Myocardial ischemia-reperfusion injury: a neglected therapeutic target

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Acute myocardial infarction (MI) is a major cause of death and disability worldwide. In patients with MI, the treatment of choice for reducing acute myocardial ischemic injury and limiting MI size is timely and effective myocardial reperfusion using either thombolytic therapy or primary percutaneous coronary intervention (PPCI). However, the process of reperfusion can itself induce cardiomyocyte death, known as myocardial reperfusion injury, for which there is still no effective therapy. A number of new therapeutic strategies currently under investigation for preventing myocardial reperfusion injury have the potential to improve clinical outcomes in patients with acute MI treated with PPCI.

Introduction

Coronary heart disease (CHD) is the leading cause of death and disability worldwide. According to the WHO, 7,254,000 deaths worldwide (12.8% of all deaths) resulted from CHD in 2008. The effects of CHD are usually attributable to the detrimental effects of acute myocardial ischemia-reperfusion injury (IRI). IRI typically arises in patients presenting with an acute ST-segment elevation myocardial infarction (STEMI), in whom the most effective therapeutic intervention for reducing acute myocardial ischemic injury and limiting the size of myocardial infarction (MI) is timely and effective myocardial reperfusion using either thrombolytic therapy or primary percutaneous coronary intervention (PPCI). However, the process of myocardial reperfusion can itself induce further cardiomyocyte death, a phenomenon known as myocardial reperfusion injury (1–3). Although the process of myocardial reperfusion continues to improve with more timely and effective reperfusion and with advances in PCI technology and antiplatelet and antithrombotic agents for maintaining the patency of the infarct-related coronary artery, there is still no effective therapy for preventing myocardial reperfusion injury. In this respect, myocardial reperfusion injury remains a neglected therapeutic target for cardioprotection in PPCI patients. In this article, the pathophysiology of myocardial lRI and the emerging therapeutic strategies for protecting the heart from its detrimental effects are reviewed.

Pathophysiology of myocardial ischemic injury

Acute occlusion of the coronary artery in the STEMI patient subjects the myocardium supplied by that vessel to acute myocardial ischemia, thereby demarcating the area at risk (AAR) of potential MI, should the acute coronary occlusion be sustained or permanent. If the period of acute myocardial ischemia is prolonged (more than 20 minutes) a "wave front" of cardiomyocyte death begins in the subendocardium and extends transmurally over time toward the epicardium (4).

The deprivation of oxygen and nutrient supply results in a series of abrupt biochemical and metabolic changes within the myocardium (Figure 1). The absence of oxygen halts oxidative phosphorylation, leading to mitochondrial membrane depolarization, ATP depletion, and inhibition of myocardial contractile

function. This process is exacerbated by the breakdown of any available ATP, as the F_1F_0 ATPase functions in reverse to maintain the mitochondrial membrane potential, resulting in ATP hydrolysis and an increase in mitochondrial inorganic phosphate. In the absence of oxygen, cellular metabolism switches to anaerobic glycolysis, resulting in the accumulation of lactate, which reduces intracellular pH (to <7.0). The intracellular accumulation of protons activates the Na^+-H^+ ion exchanger, which extrudes protons from the cell in exchange for Na^+ entry. The lack of ATP during ischemia ceases function of the $3Na^+$-$2K^+$ ATPase, thereby exacerbating the intracellular Na^+ overload. In response, the reverse activation of the $2Na^+$-Ca^{2+} ion exchanger results in intracellular Ca^{2+} overloading as the cell tries to extrude Na^+ (5).

Pathophysiology of myocardial reperfusion injury

After the onset of acute myocardial ischemia in patients with STEMI, timely myocardial reperfusion using PPCI is essential to salvage viable myocardium, limit MI size, preserve LV systolic function, and prevent the onset of heart failure. However, the reperfusion of acutely ischemic myocardium can independently induce cardiomyocyte death (1–3), although this concept has been difficult to accept over the years. The four recognized forms of myocardial reperfusion injury are discussed in detail below, the first two reversible and the second two irreversible.

