Downregulation of Peroxisome Proliferator–Activated Receptor-α Gene Expression in a Mouse Model of Ischemic Cardiomyopathy Is Dependent on Reactive Oxygen Species and Prevents Lipotoxicity

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**Background**—The peroxisome proliferators–activated receptor-α (PPARα), a transcription factor that modulates fatty acid metabolism, regulates substrate preference in the heart. Although in acute ischemia there is a switch in substrate preference from fatty acids to glucose, metabolic gene expression in repetitive ischemia is not well described. In a mouse model of ischemic cardiomyopathy induced by repetitive ischemia/reperfusion (I/R), we postulated that downregulation of PPARα is regulated by reactive oxygen species and is necessary for maintaining contractile function in the heart.

**Methods and Results**—Repetitive closed-chest I/R (15 minutes) was performed daily in C57/BL6 mice, mice overexpressing extracellular superoxide dismutase, and mice treated with the PPARα agonist-WY-14,643. Echocardiography, histology, and candidate gene expression were measured at 3, 5, 7, and 28 days of repetitive I/R and 15 and 30 days after discontinuation of I/R. Repetitive I/R was associated with a downregulation of PPARα-regulated genes and both myosin heavy chain isoform transcript levels, which was reversible on discontinuation of I/R. Overexpression of EC-SOD prevented the downregulation of PPARα-regulated genes and myosin iso-genes by repetitive I/R. Furthermore, reactivation of PPARα in mice exposed to repetitive I/R worsened contractile function, induced microinfarctions, and increased intramyocardial triglyceride deposition, features suggestive of cardiac lipotoxicity.

**Conclusions**—Metabolic and myosin isoform gene expression in repetitive I/R is mediated by reactive oxygen species. Furthermore, we suggest that downregulation of PPARα in repetitive I/R is an adaptive mechanism that is able to prevent lipotoxicity in the ischemic myocardium. (Circulation. 2005;112:407-415.)

**Key Words:** free radicals ■ hibernation ■ ischemia ■ metabolism ■ reperfusion

In response to pressure overload or hypoxia, the heart switches its substrate preference from fatty acids to glucose.1,2 This substrate switch is caused by the downregulation of peroxisome proliferators–activated receptor-α (PPARα) gene expression and activity.3 PPARα is a nuclear transcription factor that regulates the expression of nearly all proteins and enzymes modulating fatty acid uptake and oxidation.3 We have previously shown that pressure overload–induced hypertrophy is associated with a downregulation of PPARα and that pharmacological reactivation of PPARα worsens contractile function, suggesting that substrate switching in the stressed heart may preserve contractile function.1

Acute total ischemia is characterized by a transient switch in myocardial metabolism to anaerobic glycolysis.4 In situations of chronically reduced coronary blood flow or after repetitive brief myocardial ischemia/reperfusion (I/R), there is reversible contractile dysfunction associated with increased use of glucose,5,6 a phenomenon called myocardial hibernation. Although short-term studies have described some features of myocardial metabolism during hibernation,7 metabolic gene expression in this condition is not well described.

We have previously characterized a murine model of ischemic cardiomyopathy induced by repetitive I/R.8 In this model, repetitive I/R leads to the development of a reversible ischemic cardiomyopathy associated with interstitial fibrosis and ventricular dysfunction but without myocardial infarction,8 features resembling human hibernating myocardium.5 In this study, we investigated whether PPARα gene expression is altered in repetitive I/R. Because ventricular dysfunction and fibrosis in this model are dependent on reactive...
oxygen species (ROS), we determined whether ROS also regulates metabolic and myosin iso-gene expression. We found that repetitive I/R was associated with a downregulation of PPARα-regulated genes and myosin iso-gene expression. Downregulation of PPARα-regulated genes and myosin iso-gene expression was regulated by ROS. Unexpectedly, reactivation of PPARα in repetitive I/R was associated with worsened contractile function, microinfarctions, and increased intramyocardial triglyceride deposition, features characteristic of cardiac lipotoxicity.

