, the 30- to 50-minute ischemic period, and the 50- to 80-minute ischemic period (Figs 2A and B and 3). Furthermore, dialysate glucose was significantly higher during the 50- to 80-minute ischemic period compared with both the aerobic

Table 1. Hemodynamic and Contractile Responses in the LAD Perfusion Bed

Response	Aerobic Period (0 to 30 min)	Ischemic Period (30 to 50 min)	Ischemic Period (50 to 80 min)	Reperfusion Period (80 to 110 min)
HR (beats/min)				
Euglycemic	98 ± 8 (8)	95 ± 6 (8)	99 ± 8 (8)	101 ± 7 (7)
Hyperglycemic	125 ± 12 (7)	111 ± 13 (6)	117 ± 14 (7)	120 ± 12 (6)
LV peak systolic pressure (mm Hg)				
Euglycemic	80.3 ± 3.4 (8)	82.0 ± 4.0 (8)	81.8 ± 3.2 (8)	78.8 ± 2.8 (7)
Hyperglycemic	94.2 ± 9.7 (7)	81.3 ± 5.5 (7)	82.4 ± 6.6 (7)	105.5 ± 23.7 (6)
LV end diastolic pressure (mm Hg)				
Euglycemic	4.7 ± 1.0 (8)*†	7.3 ± 1.9 (8)	8.0 ± 2.0 (8)	4.2 ± 1.1 (7)†
Hyperglycemic	4.8 ± 0.6 (7)	5.6 ± 0.6 (7)	5.6 ± 0.3 (7)	5.5 ± 0.7 (6)
LV peak dp/dt (mm Hg/s)				
Euglycemic	1,525 ± 119 (8)*	1,257 ± 107 (8)	1,291 ± 129 (8)	1,282 ± 126 (7)
Hyperglycemic	1,960 ± 236 (7)*†	1,511 ± 233 (7)	1,513 ± 208 (7)	1,930 ± 532 (6)*†
LAD bed $M\dot{V}o_2$ (mL/min)				
Euglycemic	3.05 ± 0.44 (8)*	2.15 ± 0.23 (8)		2.19 ± 0.25 (7)
Hyperglycemic	3.38 ± 0.47 (7)*	2.36 ± 0.26 (7)		2.34 ± 0.43 (6)
LAD MAP (mm Hg)				
Euglycemic	82.1 ± 4.6 (8)*†	29.1 ± 3.9 (8)	30.1 ± 3.8 (8)	69.8 ± 8.5 (7)*†
Hyperglycemic	94.5 ± 6.6 (7)*†	30.3 ± 1.2 (7)	27.5 ± 1.6 (7)	70.8 ± 7.6 (6)*†

NOTE. Values are the mean ± SE. Numbers in parentheses denote the n. Significance results taken from pairwise comparisons using Fischer's LSD.

Abbreviations: HR, heart rate; LV, left ventricular; MAP, mean arterial pressure.

*Significantly different from 30 to 50-minute ischemic period, $P < .05$.

†Significantly different from 50 to 80-minute ischemic period, $P < .05$.

Table 2. Regional Myocardial Blood Flow (mL · g^{-1} · min^{-1})

Region	Aerobic Period	Ischemic Period	Reperfusion Period
LAD BED			
EPI			
Euglycemic	1.08 ± 0.19 (8)*	0.70 ± 0.12 (8)	1.04 ± 0.15 (5)*
Hyperglycemic	0.97 ± 0.26 (5)	0.59 ± 0.19 (5)	0.97 ± 0.26 (4)
MID			
Euglycemic	0.92 ± 0.16 (8)*	0.53 ± 0.15 (8)	1.12 ± 0.28 (5)*
Hyperglycemic	0.85 ± 0.13 (5)*	0.55 ± 0.13 (5)	1.15 ± 0.34 (4)*
ENDO			
Euglycemic	1.01 ± 0.17 (8)*	0.35 ± 0.08 (8)†	1.31 ± 0.29 (5)*
Hyperglycemic	0.85 ± 0.13 (5)*	0.27 ± 0.05 (5)†	0.76 ± 0.20 (4)*
CFX BED			
EPI			
Euglycemic	1.14 ± 0.23 (8)	1.22 ± 0.23 (8)	1.02 ± 0.16 (5)
Hyperglycemic	1.34 ± 0.38 (5)	1.37 ± 0.47 (5)	1.13 ± 0.39 (4)
MID			
Euglycemic	0.89 ± 0.19 (8)	1.06 ± 0.19 (8)	0.95 ± 0.20 (5)
Hyperglycemic	0.97 ± 0.36 (5)	1.04 ± 0.40 (5)	0.89 ± 0.38 (4)
ENDO			
Euglycemic	0.82 ± 0.15 (8)	1.12 ± 0.19 (8)	1.06 ± 0.21 (5)
Hyperglycemic	0.77 ± 0.17 (5)	1.20 ± 0.44 (5)	0.57 ± 0.17 (4)