Reperfusion-induced arrhythmias. The sudden reperfusion of acutely ischemic myocardium in STEMI patients undergoing PPCI may be accompanied by ventricular arrhythmias, which usually self-terminate or are easily treated (6).

Myocardial stunning. The reversible post-ischemic contractile dysfunction that occurs on reperfusing acute ischemic myocardium is referred to as myocardial stunning. This form of reperfusion injury results from the detrimental effects of oxidative stress and intracellular calcium overload on the myocardial contractile apparatus (7).

Microvascular obstruction. Microvascular obstruction (MVO) was first described by Krug et al. in 1966 as the "inability to reperfuse a previously ischemic region" (8). The major contributing factors include capillary damage with impaired vasodilatation, external capillary compression by endothelial cell and cardiomyocyte swelling, micro-embolization of friable material released from the atherosclerotic plaque, platelet micro-thrombi, the release of soluble vasomotor and thrombogenic substances, and neutrophil
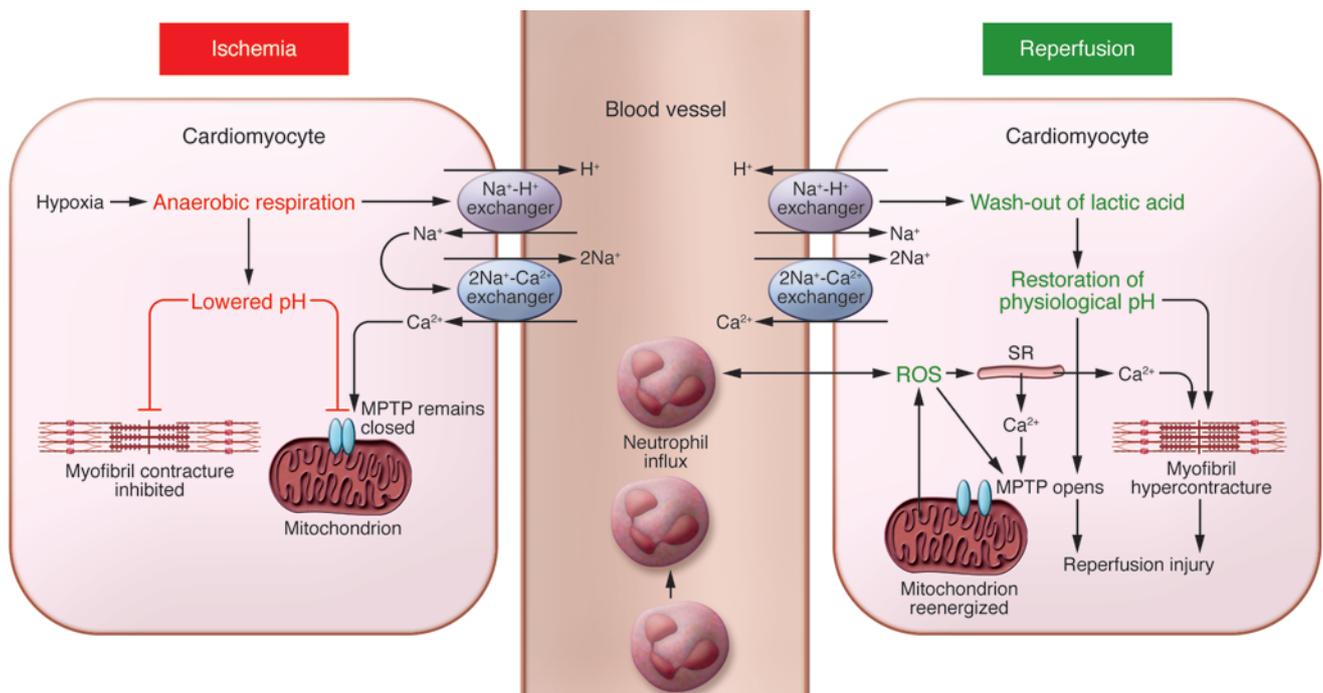
Figure 1
Schematic illustrating the main proponents of acute myocardial IRI. During acute myocardial ischemia, the absence of oxygen switches cell metabolism to anaerobic respiration, resulting in the production of lactate and a drop in intracellular pH. This induces the Na^+-H^+ exchanger to extrude H^+ and results in intracellular Na^+ overload, which activates the $2Na^+$-Ca^{2+} exchanger to function in reverse to extrude Na^+ and leads to intracellular Ca^{2+} overload. The Na^+-K^+ ATPase ceases to function in ischemia, exacerbating intracellular Na^+ overload. The acidic conditions during ischemia prevent the opening of the MPTP and cardiomyocyte hypercontracture at this time. During reperfusion, the electron transport chain is reactivated, generating ROS. Other sources of ROS include xanthine oxidase (endothelial cells) and NADPH oxidase (neutrophils). ROS mediate myocardial reperfusion injury by inducing the opening of the MPTP, acting as a neutrophil chemoattractant, and mediating dysfunction of the sarcoplasmic reticulum (SR). This contributes to intracellular Ca^{2+} overload and damages the cell membrane by lipid peroxidation, inducing enzyme denaturation and causing direct oxidative damage to DNA. Reperfusion and reactivation of the Na^+-H^+ exchanger result in washout of lactic acid, resulting in the rapid restoration of physiological pH, which releases the inhibitory effect on MPTP opening and cardiomyocyte contracture. The restoration of the mitochondrial membrane potential drives calcium into the mitochondria, which can also induce MPTP opening. Several hours after the onset of myocardial reperfusion, neutrophils accumulate in the infarcted myocardial tissue in response to the release of the chemoattractants ROS, cytokines, and activated complement.