**Methods**

**Brief Repetitive I/R**

All animals received humane care in compliance with the Guide for the Care and Use of Laboratory Animals (NIH publication 85–23, revised 1985). Wild-type (WT) C57/BL6 mice were obtained commercially (Harlan Sprague-Dawley), and extracellular superoxide dismutase (EC-SOD) mice were developed as previously described. Heterozygous EC-SOD and WT mice underwent initial surgery at 8 to 10 weeks of age using a closed-chest model of I/R. Briefly, an 8–0 Prolene (Ethicon) suture was placed around the left anterior descending artery; both ends of it were threaded through a piece of PE-10 plastic tube (Becton Dickinson), exteriorized through the thorax wall, and stored subcutaneously. After 7 to 9 days of recovery, the skin was reopened, and the ends of the suture were attached to heavy metal picks. The picks were pulled apart until ST elevation occurred in ECG. The 15-minute ischemia was followed by reperfusion (resolution of ST elevation) until the next day. This brief repetitive I/R was performed daily for 3, 5, 7, and 28 days in WT and EC-SOD mice. For the recovery studies, mice underwent a 7-day I/R protocol, and the animals stayed in the vivarium for 15 and 30 days. For the PPARα reactivation studies, C57/BL6 mice were fed the specific PPARα agonist WY-14,643 in powdered chow at a concentration of 0.01% wt/wt for 8 days (1 day before first I/R until the end of I/R). Sham animals underwent the initial surgery only and waited for the same period of time as I/R groups.

**Gene Expression**

The methods for RNA extraction and quantitative RT-PCR have been described previously. The nucleotide sequences for primers and probes have also been previously published. The transcript for the constitutive gene cyclophilin was used as housekeeping gene for data normalization for human studies. Cyclophilin gene expression did not change with I/R. Internal standards were prepared using the T7 RNA polymerase method (Ambion).

**Echocardiography**

Echocardiographic measurements were performed 3 to 4 hours after the last ischemic episode with an 8-MHz probe (Sequoia C256, Acuson) as previously described. The measurements were performed on 3 different images of each mouse, and calculations were done using previously described methods.

**Histology**

Hearts were fixed in zinc-formalin and embedded in paraffin (Z-fix, Anatech). Sections were made at 200-μm intervals from base to apex and stained with hematoxylin and eosin for the initial evaluation. The area below the suture was identified; then, serial sections were stained with picrosirius red to identify collagen as previously described. Additional hearts were used for frozen sections and slides from the ischemic area were stained with oil red O staining as previously described.

**Statistical Analysis**

Data are expressed as mean±SEM. Differences between the groups were calculated by Student t test. A value of P<0.05 was considered significant.

**Results**

**PPARα-Regulated Gene Expression**

PPARα, a nuclear receptor that regulates fatty acid metabolism, modulates substrate preference in the heart. We have previously reported that hypoxia and pressure overload downregulate PPARα-regulated gene expression in the rodent heart. Similarly, PPARα transcript levels were decreased in mouse hearts exposed to 7 and 28 days of repetitive I/R (Figure 1). Medium-chain acyl CoA dehydrogenase (MCAD) and muscle carnitine palmitolyl transferase 1 (mCPT1), PPARα-regulated genes involved in the transport and oxidation of fatty acids in the mitochondria, also demonstrated significant downregulation at 7 and 28 days of I/R (Figure 1). Interestingly, repetitive I/R induced a transient increase in the expression of uncoupling protein 3 (UCP3), another PPARα-regulated gene involved in uncoupling the electron transport chain (Figure 1). PPARα, MCAD, mCPT1, and UCP3 transcript levels were increased 15 and 30 days after the discontinuation of I/R (Figure 1). Thus, our findings suggest that repetitive I/R causes a reversible downregulation of fatty acid metabolism.