NOTE. Values are the mean ± SE. Numbers in parentheses denote the n.

Abbreviations: EPI, subepicardium; MID, midmyocardium; ENDO, subendocardium.

*$P < .05$ *v* ischemic period.

†$P < .05$ *v* subepicardium.

and 30- to 50-minute ischemic period in all three regions (Figs 2 and 3).

Comparing euglycemic and hyperglycemic groups within periods, dialysate glucose in the hyperglycemic group was significantly higher in all three perfusion beds during the 50- to 80-minute ischemic period and reperfusion period (Figs 2A and B and 3).

Dialysate Lactate

Euglycemic group. There were no significant differences in either the CFX midmyocardium (1.2 ± 0.25, 1.09 ± 0.27, 1.25 ± 0.29, and 1.4 ± 0.27 for control, 30- to 50-minute ischemic, 50- to 80-minute ischemic, and reperfusion periods, respectively) or the LAD subepicardium (1.53 ± 0.32, 1.97 ± 0.21, 1.85 ± 0.25, and 1.81 ± 0.19 for control, 30- to 50-minute ischemic, 50- to 80-minute ischemic, and reperfusion periods, respectively) in the euglycemic group. However, LAD subendocardial dialysate lactate was significantly greater during both ischemic periods (3.45 ± 0.53 and 3.46 ± 0.53 mmol/L in the 30- to 50-minute and 50- to 80-minute ischemic periods, respectively) compared with the aerobic period (1.49 ± 0.40 mmol/L).

Hyperglycemic group. Dialysate lactate in the CFX midmyocardium was significantly greater in the 50- to 80-minute ischemic period (2.70 ± 0.73) and reperfusion period (3.26 ± 0.74) compared with the aerobic period (2.07 ± 0.46). LAD subepicardial dialysate lactate was significantly greater in the 30- to 50-minute ischemic period (3.26 ± 0.87 mmol/L), the 50- to 80-minute ischemic period (3.97 ± 1.05), and the reperfusion period (3.97 ± 1.01) compared with the aerobic period (2.48 ± 0.57). LAD subendocardial dialysate lactate was also significantly greater in both ischemic periods (3.95 ± 0.50 and 5.10 ± 0.82 mmol/L for the 30- to 50-minute and 50- to 80-minute ischemic periods, respectively) compared with the aerobic period (2.14 ± 0.44).

Compared with the euglycemic group, the hyperglycemic group exhibited significantly higher dialysate lactate concentrations in the LAD subepicardium and CFX midmyocardium during all periods, and greater dialysate lactate levels in the LAD subendocardium after glucose infusion.

Glucose, Lactate, and FFA Metabolism

Glucose, lactate, and FFA measurements are presented in Table 3. Glucose extraction, percent glucose extraction,

