Glycolytic buffering affects cardiac bioenergetic signaling and contractile reserve similar to creatine kinase

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Harrison, Glenn J., Michiel H. van Wijhe, Bas de Groot, Francina J. Dijk, Lori A. Gustafson, and Johannes H. G. M. van Beek. Glycolytic buffering affects cardiac bioenergetic signaling and contractile reserve similar to creatine kinase. Am J Physiol Heart Circ Physiol 285: H883–H890, 2003. First published April 24, 2003; 10.1152/ajpheart.00725.2002.—Creatine kinase (CK) and glycolysis represent important energy-buffering processes in the cardiac myocyte. Although the role of compartmentalized CK in energy transfer has been investigated intensely, similar duties for intracellular glycolysis have not been demonstrated. By measuring the response time of mitochondrial oxygen consumption to dynamic workload jumps (t_{mito}) in isolated rabbit hearts, we studied the effect of inhibiting energetic systems (CK and/or glycolysis) on transcytosolic signal transduction that couples cytosolic ATP hydrolysis to activation of oxidative phosphorylation. Tyrode-perfused hearts were exposed to 15 min of the following: 1) 0.4 mM iodoacetamide (IA; n = 6) to block CK (CK activity <3% vs. control), 2) 0.3 mM iodoacetic acid (IAA; n = 5) to inhibit glycolysis (GAPDH activity <3% vs. control), or 3) vehicle (control, n = 7) at 37°C. Pretreatment t_{mito} was similar across groups at 4.3 ± 0.3 s (means ± SE). No change in t_{mito} was observed in control hearts; however, in IAA- and IA-treated hearts, t_{mito}, decreased by 15 ± 3% and 40 ± 5%, respectively (P < 0.05 vs. control), indicating quicker energy supply-demand signaling in the absence of ADP/ATP buffering by CK or glycolysis. The faster response times in IAA and IA groups were independent of the size of the workload jump, and the increase in myocardial oxygen consumption during workload steps was unaffected by CK or glycolysis blockade. Contractile function was compromised by IAA and IA treatment versus control, with contractile reserve (defined as increase in rate-pressure product during a standard heart rate jump) reduced to 80 ± 8% and 80 ± 10% of baseline, respectively (P < 0.05 vs. control), and significant elevations in end-diastolic pressure, suggesting raised ADP concentration. These results demonstrate that buffering of phosphate metabolites by glycolysis in the cytosol contributes appreciably to slower mitochondrial activation and may enhance contractile efficiency during increased cardiac workloads. Glycolysis may therefore play a role similar to CK in heart muscle.

glycolysis; energy transduction; mitochondria; regulation of oxidative phosphorylation


Compared with CK, glycolysis has received little attention as an integral part of myocyte energy transfer and signaling. Previous studies (41) have illustrated a degree of functional compartmentalization of glycolytic metabolism and energy stores plus the preferential use of glycolytic rather than oxidatively produced ATP by the sarcolemma. Whereas anaerobic glycolysis may only produce as little as 3–7% of the total ATP under aerobic conditions in ex vivo preparations (8, 17), its contribution to cellular bioenergetics may increase significantly during ischemia and hypoxia (31). In addition, ATP from glycolysis may be used in the heart early during dynamic workload changes (7), as observed previously in skeletal muscle (24).

We can measure the time course of oxygen consumption (Vo2) in response to pacing-induced workload steps, which reflects the transcytosolic energy signaling speeds between myofibrils and ion pumps and the mitochondria in isolated rabbit hearts to match ATP synthesis to hydrolysis (7, 37, 38). The mitochondrial delay time (t_{mito}) is sensitive to altered exogenous substrate (35) and ischemia (46). Recently, we (10) observed that inhibition of CK led to a dose-dependent quickening in t_{mito} combined with a loss of isovolumic contractile reserve, findings that argued against obligatory cytosolic energy transfer via CK rather suggesting that CK is an active ADP/ATP buffer that locally increases the effectiveness of ATP consuming processes, but thereby effectively slows the signal from the myocardial and myocyte bioenergetics is centered on

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sites of ATP hydrolysis to oxidative phosphorylation. Similarly, the local glycolytic buffering near ion pumps and myofibrils might directly enhance contractile function (41). We investigate here whether such buffering by glycolysis slows the activation of oxidative phosphorylation in a similar way as the CK system.