**Myosin Iso-Gene Expression**

Myosin heavy chain (MHC), the main component of myosin, exists in 2 distinct isoforms. Myosin composed of predominantly MHC-β isoform has a decreased ATPase activity compared with MHC-α which results in decreased contractile velocity, but greater economy in force generation. Because hibernating myocardium is characterized by contractile dysfunction, we investigated myosin iso-gene expression in this model of repetitive I/R. Repetitive I/R caused a decrease in both MHC-α and MHC-β expression (Figure 2). Although MHC-α transcript levels returned to baseline after discontinuation of I/R, MHC-β expression remained downregulated.

**Gene Expression in EC-SOD Mice**

Because hypoxia regulates both myosin iso-gene expression and PPARα-regulated gene expression, we postulated that ischemia (which causes tissue hypoxia) regulates PPARα and myosin iso-gene expression in repetitive I/R. However, we have also shown that repetitive I/R induces a transient inflammatory response dependent on ROS that leads to interstitial fibrosis and ventricular dysfunction. Furthermore, there is increased carbonylated protein, an indirect marker of oxidative stress, in hearts exposed to repetitive I/R (Sharma et al, unpublished observation). To determine whether metabolic and myosin iso-gene expression is dependent on ischemia itself or ROS production, we performed repetitive I/R in mice that overexpress EC-SOD, an enzyme that scavenges ROS. We have previously demonstrated that EC-SOD overexpression attenuated ROS-dependent cytokine expression and interstitial fibrosis and improves contractile function in heart exposed to repetitive I/R. These observations provide indirect evidence that EC-SOD overexpression reduces oxidative stress.
stress–induced injury. Surprisingly, repetitive I/R failed to decrease the expression of PPARα and mCPT1 in EC-SOD mice (Figure 3). MCAD and UCP3 transcript levels were only transiently downregulated with repetitive I/R (Figure 3). MHC-α expression was unchanged in EC-SOD mice exposed to I/R, whereas MHC-β transcript levels were markedly and persistently increased (Figure 4). Therefore, both metabolic and myosin gene expression depends, at least in part, on ROS.

Reactivation of PPARα
To determine whether downregulation of PPARα-regulated gene expression is adaptive, we examined gene expression, contractile function, and histology in mice exposed to repetitive I/R and to the PPARα agonist-WY 14,643. We used a dose of WY-14,643 that has been shown to reactivate PPARα-regulated genes in the hypertrophied rat to normal baseline levels without causing toxicity in sham-operated mice.

Figure 1. PPARα-regulated gene expression in WT mice exposed to 3, 5, 7, and 28 days of repetitive I/R and 15 and 30 days of recovery after 7 days of I/R (n=7 in each time point). There is sustained downregulation of PPARα, mCPT1, and MCAD transcript levels starting at 7 days that returns to baseline after discontinuation of I/R. UCP3 is transiently upregulated at 3 and 5 days of I/R exposure and then markedly upregulated after discontinuation of I/R. *P<0.05 vs sham; #P<0.05 vs 7-day I/R.

Figure 2. Myosin iso-gene expression in WT mice exposed to 3, 5, 7, and 28 days of repetitive I/R and 15 and 30 days of recovery after 7 days of I/R (n=7 in each time point). There is sustained downregulation of MHC-α and MHC-β starting at 7 days of I/R. Only MHCα transcript levels return to baseline after discontinuation of I/R. MHC-β expression remains downregulated. *P<0.05 vs sham; #P<0.05 vs 7 days of I/R.
rats. Agonist treatment restored the expression of MCAD to sham levels without altering PPARα expression, confirming enhanced activity of PPARα (Figure 5A). Although fractional shortening was decreased in mice exposed to 7 days of I/R, agonist treatment was associated with even worse contractile function (Figure 5B). There was no difference between agonist-treated and control mouse hearts by hematoxylin and eosin staining (Figure 6A and B). Although 7 days of I/R induced interstitial fibrosis, agonist treatment was, surprisingly, associated with microinfarctions in the ischemic myocardium (Figure 6C and D). Equally unexpected was the observation that PPARα reactivation during brief repetitive I/R was associated with intramyocardial triglyceride accumulation in the ischemic myocardium (Figure 6E and F).