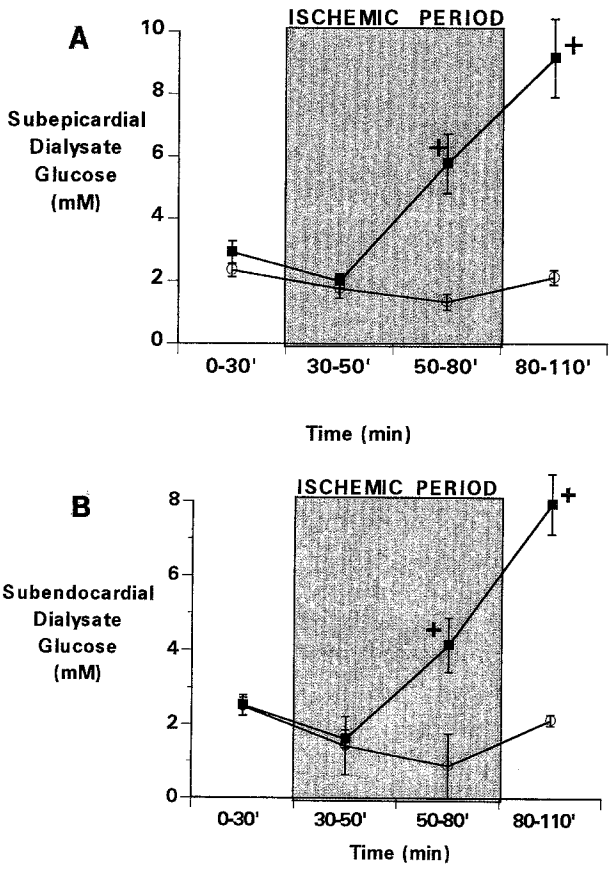

Fig 2. (A) Dialysate glucose in the LAD EPI in the euglycemic (EUG, ○) and hyperglycemic (HYP, ■) groups. Results are the mean ± SE within each period; n = 8 and 7 for euglycemic and hyperglycemic groups, respectively, through the 50- to 80-minute ischemic period, and n = 7 and 6 for the reperfusion period. †$P < .05$ *v* EUG group. (B) Dialysate glucose in the LAD ENDO in the EUG (○) and HYP (■) groups. Results are the mean ± SE within each period; n = 8 and 7 for EUG and HYP groups, respectively, through the 50- to 80-minute ischemic period, and n = 7 and 6 for the reperfusion period. †$P < .05$ *v* EUG group.

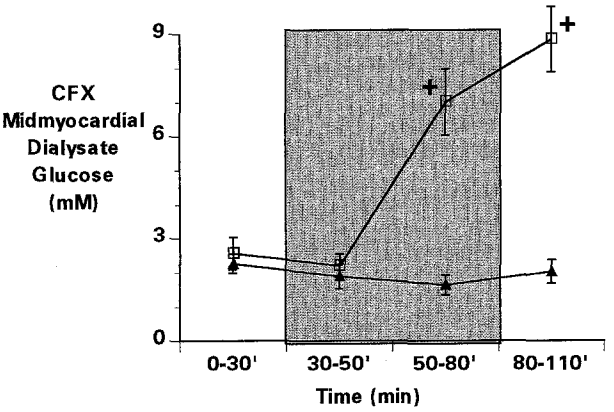

Fig 3. Dialysate glucose in the CFX MID. EUG dialysate glucose (▲); HYP dialysate glucose (□). The CFX perfusion bed served as a control bed within each animal; therefore, blood flow to the CFX midmyocardium remained at aerobic values throughout the 110-minute protocol. Results are the mean ± SE within each period; n = 8 and 7 for EUG and HYP groups, respectively, through the 50- to 80-minute ischemic period, and n = 7 and 6 for the reperfusion period. †$P < .05$ *v* EUG group.

and glucose uptake were significantly increased during the 50- to 80-minute ischemic period compared with the aerobic period in both groups. Glucose extraction, delivery, and uptake during the 50- to 80-minute ischemic period were significantly greater in the hyperglycemic group compared with the euglycemic group. Percent glucose extraction was not significantly different between hyperglycemic and euglycemic groups during any period. Percent glucose extraction was significantly greater during the 50- to 80-minute ischemic period compared with the 30- to 50-minute ischemic period in both groups. Net lactate extraction during the aerobic period switched to net lactate production during ischemia in both groups. Plasma insulin was not different between groups throughout the aerobic and ischemic periods.

Linear Regression Analysis

Correlation coefficients for dialysate glucose plotted as a function of plasma glucose were all highly significant (Fig 4), demonstrating that dialysate glucose increases as plasma glucose increases. The slope of the line of best fit was significantly higher in the nonischemic CFX bed (.555) than in the ischemic LAD subendocardium (.318) or subepicar-