The aim of the present experiments was therefore to test the effect of inhibiting glycolysis alone, or in combination with CK, on contractile function and cytosolic signaling speeds in steady state and during three levels of dynamically increased cardiac workload. This inhibition was achieved by the differential infusion of the sulfhydryl-enzyme blockers iodoacetamide (IA) and iodoacetic acid (IAA), used previously by us (10) and by others (15, 23, 32–34) to preferentially reduce CK and GAPDH activity, respectively. On the basis of our preliminary findings (36), we predicted that the removal of ATP and ADP buffering capacity of glycolysis alone or in combination with CK inhibition would speed up signaling to oxidative phosphorylation at the expense of a compromised ability to perform acute increases in myocardial workload. Such findings, compared with similar data for CK, would define the role of glycolysis in the family of local energy buffering, energy transfer, and signaling systems within the myocyte that determine cardiac function during health and disease.

METHODS

Isolated heart preparation. All experiments were approved by the local animal ethics and experimentation authority (Dier Experimentes Commissie-Free University). Eighteen male New Zealand White rabbits (2.98 ± 0.09 kg) were premedicated by an intramuscular injection of midazolam (1.5 mg/kg Dormicum; Roche) before induction of anesthesia with 0.1 mg/kg im fentanyl citrate and 3 mg/kg im flunaisone (Hynpnom; Janssen Pharmaceutica). Anesthesia was supplemented if necessary with intramuscular fentanyl (0.03 mg/kg). Animals were artificially ventilated before a median sternotomy was performed, and hearts were cannulated in kg). Animals were artificially ventilated before a median sternotomy was performed, and hearts were cannulated in

Calculation of $t_{\text{mito}}$. Detailed description of the techniques, including the oxygen transport model and the equations, assumptions, and correction values, is to be found elsewhere (37, 38). Briefly, the venous mean response time ($t_v$) is integrated from the time course of the venous PO$_2$ ($P_{\text{VO}_2}$) curve. Upon step to and from a higher heart rate. Previous studies have shown that there is an initial overshoot in rate-pressure product (RPP) before steady state is reached, which is accounted for by the RPP response time ($t_{\text{RPP}}$) and also a small initial increase in ($P_{\text{VO}_2}$) [initial deflection time ($t_{\text{d}}$)] due to transient increases in venous outflow, and hence $t_v$ is corrected for these parameters. To correct $t_v$ for delays in diffusion and transport of oxygen between the mitochondria and the oxygen electrode, we subtract the transport time ($t_{\text{transport}}$), obtaining the true response time of VO$_2$ at the level of the mitochondria during a dynamic step in workload: $t_{\text{mito}} = t_v - t_{\text{transport}}$. We therefore use $t_{\text{mito}}$ as an index of intracellular energy transfer/signaling speed during rapid increases in metabolic demand in our isovolumic preparation.

$t_{\text{transport}}$ is calculated from the venous O$_2$ response times to a combination of three experimental interventions conducted in series following the heart rate steps (37). First, a small step in arterial concentration (ACS) is made by instantaneously increasing immediately above the heart of 10% of the normal oxygenated Tyrode with identical Tyrode gassed with 95% N$_2$-5% CO$_2$ (PO$_2$ = 30 mmHg at coronary ostia) and the venous O$_2$ response to this step is assessed. Intravascular transit time is calculated with the use of a indicator-dilution step with Evans blue bound to albumin (EBS) as the intravascular indicator, infused immediately above the heart and detected in a densitometer adjacent to the venous oxygen electrode. $t_{\text{transport}}$ is derived from the ACS with a numerically somewhat smaller contribution from the EBS, but a second estimate is independently obtained from a step in perfusion flow (PFS) made by rapid extraction of ~10% of the normal coronary flow from a side arm in the aortic cannula. All ACS, EBS, and PFS steps are conducted at both intrinsic pacing rate (132 beats/min) and the highest test heart rate (220 beats/min) to assess the effect of heart rate on oxygen transport and to check the sensitivity of the preparation to small reductions in oxygen supply. These steps are repeated after the treatment to assess the effects of IAA and IA on transport. The technique has been tested and compared with other techniques such as near-infrared spectroscopy, nuclear magnetic resonance (NMR) spectroscopy (7), and heart rate measurements as extensively discussed in a review (37).