plugging (9–12). At coronary angiography in PPCI patients, MVO manifests as sluggish coronary blood flow, impaired myocardial blush grade, and a characteristic coronary flow velocity profile (13). Importantly, 30%–40% of PPCI patients in whom coronary blood flow in the infarct-related coronary artery appears normal on coronary angiography have evidence of MVO as detected by myocardial contrast echocardiography (14, 15), myocardial perfusion nuclear scanning (16), or contrast-enhanced cardiac MRI (17, 18). The presence of MVO is associated with a larger MI size, a lower LV ejection fraction, adverse LV remodelling, and worse clinical outcomes (15, 19, 20). In severe cases of MVO in which there is significant damage to the endothelium, extravasation of blood into the interstitium can produce intramyocardial hemorrhage within the area of infarction, a feature that can also be detected by cardiac MRI (21). No effective therapy currently exists for reducing MVO in patients who have undergone PPCI. Whether MVO is actually an independent causative factor of reperfusion-induced cardiomyocyte death or is merely a biomarker of severe myocardial IRI remains unclear.

Lethal myocardial reperfusion injury. Reperfusion-induced death of cardiomyocytes that were viable at the end of the index ischemic event is defined as lethal myocardial reperfusion injury (2). The major contributory factors are discussed below and include oxidative stress, calcium overload, mitochondrial permeability transition pore (MPTP) opening, and hypercontracture (3). The existence of lethal myocardial reperfusion injury has been inferred in both experimental MI models and in patients with STEMI by the observation that therapeutic interventions applied solely at the onset of myocardial reperfusion reduced MI size by 40%–50% (3). This observation suggests that lethal myocardial reperfusion injury may account for up to 50% of the final MI size. Lethal myocardial reperfusion injury attenuates the full benefits of myocardial reperfusion in terms of MI size reduction and thus represents an important target for cardioprotection in PPCI patients (see Figure 2). However, no effective therapy currently exists for reducing lethal myocardial reperfusion injury in patients who have undergone PPCI.