**Discussion**

We investigated the role of metabolic gene expression in a murine model of ischemic cardiomyopathy. The 3 main findings of our study are that (1) there was a reversible downregulation of PPARα-regulated gene expression and myosin iso-gene expression in mouse hearts exposed to the repetitive I/R, (2) decreases in PPARα and myosin iso-form expression...
transcript levels were dependent on ROS and (3), reactivation of PPARα during repetitive I/R was associated with worsened contractile function, microinfarction, and intramyocardial triglyceride accumulation in the ischemic myocardium.

Downregulation of Fatty Acid Metabolism in Repetitive I/R

Downregulation of PPARα is thought to regulate the switch from fatty acid to glucose use in the pressure-overloaded rat heart and the failing human heart. A variety of stimuli, including hypobaric hypoxia and unloading, are also capable of inducing a switch in substrate use associated with downregulation of PPARα-regulated genes. Here, we demonstrate that repetitive brief episodes of I/R are associated with a downregulation of PPARα-regulated gene expression, indicating a switch in substrate preference from fatty acids to glucose. Glucose is a more efficient fuel in terms of oxygen consumption for ATP generation than fatty acids. Fatty acid use also has detrimental effects on the postischemic heart by increasing lactate and proton production in the myocardium. The subsequent fall in intracellular pH is associated with contractile dysfunction. Therefore, switching substrate preference from fatty acids to carbohydrates in ischemia may be an adaptive mechanism that provides greater efficiency of energy conversion and ultimately the preservation of cardiomyocytes. However, in repetitive brief sublethal ischemia, the role of PPARα downregulation is not completely obvious. We suggest that repetitive I/R induces transcriptional changes favoring carbohydrate metabolism in anticipation of subsequent ischemic insults. Earlier, we called this phenomenon metabolic “programmed cell survival.”

It has been shown previously that myocardial hibernation induced by repetitive I/R is associated with a switch in substrate use from fatty acids to glucose. Our findings suggest that substrate switching in repetitive I/R is caused by a downregulation of PPARα. This observation supports the concept of PPARα as a master regulator of substrate preference in the heart. The downregulation of PPARα-regulated gene expression is completely reversible on discontinuation of I/R, indicating that these alterations in metabolic gene expression may be associated with hibernating myocardium.

Although MCAD and mCPT1 transcript levels were downregulated with repetitive I/R, UCP3 expression was transiently upregulated. Unlike MCAD and mCPT1, UCP3 does not directly regulate fatty acid oxidation. Instead, UCP3 uncouples the electron transport chain and reduces the generation of ROS in the mitochondria. Therefore, UCP3 acts like an antioxidant, reducing mitochondrial oxidative stress in response to fatty acid oxidation. Our findings suggest that UCP3 may also be regulated by a transcriptional mechanism other than PPARα. We speculate that transient upregulation of UCP3 may be a protective mechanism that helps to reduce intracellular oxidative stress during the development of fibrosis and ventricular dysfunction.

Downregulation of Myosin Iso-Gene Expression in Repetitive I/R

As mentioned earlier, MHC-β has decreased contractile velocity but conserves more ATP per contraction than MHC-α. The normal rodent heart consists of <10% MHC-β. In response to pressure overload or hypoxia, there is switch in myosin iso-gene expression, resulting in an increase in the ratio of MHC-β to MHC-α expression. This change in myosin content, which results in an increase in MHC-β composition, is thought to contribute to contractile dysfunction. In repetitive I/R, we found a decrease in both myosin isofrom transcript levels. The downregulation of both myosin isoforms may contribute to decreased contractile function in our model. However, we speculate that this decrease in myosin iso-gene expression may be a compensatory mechanism, reducing energy expenditure in the myocardium at the expense of contractile function.