Experimental protocol. All hearts were equilibrated for 30 min after instrumentation, followed by assessment of baseline hemodynamic function. During the next 30 min the first
series of steps to calculate \( t_{\text{mito}} \) was performed (as outlined in Calculation of \( t_{\text{mito}} \)) incorporating randomized heart rate steps from basal heart rate (132 beats/min) to 160, 190, and 220 beats/min and back, plus the ACS, EBS, and PFS steps at 132 and 220 beats/min. Hearts were then randomly assigned to one of three treatment groups to receive 0.4 mM IA (IA treated, \( n = 6 \)), 0.3 mM IAA (IAA treated, \( n = 5 \)), or vehicle (Tyrode buffer) infusion (control hearts, \( n = 7 \) ) over the next 15 min. Concentrations given are final values (in mM) in the perfusing solution after infusion of stock solutions into a side arm of the aortic cannula at \(-1.2 \) ml/min with an infusion pump (Vickers Medical; Hampshire, UK). Hearts were subsequently allowed 15 min for drug washout and reequilibration before the second series of \( t_{\text{mito}} \) steps. Measurements of contractile function were made before and after IA or IAA infusion. Contractile reserve was defined as the increase in RPP (heart rate \( \times \) left ventricular developed pressure) for a 132–220 beats/min heart rate step made before and after treatment. At the end of experiments, a piece (\(-1 \) g) of left ventricle was rapidly excised into isopentane (precooled in liquid N\(_2\)) and immediately freeze-dried at \(-80^\circ\)C for biochemical analysis. The remaining heart was trimmed of extraneous tissue and used for blotted wet and dry (48 h at \(80^\circ\)C) weight measurements.

Biochemical assays. Tissue samples (5–10 mg) were cut from the freeze-dried heart sections and homogenized at 4°C for 20 s in 0.1 M potassium phosphate buffer containing 1 mM EDTA and 1 mM \( \beta \)-mercaptoethanol (pH 7.4). Triton X-100 was added to the homogenate at a final concentration of 0.1% before measurement of total CK activity coupled to NADH production at 25°C with the use of an absorbance wavelength of 340 nm (2). Adenylate kinase (AK) and GAPDH activities were assayed also at 25°C according to methods described by Bergmeyer (2). All enzyme activities are reported as international units (1 IU = \( \mu \)mol/min)/mg of dry heart weight tissue. All chemical reagents used were of analytical grade and were obtained from Sigma (St. Louis, MO) or Boehringer Mannheim (Mannheim, Germany).

The effect of IAA and IA on mitochondrial state 3 \( V_o^2 \) after the addition of 1 mM ADP was determined in a respirometer containing isolated rabbit heart mitochondria with 5 mM pyruvate and 2 mM malate as carbon sources.

Statistical analysis. All data are presented as means \( \pm \) SE except where indicated otherwise. Comparisons among treatment groups were made using one-way ANOVA with the Newman-Keuls post hoc test used to examine specific differences between group means if ANOVA had reported significance. Measurements made before and after treatment within groups were compared using the Student’s paired \( t \)-test or ANOVA for repeated measures as necessary. A value of \( P < 0.05 \) was considered statistically significant for all comparisons.

RESULTS

Biochemical evaluation of glycolytic/CK inhibition. Table 1 outlines the activities of CK, GAPDH, and AK in tissue homogenates taken from control, IAA-, and IA-treated hearts. Both IA and IAA are alkylating agents that affect cysteine residues on many enzymes. GAPDH is known to be very sensitive to IAA (23) and is the usual point of inhibition of glucose metabolism. We (10) and Tian and Ingwall (34) have used IA to cause graded inhibition of CK in the 0.1–0.4 mM concentration range. In the present study, 0.4 mM IA caused a 98% reduction in total CK activity and a concomitant reduction in GAPDH to 8% of control heart values. IAA at 0.3 mM was more effective against GAPDH (2% of control remaining) versus CK (80% of control). Because of the reserve in the CK system, CK activity alone has to be reduced to <15% of control to cause any changes in contractile function and energy signaling speeds as shown previously (10, 34). IAA-treated hearts are therefore glycolytically blocked but CK operable. The AK reaction has also been implicated in cardiac energy transfer (4, 5, 25) but its role was not studied here. AK activity was 20 times lower than CK activity in control hearts and was unaffected by IA or IAA treatment. With the use of this combination of only partially selective chemical inhibitors, the effects of CK and/or glycolytic blockade can be separated in the isolated perfused heart.