Mediators of myocardial reperfusion injury

Experimental studies have identified several critical factors that act in concert to mediate the detrimental effects of myocardial reperfusion injury (see Figure 1).
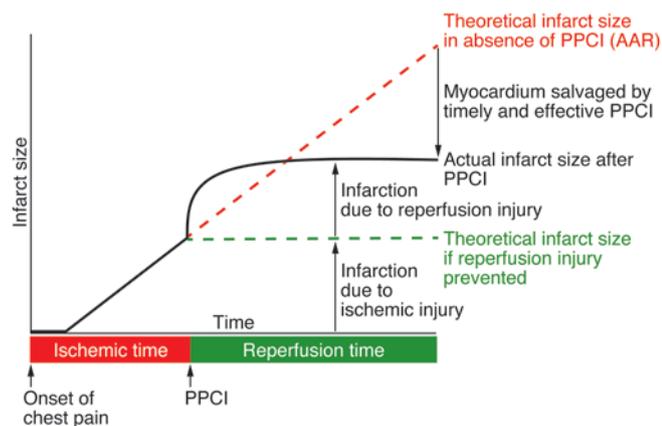
Figure 2
This figure illustrates the individual contributions of acute myocardial ischemic injury and myocardial reperfusion injury to final MI size (expressed in arbitrary units) in STEMI patients up to 24 hours following PPCI. The black solid line depicts the individual contributions to final MI size of acute myocardial ischemic injury and of myocardial reperfusion injury. The green dashed line depicts the theoretical MI size following PPCI based on acute myocardial ischemia alone in the absence of myocardial reperfusion injury. The red dashed line depicts the theoretical MI size based on acute myocardial ischemia alone in the absence of PPCI. The presence of myocardial reperfusion injury attenuates the benefit of PPCI in terms of the reduction of MI size. Therefore, the administration of a therapeutic strategy as an adjunct to PPCI that is capable of reducing myocardial reperfusion injury would result in a smaller MI size (as depicted by the green dashed line) and take advantage of full benefits of myocardial reperfusion. Figure modified from *Cardiovascular Research* (93).

Oxidative stress. In the first few minutes of myocardial reperfusion, a burst of oxidative stress (22, 23) is produced by a variety of sources (Figure 1). This detrimental oxidative stress mediates myocardial injury and cardiomyocyte death through a number of different mechanisms (Figure 1). Based on these observations, antioxidant therapy was naturally considered to be an appropriate option to prevent such injury. However, both experimental and clinical studies have reported mixed results with the administration of antioxidant therapy at the onset of myocardial reperfusion. The reason for this may in part be due the inability of the antioxidant to enter the cell (24). In this regard, the discovery of mitochondrial-specific antioxidants may be more effective (25).

Intracellular Ca^{2+} overload. Intracellular and mitochondrial Ca^{2+} overload begins during acute myocardial ischemia and is exacerbated at the time of myocardial reperfusion due to disruption of the plasma membrane, oxidative stress-induced damage to the sarcoplasmic reticulum, and mitochondrial re-energization (Figure 1). Mitochondrial re-energization allows the recovery of the mitochondrial membrane potential that drives the entry of Ca^{2+} into mitochondria via the mitochondrial Ca^{2+} uniporter and subsequently induces the opening of the MPTP (Figure 1). Experimental studies have shown that pharmacologic antagonists of the sarcolemmal Ca^{2+} channel (26) or the mitochondrial Ca^{2+} uniporter (27), administered at the onset of myocardial reperfusion, reduce MI size by up to 50%. However, not all experimental studies using this therapeutic strategy have been positive. Clinical studies of calcium channel blockers administered at the onset of myocardial reperfusion have not shown beneficial results (28). The recent

identification of the mitochondrial Ca^{2+} uniporter (29) may result in the discovery of a new class of specific inhibitors for targeting lethal myocardial reperfusion injury.

The rapid restoration of physiological pH at the time of reperfusion. During acute myocardial ischemia the intracellular pH decreases to less than 7.0, whereas at reperfusion, physiological pH is rapidly restored by the washout of lactate and the activation of the Na^+-H^+ exchanger as well as the Na^+-HCO^- symporter. This pH shift contributes to the cardiomyocyte death of lethal myocardial reperfusion injury (30) by permitting MPTP opening and cardiomyocyte rigor hypercontracture in the first few minutes of reperfusion. Reperfusion of ischemic animal hearts with an acidic buffer can reduce MI size (31). Therefore, a potential treatment strategy for preventing lethal myocardial reperfusion injury would be to slow the normalization of physiologic pH at the time of myocardial reperfusion, which may be achieved via the pharmacologic inhibition of the Na^+-H^+ exchanger (5) or by slowing the process of myocardial reperfusion, as in the case of ischemic postconditioning (IPost) (32), which has been termed "the pH hypothesis" by Cohen and Downey (33).