ROS Regulation of Gene Expression in Repetitive I/R

Because hypoxia regulates metabolic and myosin iso-gene expression, we hypothesized that ischemia itself modulates PPARα and myosin iso-gene transcript levels in response to repetitive I/R. Surprisingly, repetitive I/R in mice that overexpress EC-SOD, a scavenger of ROS, failed to downregulate both PPARα-regulated genes or myosin iso-gene expression. Therefore, downregulation of PPARα and myosin iso-gene expression is dependent on ROS, not ischemia itself. Furthermore, although acute I/R can rapidly induce gene expression, sustained downregulation of PPARα-regulated genes developed only after 7 days of I/R, suggesting a cumulative or delayed regulation by ROS.

Mitochondrial substrate oxidation is an important source of intracellular oxidative stress. Hyperglycemia and high circulating fatty acids can induce cellular dysfunction by increasing mitochondrial ROS generation. Furthermore, cardiac-specific overexpression of PPARα induces cardiac...
dysfunction in the presence of high circulating fatty acids associated with increased ROS generation. Therefore, increased mitochondrial ROS generation may contribute to contractile dysfunction in postischemic hearts that oxidizes fatty acid. We speculate that ROS-mediated downregulation of PPARα is an important protective mechanism that reduces intracellular ROS generation. Therefore, ROS not only contributes to inflammation and fibrosis, as we have previously shown, but also regulates changes in metabolism that may protect the heart from further oxidative stress (Figure 7). Whether ROS directly regulates PPARα expression or indirectly modulates transcript levels via changes in cytokines or contractility is unclear. Invariably, the role of ROS in ischemia and reperfusion is complex, and the mechanism of ROS regulation of PPARα and myosin iso-gene expression requires further investigation. Although PPARα expression did not change in EC-SOD mice exposed to repetitive I/R, both MCAD and UCP3 transcript levels were transiently downregulated. Furthermore, mCPT1 transcript levels are also slightly decreased (although not significantly) on day 3 of I/R. These observations suggest that mechanisms other than ROS also regulate PPARα activity in hearts exposed to I/R.

Interestingly, MHC-β transcript levels were markedly increased in the EC-SOD mice exposed to I/R, suggesting that in the absence of oxidative stress, I/R induces the fetal pattern of myosin iso-gene expression. Furthermore, these observations also suggest that ROS profoundly decreases the expression of MHC-β. Perhaps the inability of MHC-β transcript levels to return to baseline after discontinuation of I/R reflects the profound affect of ROS on MHC-β gene expression. Because the EC-SOD mice have improved contractile function compared with control mice in response to repetitive I/R, our findings suggest that increased MHC-β expression does not negatively affect cardiac function in the setting of repetitive I/R.

**Contractile Dysfunction Associated With PPARα Reactivation**

Reactivation of PPARα in pressure overload–induced hypertrophy is associated with contractile dysfunction. Here, we also demonstrate that reactivation of PPARα in repetitive I/R worsens contractile function, suggesting that downregulation of fatty acid metabolism in the repetitive I/R is necessary to maintain cardiac function. As mentioned, enhanced fatty acid metabolism in the postischemic heart may increase the
generation of intracellular ROS and increase lactate and proton production. Furthermore, PPARα activation can decrease glucose use by increasing the expression of pyruvate dehydrogenase kinase 4 (PDK-4), which phosphorylates and inhibits the pyruvate dehydrogenase complex. Therefore, this decrease in glucose use caused by reactivation of PPARα in repetitive I/R may worsen energy efficiency and contribute to contractile dysfunction.

The observation of microinfarctions in the ischemic myocardium was totally unexpected. ROS generation from disorder fatty acid metabolism can also induce apoptosis. Furthermore, cardiac-specific overexpression of PPARα enhances ROS production. We have previously reported that inflammation and fibrosis in repetitive I/R depend on ROS. Therefore, we speculate that reactivation of PPARα, which results in a switch back from glucose to fatty acid use, enhances ROS generation in myocardium, subsequently increasing inflammation, fibrosis, and apoptosis (Figure 7). Although we can speculate that these mechanisms may worsen fibrosis and contractile function, it is unclear how they manifest as localized microinfarction. It is conceivable that focal areas of increased wall stress, in conjunction with increased ROS generation in agonist-treated hearts, induce microinfarctions.