Effect of IA and IAA on contractile function. There was no difference in the wet (7.7 \( \pm \) 0.4 g wet wt) and dry (1.3 \( \pm \) 0.1 g dry wt) heart weights or in the initial coronary flow rates (12.6 \( \pm \) 0.4 ml \( \cdot \) min\(^{-1} \cdot \) g wet wt\(^{-1} \)) between the three groups of hearts. Table 2 summarizes the isovolumic contractile indexes measured in hearts from the three treatment groups measured immediately before infusion of the test compound and after the 15-min washout period. As shown, there was no difference in any parameter between groups’ pretreatment, but significant systolic and diastolic impairment after inhibition of glycolysis. Systolic left ventricular pressure was significantly decreased in the IAA group, whereas end-diastolic pressure was elevated in both the IA- and IAA-treated hearts, findings consistent with our and others previous use of these compounds (10, 15, 32, 34) and potentially reflecting an increase in the free cytosolic [ADP]. Contractile function stabilized after the washout period and no significant deterioration was observed during the second series of heart rate steps.

Contractile reserve in our preparation is defined as the relative increase in RPP during a step in heart rate from 132 to 220 beats/min, measured before and after treatment. Pretreatment contractile reserve was 3,385 \( \pm \) 293, 5,243 \( \pm \) 1,062, and 5,475 \( \pm \) 702 mmHg/min in control, IAA, and IA heart groups, respectively \( (P > 0.05) \). Contractile reserve was significantly reduced with the use of IAA (4,092 \( \pm \) 777 mmHg/min) and IA (4,239 \( \pm \) 652 mmHg/min) treatment (\(-80% \) of pretreatment; \( P < 0.05 \)), a finding reported previously.
for CK inhibition (10, 34) but not for glycolytic blockade. Contractile reserve in control hearts was well maintained at ~109% of pretreatment values (3,583 ± 172 mmHg/min).

Reduction in contractile reserve was apparently independent of changes in oxidative metabolism of the hearts: the MV02 increases during the 132–220 beats/min heart rate step for control, IAA-, and IA-treated hearts changed nonsignificantly posttreatment at −9.5 ± 2.2%, −4.7 ± 3.5%, and −7.0 ± 1.5%, respectively. ttransport, which characterizes oxygen transport, was not affected by the IA or IAA infusion (see Table 3). We have previously shown in mitochondria isolated from 0.4 mM IA-treated hearts that state 3 and 4 V02 stimulated with ADP is unaffected, suggesting no damage to oxidative phosphorylation in these hearts (10). State 3 respiration in isolated mitochondria was not affected by addition of up to 1.5 mM IA or up to 1 mM IAA, appreciably higher concentrations than used during perfusion. These data therefore show that oxidative metabolism and oxygen transport in the heart is not compromised by the IA and IAA infusion.

Taken together, the above observations show that contractile function can be effectively maintained at low cardiac workloads without appreciable CK and/or glycolytic activity but the ability of hearts to undergo rapid increases in performance is compromised, reflecting the important role of these energy systems in buffering local ATP levels.

### Energy signaling speeds in absence of glycolysis or CK

We estimate the delay time in ATP hydrolysis-to-synthesis coupling from the time course of VO2 during three levels of submaximal workload elevations by pacing hearts from baseline (132 beats/min) to 160, 190, and 220 beats/min and back. This index, tmito, is calculated from the (PV02) curve (t~9–12 s) and corrected for tpp (−1 to 3 s), tdd (−1 to 2 s), and ttransport (−4 to 6 s) (see METHODS). From Table 3 it can be seen that neither pre- nor posttreatment ttransport values differ among groups and there is a small, similar posttreatment increase in ttransport across the groups (~12%). Table 4 gives the mean pretreatment tv values for control, IAA, and IA heart groups during heart rate steps from 132 to 160, 190, and 220 beats/min, respectively. tv is calculated during both the upward and downward heart rate step, but there was no difference (P > 0.05 data not shown), and thus the average is given. Two-way ANOVA revealed that there was no pretreatment difference between heart groups at any heart rate; however, tmito significantly increased in all groups with increasing heart rate (P < 0.05), with a parallel prolongation in the tpp values from 1.3 ± 0.1 s (160 beats/min) to 2.1 ± 0.2 s (220 beats/min), a finding observed previously (6). The dd did not differ significantly with heart rate either before or after treatment in any group.
Posttreatment changes in \( t_{\text{mito}} \) (see Fig. 1) show that inhibition of glycolysis alone (IAA) or in combination with CK (IA) causes a significant quickening in the response time of ATP synthesis to a step in energy utilization compared with the unchanged \( t_{\text{mito}} \) values over time in control hearts. The faster \( t_{\text{mito}} \) in the IAA and IA groups was independent of the size of the workload transition tested, with all heart rate steps being significantly different from pretreatment \( (P < 0.05; \text{two-way ANOVA}) \). In addition, the change in pre- to posttreatment \( t_{\text{mito}} \) observed in IA-treated hearts was significantly larger than that in the IAA group.