Table 3. Glucose, Lactate, and FFA Metabolism in the LAD Perfusion Bed

Parameter	Aerobic Period (0 to 30 min)	Ischemic Period (30 to 50 min)	Ischemic Period (50 to 80 min)	Reperfusion Period (80 to 110 min)
Plasma arterial glucose (mmol/L)				
Euglycemic	5.15 ± 0.30	4.37 ± 0.46	3.77 ± 0.35	4.88 ± 0.32
Hyperglycemic	5.36 ± 0.63‡	4.49 ± 0.49	13.17 ± 1.27*†	17.10 ± 1.91*†‡
Glucose extraction (mmol/L)				
Euglycemic	0.00 ± 0.05*‡	0.27 ± 0.05	0.55 ± 0.12*	0.17 ± 0.06‡
Hyperglycemic	−0.03 ± 0.02‡	0.20 ± 0.07	1.33 ± 0.24*†	0.29 ± 0.12‡
Percent glucose extraction				
Euglycemic	0.13 ± 0.96‡	7.21 ± 1.66	16.30 ± 3.80*	3.68 ± 1.39‡
Hyperglycemic	−0.45 ± 0.39‡	4.19 ± 1.32	10.46 ± 1.72*	1.55 ± 0.55‡
Glucose delivery (μmol/min)				
Euglycemic	215.6 ± 18.4*‡	73.0 ± 7.5	64.2 ± 6.0	191.0 ± 15.5*‡
Hyperglycemic	240.6 ± 39.2*	83.5 ± 15.6	244.7 ± 27.3*†	724.7 ± 95.4*†‡
Glucose uptake (μmol/min)				
Euglycemic	0.39 ± 2.58‡	6.40 ± 1.40	12.26 ± 2.96	8.86 ± 3.32
Hyperglycemic	−2.13 ± 1.16‡	5.36 ± 2.42	31.43 ± 6.65*†	14.88 ± 6.62*‡
Arterial lactate (mmol/L)				
Euglycemic	1.82 ± 0.30	1.77 ± 0.29	1.76 ± 0.28	2.06 ± 0.30
Hyperglycemic	2.98 ± 0.55†‡	3.23 ± 0.70†	4.02 ± 0.98†	5.18 ± 0.24*†
Net lactate extraction (mmol/L)				
Euglycemic	0.54 ± 0.08*‡	−1.13 ± 0.45	−0.34 ± 0.25*	0.20 ± 0.05*
Hyperglycemic	0.53 ± 0.10*‡	−0.73 ± 0.19	−0.65 ± 0.24	0.33 ± 0.07*‡
Arterial FFA (mmol/L)				
Euglycemic	0.53 ± 0.05	0.56 ± 0.05	0.53 ± 0.06	0.49 ± 0.07
Hyperglycemic	0.68 ± 0.04†‡	0.64 ± 0.05†	0.54 ± 0.06*	0.47 ± 0.07*‡
FFA extraction (mmol/L)				
Euglycemic	0.09 ± 0.02	0.11 ± 0.02	0.12 ± 0.03	0.06 ± 0.01
Hyperglycemic	0.08 ± 0.01*	0.19 ± 0.03	0.12 ± 0.02*	0.05 ± 0.03*‡
Plasma insulin (μU/mL)				
Euglycemic	3.69 ± 1.06	3.91 ± 1.06	3.80 ± 1.06	3.74 ± 1.13
Hyperglycemic	3.03 ± 1.22	2.83 ± 1.22	4.88 ± 1.22	11.35 ± 1.34*†‡

NOTE. Values are the mean ± SE; n = 8 and n = 6 for euglycemic and hyperglycemic groups, respectively, except during reperfusion, when n = 7 and n = 5.

*$P < .05$ *v* 30 to 50-minute ischemic period.

†$P < .05$ *v* euglycemic group.

‡$P < .05$ *v* 50 to 80-minute ischemic period.