The MPTP: an important target for cardioprotection. Many of the above proponents of myocardial reperfusion injury appear to converge on the MPTP. The MPTP is a nonselective channel of the inner mitochondrial membrane, the opening of which results in mitochondrial membrane depolarization and uncoupling of oxidative phosphorylation, leading to ATP depletion and cell death (34, 35). In the setting of acute myocardial IRI, the MPTP has been shown to remain closed during ischemia and only open at reperfusion in response to mitochondrial Ca^{2+} and phosphate overload, oxidative stress and relative ATP depletion, and rapid pH correction (36). As such, preventing MPTP opening at the time of reperfusion by administering known MPTP inhibitors (such as the immunosuppressant cyclosporin A) at the onset of myocardial reperfusion has been reported in experimental studies to reduce MI size by 40%–50% in small and large animal MI models (37–40) and protect human atrial trabeculae subjected to simulated IRI (41). As such, the MPTP provides an important therapeutic target for preventing lethal myocardial reperfusion injury (see section below).

Inflammation: guilty mediator or innocent bystander. It is unclear whether the inflammatory response that accompanies an acute MI contributes to the pathogenesis of lethal myocardial reperfusion injury or whether it is a reaction to the acute myocardial injury (42). Although experimental studies have reported significant reduction of MI with therapeutic strategies designed to inhibit the inflammatory process at the time of myocardial reperfusion using antibodies against cell-adhesion molecules (43–45) and the inhibition of complement activation (46), corresponding clinical studies using this therapeutic approach have been largely negative (47–49).

Late myocardial reperfusion injury: extending the window of cardioprotection. The previously described stimulators of myocardial reperfusion injury all appear to operate in the first few minutes of myocardial reperfusion, providing a narrow window for reducing MI size in PPCI patients. However, several other important processes such as apoptosis and inflammation, which are also initiated during ischemia and continue over several hours into reperfusion, may contribute to the development of lethal myocardial reperfusion injury. These contributing pathways provide a potential second therapeutic window for reducing MI size, even well after myocardial reperfusion has taken place. Consistent with this proposal are

Table 1

Mechanical therapeutic interventions for preventing myocardial reperfusion injury in patients undergoing PPCI

Study	N	Therapeutic intervention	Result
IPost			
Staat et al. 2005 (66)	30	Four 60-s cycles of low-pressure inflation/deflation of angioplasty balloon	Reduction of MI size by 36% (72-hr AUC CK); improved myocardial blush grade
Thibault et al. 2008 (94)	38	Four 60-s cycles of low-pressure inflation/deflation of angioplasty balloon	Reduction of MI size by 40% (72-hr AUC CK); reduction of MI size by 39% at 6 mo, as assessed with SPECT; 7% increase in EF, as assessed with ECG, at one year
Lonborg et al. 2010 (95)	118	Four 60-s cycles of low-pressure inflation/deflation of angioplasty balloon	Reduction of MI size by 19% at 3 mo, as assessed with CMR; 31% increase in myocardial salvage index
Sorensson et al. 2010 (96)	76	Four 60-s cycles of low-pressure inflation/deflation of angioplasty balloon	No difference in 48-hr AUC CK-MB or Trop-T; no difference in myocardial salvage, as assessed with CMR, on days 7–9; significant increase in myocardial salvage in patients with large AAR (>30% of LV)
Tarantini et al. 2012 (97)	78	Four 60-s cycles of low-pressure inflation/deflation of angioplasty balloon; IPost protocol delivered in stent	Trend toward increased MI size; increased adverse events
Freixa et al. 2012 (98)	79	Four 60-s cycles of low-pressure inflation/deflation of angioplasty balloon; IPost protocol delivered in stent	Worse myocardial salvage; no difference in MI size
Engstrom et al. 2012 (99); DANAMI-3	2,000	Four 30-s cycles of low-pressure inflation/deflation of angioplasty balloon	Ongoing phase 3 study investigating the effect of IPost on death and HHF
RIC			
Botker et al. (77, 100)	142	Four 5-min cycles of inflation/deflation of cuff placed on upper arm, in the ambulance	Increase in myocardial salvage index from 0.55 to 0.75; patients with anterior STEMI with occluded arteries showed most benefit
Rentoukas et al. 2010 (101)	92	Three 4-min cycles of inflation/deflation of cuff placed on upper arm, delivered at hospital prior to PPCI	Less STR and smaller Trop-I peak; additive effect with i.v. morphine
Therapeutic hypothermia			
Gotberg et al. 2010 (102); RAPID-MI-ICE	20	Cooling to 35°C prior to PPCI by i.v. infusion of 1–2 liters of cold saline and cooling with Philips InnerCool RTx Endovascular System	Reduction in MI size as % of AAR, as assessed with CMR at 4 days (30% vs 48%); 43% reduction in peak and cumulative Trop-T release
Erlinge et al. 2012 (103); CHILL-MI	120	Cooling to 35°C prior to PPCI by i.v. infusion of 1–2 liters of cold saline and cooling with Philips InnerCool RTx Endovascular System	Ongoing multicenter study investigating whether cooling prior to PPCI reduces MI size (as a % of AAR) on CMR at 4 days
Therapeutic hyperoxemia			
O'Neill et al. 2007 (78); AMIHOT I	269	IC hyperbaric hyperoxemic reperfusion started after PPCI and continued for 90 min	No difference in 14-day MI size as assessed with SPECT; patients with anterior STEMI <6 h showed improvements[A]
Stone et al. 2009 (104); AMIHOT II	281	IC hyperbaric hyperoxemic reperfusion started after PPCI and continued for 90 min	No difference in 14-day MI size as assessed with SPECT[A]