**DISCUSSION**

The current study evaluated the effects of inhibiting two potentially important energy-buffering systems, namely, CK and glycolysis, on the dynamics of activation of oxidative phosphorylation during pacing-induced workload transitions in isolated rabbit hearts. The findings illustrate that glycolytic bypass by giving pyruvate alone led to smaller \( t_{\text{mito}} \) values (Fig. 2). This ATP synthesis/hydrolysis coupling rate was further accelerated when glycolysis was chemically inhibited more fully and also when CK was additionally blocked. Both observations were independent of the size of the workload step tested (Fig. 1) and any apparent loss of oxidative capacity. The loss of ADP/ATP buffering near ion pumps and myofibrillar ATPase's lead to a loss of contractile reserve during elevated myocardial performance states, despite quicker activation of oxidative...
phosphorylation and the ability of hearts to maintain low levels of cardiac function. This was observed previously for CK inhibition (10, 15, 34) but not for glycolytic blockade.

Acute inhibition of CK and GAPDH was achieved using the combination of the alkylating compounds IA and IAA. We (10) and others (15, 23, 32–34) have shown that controlled infusion can provide irreversible blockade of desired energetic pathways with maintained preparation stability. In mitochondria isolated from IA-treated hearts, we have found no loss of oxidative capacity analyzed by ADP-stimulated state 3 and 4 $V_{O_{2}}$ (10) and isolated mitochondrial oxidative capacity was not affected by the IA and IAA concentrations used (see RESULTS). Moreover, by using CK histochemistry, we have observed homogeneous patterns of CK inhibition within tissue slices and myocytes from IA- and IAA-treated hearts (10), indicating that 15 min of infusion allows complete cellular distribution of IA.

The activity of another putative energy transfer enzyme, AK, remained unchanged in both IA and IAA-treated hearts (see Table 1 and Refs. 10 and 34). Acute chemical inhibition of CK gave the same result as targeted gene deletion of CK in knockout mice (9, 10).

Blocking fatty acid usage, which is the preferred myocardial substrate in vivo, increases $t_{mito}$ in an ex vivo heart muscle preparation (45). The present ex vivo preparation, with glucose and pyruvate but no fatty acids as exogenous substrate, is therefore not representing the normal in vivo situation, but is appropriate to study the effect of glycolysis on $t_{mito}$, because it can be supplied with a high level of glucose and glycolysis is subsequently inhibited, whereas pyruvate provides for sustained energy metabolism.

Contractile function in absence of CK and/or glycolysis. Isolated hearts in the present and other studies (10, 15, 32–34) treated with IA can maintain low to medium workloads with CK activity <10% of baseline. This extends to MM and MM/mito CK knockout mice and rats with phosphocreatine replaced with $\gamma$-guanidinopropionate in vitro (29) and in vivo (22). Unchanged CK fluxes are observed during substrate deprivation (12), hypoxia/cyanide infusion (20), or threefold increases in work by pacing or catecholamine stress (16).

The diastolic dysfunction indicated by EDP increases in IA and IAA hearts, respectively (see Table 2) may reflect elevated free [ADP] near myofibrils as measured using $^{31}$P NMR in IA-treated rat hearts (34). However, due to the importance of bound, compartmentalized CK in the myocardium (39, 44), increases in measured [ADP] may not reflect changes in subcompartments (20). Interestingly, blockade of glycolysis with IAA decreased left ventricular systolic pressure and increased end-diastolic pressure at baseline heart rate more than CK inhibition with IA, potentially pointing to the importance of glycolytic buffering of ATP near myofibrils in maintaining contractile function. Interestingly, if binding of glycolytic enzymes to the contractile elements in insect flight muscle is genetically disturbed, the capacity to fly in these animals is severely reduced (43).