An equally unexpected finding is the increased intramyocardial triglyceride accumulation in the ischemic regions of PPARα-reactivated hearts. PPARα regulates the expression of genes involved in both the uptake (eg, fatty acid transporters, acyl CoA synthetase) and oxidation of fatty acids (eg, MCAD, mCPT1). We expected that intramyocardial triglyceride accumulation would decrease in the ischemic myocardium as the machinery for fatty acid oxidation is increased by PPARα reactivation. However, metabolism is not solely regulated by transcriptional mechanisms. In fact, much of metabolism is regulated at the posttranscriptional level (eg, Randle effect, malonyl CoA, allosteric inhibition by glycolytic intermediates, phosphorylation of enzymes). Our findings suggest that in repetitive I/R, reactivation of PPARα enhances the uptake of fatty acids greater than fatty oxidation, implying that fatty acid oxidation is inhibited by a yet-to-be-determined posttranscriptional mechanism. Therefore, reactivation of PPARα activity in repetitive I/R is associated with a mismatch between uptake and oxidation of fatty acids, leading to intramyocardial triglyceride deposition. Nonetheless, this accumulation of intramyocardial triglycerides associated with contractile dysfunction and microinfarctions strongly suggests cardiac lipotoxicity.

Recently, a study by Yue et al suggested that PPARα activation in acute I/R protects the heart. In this study, however, acute I/R induced a rapid rise in serum free fatty acid levels that was abolished with PPARα activation. This transient increase in free fatty acid delivery and subsequent increase in oxidation may be detrimental to the postischemic heart. Thus, despite an increase in myocardial fatty acid enzyme machinery in PPARα agonist–treated mice exposed to acute I/R, fatty acid oxidation would actually decrease because of the systemic effect of the PPARα agonist that decreases free fatty acid delivery to the myocardium. This interplay between the systemic and localized effects of PPARα activation probably explains why drugs that activate PPARα (eg, fibrates) have not been shown to increase cardiovascular complications in patients.

Prevention of Cardiac Lipotoxicity in Repetitive I/R

Collectively, the term cardiac lipotoxicity refers to a constellation of findings characterized by altered fatty acid metabolism, intramyocardial triglyceride overload, and contractile dysfunction. The accumulation of triglyceride within the myocardium, a marker of lipotoxicity, is caused by a mismatch between the uptake and oxidation of fatty acids and is a feature of a number of pathological processes. Cardiac overexpression of genes regulating fatty acid metabolism such as acyl CoA synthetase, fatty acid transport protein 1, and PPARα induces lipotoxic cardiomyopathy. Loss of function mutation in the leptin receptor induces obesity and type II diabetes in rodents associated with intramyocardial lipid overload and contractile dysfunction. Patients with congenital lipodystrophy, a rare disorder in which the absence of adipocytes results in the accumulation of lipid in nonadipose tissues, or with inherited mitochondrial fatty acid oxidation defects develop premature cardiomyopathy. We have shown that patients with nonscared cardiomyopathy and intramyocardial triglyceride overload also have increased PPARα-regulated gene expression.