[A]Pooled analysis of the results from AMIHOT I/II suggested beneficial effects on MI size. CMR, cardiac MRI; EF, ejection fraction; HHF, hospitalization for heart failure; STR, ST-segment resolution. CK, creatine kinase; CK-MB, CK (muscle and brain iso-enzyme); IC, intracoronary; Trop-T, troponin-T; Trop-I, troponin-I.

experimental data demonstrating an increase in MI size as reperfusion time progresses, suggesting a wave front of reperfusion injury that progress with time (3, 50, 51). However, this is a controversial area of research, and some experimental studies have failed to demonstrate an increase in MI size with reperfusion time (52).

Several experimental studies have reported that administering cardioprotective agents such as erythropoietin (anti-apoptotic) (53), PI3K-γ/δ inhibitors (anti-inflammatory) (54), intracoronary aqueous oxygen (55), and IPost (anti-apoptotic and anti-inflammatory) (56) from 30 minutes to 24 hours into myocardial reperfusion may still limit acute MI size at 72 hours. Whether this therapeutic window exists in patients with STEMI who are under-

going PPCI is of great investigational interest, as such a window would allow a cardioprotective intervention to be administered some hours after the PPCI procedure. This area of research is still in its infancy but may suggest or provide an additional therapeutic window to target late into the reperfusion phase.

Therapeutic strategies for reducing acute myocardial ischemic injury

For patients presenting with a STEMI, the most effective and well-established therapeutic strategy for reducing acute myocardial ischemic injury and limiting MI size is timely myocardial reperfusion using either thrombolytic therapy or PPCI. The duration

Table 2

Pharmacologic therapies for preventing myocardial reperfusion injury in PPCI patients