The compromised contractile reserve observed after CK inhibition in the present study confirms previous observations in response to chronotropic (10), inotropic (34), and pressure-volume (15) cardiac work elevations. The report of functional loss due to glycolytic blockade is new to our knowledge. The equivalent reduction of contractile reserve in both IA- and IAA-treated hearts (Table 2) suggests the preferential use of glycolytically produced ATP during rapidly increased workloads. Such a function during dynamic exercise has been described for skeletal muscle (24). The reduced contractile reserve in hearts void of energetic buffers may be due to increased ADP affecting Ca$^{2+}$ handling (32–34). The free energy of ATP hydrolysis is directly reduced after CK inhibition, affecting sarcoplasmic reticulum Ca$^{2+}$ pumps (34) and CK blockade abolishes the increased cytosolic Ca$^{2+}$ transient during inotropic stimulation (33).

Energy signaling speeds after CK and/or glycolysis inhibition. The significant reductions in $t_{mito}$ observed in response to glycolytic blockade alone and in combination with CK provides evidence that both of these energetic pathways act as cytosolic buffers that slow the metabolic signal to the mitochondria in response to dynamically varying ATP hydrolysis. The reduction in $t_{mito}$ with 0.4 mM IA treatment in the present study (40 ± 5%) compared favorably with that seen in our previous study (46 ± 6%) (10). The reduction in $t_{mito}$ by preferential glycolytic inhibition by IAA demonstrates the contribution of glycolysis in energy transfer, which was suggested previously (36, 37).

Pyruvate was added to the perfusate in the present experiments to provide oxidizable substrate in the absence of glycolysis. Positive inotropism of pyruvate has been demonstrated both in isolated myocytes (19) and intact hearts (11, 18, 47), causing improved cytosolic phosphorylation potential, redox state, and augmented cytosolic free Ca$^{2+}$ transients under normal workload conditions. In the current study, pyruvate increased left ventricular systolic pressure at baseline heart rate versus glucose-only hearts (113 ± 5 vs. 90 ± 3 mmHg; $P < 0.05$) (10). Pretreatment $t_{mito}$ Values in the present experiments with glucose + pyruvate are significantly shorter than for glucose-only hearts, as shown in Fig. 2 (4.3 ± 0.3 vs. 6.5 ± 0.5 s; $P < 0.05$) (10). We have previously observed pyruvate alone to lower $t_{mito}$ at 28°C versus glucose (35), but this was not in combination with glucose. This effect is probably not due to altered mitochondrial aerobic capacity, because the latter does not affect $t_{mito}$ (37).

Phosphofructokinase in inhibited by elevated citrate caused by pyruvate infusion (18) and GAPDH and pyruvate kinase action are also antagonized by pyruvate (42). Indeed, 2.5 mM pyruvate outcompetes 5 mM $[^{13}$C]glucose, thereby limiting glycolysis to producing <2% of oxidative metabolism (3). Thus $t_{mito}$ appears to be quickened both by the inhibiting effects of pyruvate itself on glycolysis and by further iodoacetic acid blockade (Fig. 2).

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In conclusion, there exists a complex system of bioenergetic buffering and signal transduction that includes not only the energy transfer enzymes CK (27, 28, 40) and AK (4, 25) but also the glycolytic pathway (3, 41). All of these systems affect the complex regulation of oxidative phosphorylation from the cytosol. The present findings show that inhibition of glycolytic buffering of phosphate metabolites (ADP/ATP/Pi) near ion pumps and myofibrils compromises contractile reserve during increased workloads and accelerates the transcytosolic energy signal for oxidative phosphorylation. The quicker ATP hydrolysis/synthesis coupling speeds during inhibition of glycolysis were further shortened when CK was additionally inhibited, without further deterioration of contractile reserve. We suggest that the faster energy transfer speeds observed during our dynamic workload steps in the absence of glycolytic buffering, alone or in combination with CK, reflects a function of glycolysis as a complementary temporal energy buffer in the cytosol that helps maintain myocardial contractility.

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DISCLOSURES

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