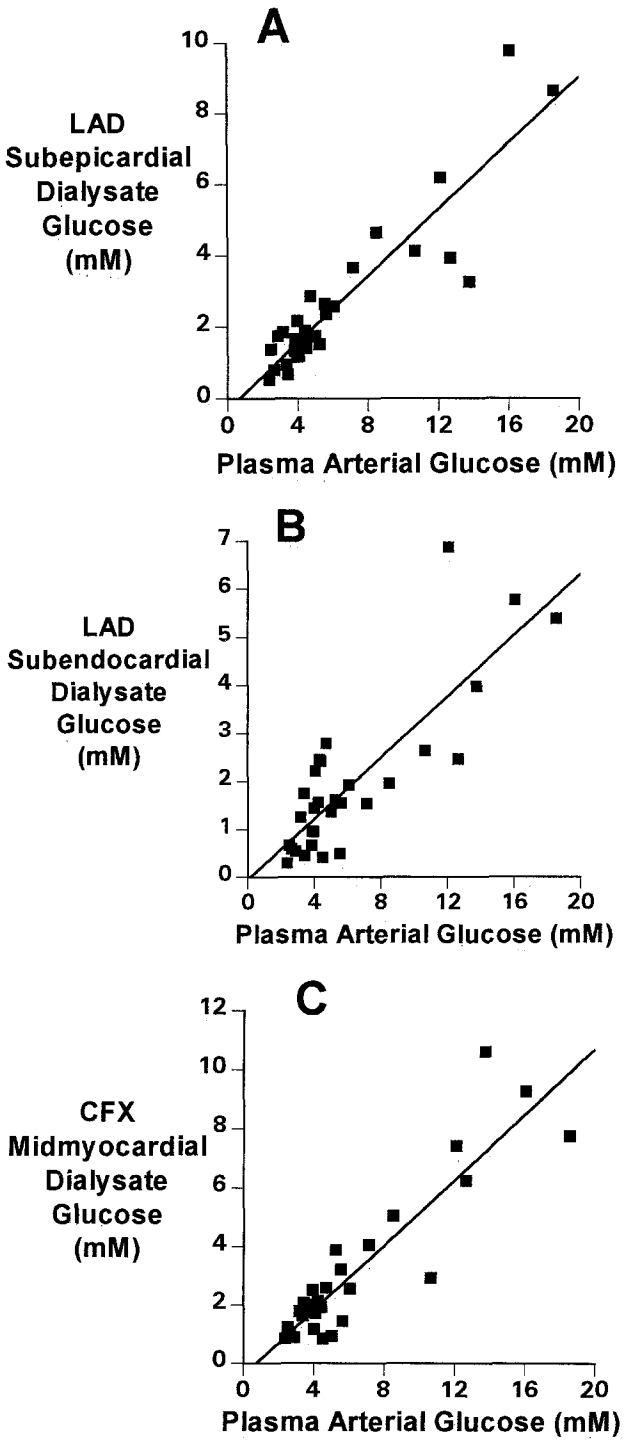

Fig 4. Dialysate glucose plotted as a function of plasma arterial glucose during both ischemic periods for both EUG and HYP groups. (A) LAD EPI dialysate glucose (r = .91, y = 0.47x − 0.31). (B) LAD ENDO dialysate glucose (r = .83, y = 0.32x − 0.49). (C) CFX MID dialysate glucose (r = .91, y = 0.55x − 0.41). The slope of the line of best fit was significantly higher in the nonischemic CFX bed (0.55) than in the ischemic LAD subendocardium (0.32) or subepicardium (0.47), and was significantly lower in the LAD subendocardium than in the LAD subepicardium.

dium (.468). In addition, dialysate glucose concentration was significantly lower in the LAD subendocardium during ischemia than in the LAD subepicardium. These results show that for a given plasma glucose concentration, interstitial glucose decreased progressively with decreasing blood flow (Fig 4).

Dialysate glucose in the LAD perfusion bed was significantly correlated with glucose delivery (LAD pump flow · arterial glucose concentration) during the ischemic period (r = .74 and r = .75 for the subepicardium and subendocardium, respectively). Equations for the lines of best fit for both subepicardial and subendocardial dialysate glucose as a function of glucose delivery were y = 0.019x + 0.458 and y = 0.014x + 0.32, respectively. Glucose extraction also correlated significantly with dialysate glucose in both the LAD subepicardium and the LAD subendocardium (r = .60 and r = .53 for subepicardium and subendocardium, respectively).

DISCUSSION

The results of the present investigation demonstrate that hyperglycemia results in a greater than twofold increase in myocardial interstitial glucose, glucose extraction, and glucose uptake during ischemia despite there being no change in arterial FFA and plasma insulin. In 1953, Goodale and Hackel[1] demonstrated that glucose extraction was positively correlated with arterial glucose levels in normal dogs (r = .79); however, insulin and FFA levels were not controlled in their study. Barrett et al[3] found that hyperglycemia (~10 mmol/L) did not significantly increase myocardial glucose uptake in dogs treated with somatostatin to suppress insulin secretion under normal flow conditions; however, the effects of ischemia were not assessed. Russell and Oliver[15] showed a significant increase in glucose extraction during coronary artery occlusion in hyperglycemic dogs with uncontrolled insulin secretion. Hyperglycemia resulted in an increase in glucose uptake in isolated perfused rabbit hearts compared with a control group with similar FFA and insulin concentrations under ischemic conditions.[18] The present investigation is the first to show that hyperglycemia results in increased glucose extraction in ischemic myocardium under conditions of stable FFA and insulin levels. Furthermore, the increase in glucose extraction corresponded with an increase in myocardial interstitial glucose levels, as estimated by microdialysis.