Study	N	Therapeutic intervention	Result
Adenosine			
Ross et al. 2005 (105); AMISTAD II	2,118	Infusion of adenosine 50 or 70 μg/kg/min i.v. for 3 h after PPCI	No difference in death or HHF at 6 months; post hoc analysis showed benefits in patients presenting within 3.2 h
Anti-inflammatory agents			
Armstrong et al. 2007 (72); APEX-MI	5,745	Pexelizumab 2 mg/kg i.v. bolus given over 10 min prior to PPCI, followed by 0.05 mg/kg/h infusion for 24 hours	No difference in deaths at 30 days (pexelizumab, 4.1%; placebo, 3.9%)
Atar et al. 2009 (49); FIRE	232	Bolus of FX06 (200 mg i.v.) immediately prior to guidewire crossing obstruction, repeated within 10 min	No difference in MI size at 5 days or 4 months, as assessed with CMR; no difference in MI size, as assessed with Trop
Atrial natriuretic peptide			
Kitakaze et al. 2007 (106); J-WIND	569	Carperitide infusion at 0.025 μg/kg/min i.v. increase in LVEF at 6–12 months	Reduction in MI size by 14.7% (total CK AUC); 2.5% increase in LVEF at 6–12 months
Atorvastatin			
Kim et al. 2010 (107); STATIN STEMI	171	Oral atorvastatin 80 mg vs atorvastatin I 10 mg prior to PPC	No effect on death, MI, or revascularization; no difference in MI size (CK-MB maximum); improved myocardial perfusion (blush grade, STR)
Hahn et al. 2011 (108)	173	Oral atorvastatin 80 mg versus atorvastatin no difference in perfusion	No effect on MI size, as assessed with SPECT at days 5–14; no difference in perfusion
Post et al. 2012 (109); REPARATOR	52	Oral atorvastatin 80 mg versus atorvastatin 10 mg prior to PPCI	No effect on indexed LVESV; no difference in MI size or perfusion size or perfusion
CsA			
Piot et al. 2008 (80), Mewton et al. 2010 (110)	58	CsA (2.5 mg/kg i.v.) 10 min prior to PPCI	Reduction of 44% in MI size (72-h AUC total CK); 20% reduction in MI size (as assessed with CMR in a subset of 27 patients); 28% reduction in MI size and smaller LVESV on CMR at 6 months
Ovize et al. 2012 (111); CIRCUS	972	CsA (2.5 mg/kg i.v.) 10 min prior to PPCI	Ongoing phase 3 clinical trial investigating whether CsA improves clinical outcomes at one year (total mortality; HHF; increase in LV end-diastolic volume >15%
Delcasertib			
Lincoff et al. 2011 (112); PROTECTION-AMI	1,083	Delcasertib at 50, 150, and 450 mg/h i.v. for 24 hours started prior to PPCI	No difference in infarct size as assessed with CK-MB AUC; takes 5–30 min to reach steady state after beginning infusion
EPO			
Voors et al. 2010 (113); HEBE-III	529	EPO epoetin-α 60,000 IU i.v. within 3 hours after PPCI	No difference in LVEF at 6 weeks; no difference in MI size (as assessed using AUC CK-MB or Trop-T); more major cardiac adverse events occurred with EPO
Ott et al. 2010 (114); REVIVAL-3	138	EPO epoetin-β 33,000 IU i.v. immediately after PPCI, repeated after 24 and 48 hours	No difference in LVEF at 6 months, as assessed with CMR; no difference in MI size (assessed with CMR after 5 d and 6 mo)
Ludman et al. 2011 (115)	52	EPO epoetin-β 50,000 IU i.v. prior to PPCI, repeated after 24 hours	No difference in MI size at 3 days using CMR and Trop-T; 2-fold higher incidence of MVO on CMR
Exenatide			
Lonborg et al. 2011 (81, 116)	107	Infusion of exenatide (25 mg in 250 ml saline i.v.) started 15 min prior to PPCI and infused for 6 h	Increase in myocardial salvage index at 90 days by CMR (0.71 vs. 0.62); reduced MI size as % of AAR at 90 days as assessed with CMR (0.30 vs. 0.39); reduced MI size was observed for patients presenting <132 min (8% vs 11%)
GIK therapy			
Mehta et al. 2005 (117); CREATE-ECLA	20,201	Infusion of GIK (25% glucose, 50 U/l insulin, 80 mEq/l potassium infused i.v. at 1.5 ml/kg/h) for 24 h	No difference in 30-d mortality (9.7% placebo vs 10.0% GIK); majority of patients were treated by thrombolysis and not PPCI; GIK was begun after reperfusion in 68% of patients
Selke et al. 2012 (58); IMMEDIATE	357	Infusion of GIK solution (i.v. for 12 h), begun in the ambulance for suspected STEMI patients	No difference in progression to MI, but reduction in composite outcome of cardiac arrest or in-hospital mortality was 6.1% with GIK vs 14.4% with placebo
Sodium nitrite			
Frenneaux et al. 2012 (118); NIAMI	200	Sodium nitrite i.v. prior to PPCI	Ongoing phase 2 trial to investigate whether i.v. sodium nitrite reduces MI size (expressed as % of the AAR on CMR)
Mathur et al. 2012 (119); NITRITE-AMI	80	Intracoronary bolus of sodium nitrite given 5 min prior to PPCI	Ongoing phase 2 trial investigating whether intracoronary sodium nitrite reduces MI size (48-h total AUC CK)
TRO40303			
Atar et al. 2012 (120); MitoCare	180	Peripheral i.v. infusion of TRO40303 prior to PPCI	Ongoing phase 2 trial investigating whether TRO40303 reduces MI size (72-h AUC CK and Trop-I)