Although the mechanism of lipotoxicity is unclear, free-radical generation from disordered fatty acid metabolism has been shown to induce apoptosis, a phenomenon called lipoapoptosis. We did not observe intramyocardial triglyceride deposition in mouse hearts exposed to repetitive I/R. However, restoration of PPARα activity to baseline in hearts...
exposed to repetitive I/R was associated with markedly increased intramyocardial triglyceride accumulation, worsened contractile function, and microinfarction, features suggestive of cardiac lipotoxicity. We suggest that the downregulation of PPARα in hearts exposed to repetitive I/R is an adaptive mechanism that improves energy efficiency and reduces fatty acid–mediated oxidative stress, which subsequently decreases inflammation, fibrosis, and apoptosis (Figure 7). Further support for this concept comes from the finding that normal PPARα-regulated gene expression does not contribute to cardiac dysfunction in EC-SOD mice. We have previously demonstrated that EC-SOD mice have improved contractile function in response to repetitive I/R,8 yet there is no difference in PPARα-regulated gene expression in EC-SOD mice. Presumably, overexpression of EC-SOD, which scavenges ROS, protects the ischemic myocardium from fatty acid–mediated oxidative stress. Interestingly, the observation that ROS regulates PPARα-regulated gene expression suggests a complex but coordinated ROS-mediated transcriptional response that may ultimately reduce the generation of free radicals and protect against cardiac lipotoxicity and lipoapoptosis.

Study Limitations

In our hands, metabolic gene expression correlates well with cardiac substrate use.1 Nonetheless, we did not actually measure cardiac substrate use in this study because of several technical limitations. Although cardiac substrate use would have strengthened the message of this study, substrate switching from fatty acids to glucose metabolism in repetitive I/R and hibernating myocardium has been described in the literature.41 Furthermore, reactivation of PPARα with WY-14,643 has been shown to increase fatty acid oxidation and to decrease glucose use in the heart.1 Future studies using micro-PET to qualitatively elucidate in vivo cardiac metabolism in repetitive I/R will undoubtedly enhance our understanding of substrate switching in repetitive I/R.

Conclusions

Repetitive I/R is associated with a reversible downregulation of PPARα-regulated gene expression and myosin iso-gene expression that are dependent on ROS. Because restoration of PPARα activity in repetitive I/R was associated with intramyocardial triglyceride accumulation and cardiac dysfunction, we speculate that downregulation of PPARα transcript levels is an adaptive mechanism that prevents cardiac lipotoxicity.

Acknowledgments

This study was supported by grants from the NHLBI (RO1-HL073162-01 and T32-HL 07591 to Dr Taegtmeyer) and (HL-42550 to Dr Entman). Dr Dewald was supported by the Deutsche Forschungsgemeinschaft and BONFOR (DE801/1-1). We thank Thuy Pham, Christine Peigney, and Helge Doerr for their expert technical assistance.

References


19. Lopaschuk GD, Warmbolt RB, Barr RL. An imbalance between glycolysis and glucose oxidation is a possible explanation for the detrimental effects of high levels of fatty acids during aerobic reperfusion of ischemic hearts. J Pharmacol Exp Ther. 1993;264:135–144.


CLINICAL PERSPECTIVE

The heart normally uses fatty acids as its chief fuel source. In the hibernating myocardium, a condition characterized by reversible cardiac dysfunction and fibrosis but no myocardial infarction, there is increased reliance of the heart on glucose. Here, we examine the role of metabolic gene expression in a mouse model of reversible ischemic cardiomyopathy induced by repetitive ischemia and reperfusion (I/R). In response to repetitive I/R, there was a reversible downregulation of the genes that modulate fatty acid metabolism and myosin heavy chain isoforms in the heart. Overexpression of extracellular superoxide dismutase, an endogenous antioxidant enzyme, in hearts exposed to repetitive I/R failed to cause the decrease in metabolic and myosin isoform gene expression. When we pharmacologically reactivate fatty acid metabolism in hearts exposed to repetitive I/R, there was worsened contractile function, microinfarctions, and triglyceride accumulation within cardiomyocytes. Our findings suggest that downregulation of fatty acid metabolic gene expression in the hibernating myocardium is an adaptive mechanism. Furthermore, modulation of myocardial metabolism may provide a pharmacological target for cardiac protection in repetitive I/R.
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_Circulation_. 2005;112:407-415; originally published online July 11, 2005;
doi: 10.1161/CIRCULATIONAHA.105.536318

_Circulation_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
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Print ISSN: 0009-7322. Online ISSN: 1524-4539

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