Interstitial glucose levels appear to depend largely on myocardial blood flow and arterial glucose, as evidenced by the significant correlations between arterial glucose and interstitial glucose levels (Fig 4). Furthermore, comparing the slope of the lines of best fit for dialysate glucose and plasma arterial glucose (.55, .47, and .32 for the CFX nonischemic midmyocardium, LAD subepicardium, and LAD subendocardium, respectively) illustrates that for a given plasma arterial glucose concentration during ischemia, interstitial glucose levels are highest in the nonischemic region and lowest in the most ischemic region (Fig 4). Interestingly, although glucose infusion resulted in significantly elevated interstitial glucose levels and arteriovenous glucose difference, when the arteriovenous glucose differ-

ence was expressed as a percent of the arterial glucose extraction ([arteriovenous glucose extraction/arterial plasma glucose] · 100), there were no significant differences between the euglycemic and hyperglycemic groups. This suggests that the fractional clearance of glucose by ischemic myocardium is unchanged by acute hyperglycemia.

Varying results have been reported for net lactate production by the myocardium in response to hyperglycemia.[15,18] Russell and Oliver[15] found no significant difference in net lactate production during ischemia in hyperglycemic dogs in vivo. Eberli et al[18] reported increased flux through glycolysis in hyperglycemic rabbits, resulting in a significant increase in net lactate production compared with a normoglycemic group. In the present study, interstitial lactate levels were significantly elevated in the most ischemic LAD subendocardium in the hyperglycemic group after glucose infusion. However, net lactate release was not different between groups during ischemia, despite significantly greater glucose uptake rates in the hyperglycemic group, suggesting that the rate of anaerobic glycolysis was not accelerated by hyperglycemia. These results suggest that hyperglycemia during ischemia causes an increase in the rate of myocardial oxidation of exogenous glucose. Studies with carbon-labeled glucose tracers need to be performed to determine definitively the metabolic fate of glucose extracted by the myocardium.[2,10,16]

During reperfusion, insulin levels increased to values normal for swine[19] in the hyperglycemic animals despite administration of somatostatin, suggesting that an increase of arterial glucose levels overrides the effects of somatostatin on suppressing insulin release. Despite the increase in plasma insulin in the hyperglycemic group, neither glucose uptake nor glucose extraction were significantly different between groups during reperfusion. Glucose uptake and glucose extraction were increased in both groups during reperfusion compared with the aerobic period. Myears et al[12] have shown that canine myocardium reperfused after 1 hour of ischemia exhibits enhanced glucose uptake and impaired utilization of palmitate. The mechanism resulting in increased glucose utilization following ischemia is not clear. It is possible that (1) glucose transport is increased by a migration of glucose transporters to the plasma membrane,[20] (2) glycolytic enzyme activity is increased, and/or (3) β-oxidation of fatty acids is inhibited.

There are several limitations to this study that should be addressed. Limitations of the cardiac microdialysis method have recently been addressed in detail.[9,21] Interpretation of the present investigation is constrained by the fact that we only manipulated two of the primary regulators of myocardial glucose uptake: plasma glucose and blood flow. To understand the determinants of myocardial glucose uptake fully, one must also independently manipulate insulin and FFA levels. Before regulation of myocardial glucose uptake in vivo is fully understood, future experiments will need to address the independent roles of blood flow, plasma FFA, and insulin on myocardial glucose uptake.

Hyperglycemic animals tended to have a greater left ventricular peak pressure and peak dp/dt during reperfusion; however, this trend was not statistically significant (Table 1). The present investigation was not designed with sufficient statistical power to address this question properly. It is possible that hyperglycemia and increased myocardial glucose uptake, in the absence of changes in insulin and FFA, result in improved postischemic function following moderate reductions in coronary blood flow. This question merits further investigation.

In conclusion, hyperglycemia, in the absence of an increase in insulin and a decrease in FFA, resulted in a greater than threefold increase in interstitial glucose levels in the nonischemic myocardium. In the ischemic myocardium, hyperglycemia resulted in a twofold elevation in interstitial glucose concentration, which corresponded to a doubling of glucose extraction and glucose uptake rates.

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