CsA, cyclosporin A; EPO, erythropoietin; LVESV, LV end-systolic volume.

of acute myocardial ischemia is a critical determinant of MI size, and as such, minimizing the time from chest pain onset to PPCI is the treatment priority. In the pre-hospital phase this includes increasing patient awareness of the symptoms of a MI (in order to minimize the delay before the emergency services are contacted), and the rapid diagnosis and transfer of STEMI patients to the PPCI center (57). At the hospital, reducing acute myocardial ischemic injury requires a treatment protocol that minimizes the door-to-PPCI time. In situations in which the transfer time to the PPCI center is prolonged, there is an opportunity for paramedics to administer a therapeutic strategy in the ambulance that can delay acute myocardial ischemic injury until PPCI has taken place. In this respect, the recently published IMMEDIATE clinical trial investigated the effects of metabolic modulation using an intravenous glucose insulin potassium (GIK) therapy administered in the ambulance to patients experiencing acute myocardial ischemia with suspected acute coronary syndrome. However, the study failed to find any difference in the primary endpoint of progression to acute MI, although patients with STEMI administered GIK therapy experienced less cardiac arrest and in-hospital mortality compared with those administered placebo (58).

Therapeutic strategies for reducing myocardial reperfusion injury

For patients presenting with a STEMI in whom acute myocardial ischemia has already taken place, the opportunity to intervene is limited to after the onset of myocardial ischemia or at the time of myocardial reperfusion (PPCI). The process of myocardial reperfusion by PPCI continues to be improved with earlier reperfusion, advances in PCI technology, and the introduction of more efficacious antiplatelet and antithrombotic agents for maintaining the patency of the infarct-related coronary artery. However, there remains no effective therapeutic agent for preventing either MVO or lethal myocardial reperfusion injury in patients with STEMI who are undergoing PPCI. Unfortunately, therapeutic targeting of the individual components of lethal myocardial reperfusion injury, including oxidative stress, calcium overload, pH correction, and, more recently, inflammation have produced disappointing results, the reasons for which are discussed below (59, 60).

However, a number of emerging therapeutic strategies for preventing lethal myocardial reperfusion injury have shown promise in small proof-of-concept clinical studies, and multicenter randomized clinical trials are currently underway to investigate the effects of these strategies on clinical outcomes in PPCI patients (Tables 1 And 2).

IPost. In contrast to unimpeded reperfusion, IPost is intermittent reperfusion of the acute ischemic myocardium, which has been reported to prevent myocardial reperfusion injury and reduce MI size by 40%–50% (61). It must be appreciated that IPost represents a form of modified reperfusion that was demonstrated in the 1980s to be beneficial in the form of gradual reperfusion (62–65). Staat et al. (66) first applied IPost to the clinical setting of PPCI: immediately after direct stenting, coronary blood reflow was allowed for 60 seconds, following which the angioplasty balloon was inflated upstream of the stent for 60 seconds to occlude coronary blood flow, and this cycle was repeated 4 times in total (Table 1). The results of this study confirmed the existence of lethal myocardial reperfusion injury in humans (67). A number of clinical studies have subsequently confirmed the beneficial effects of IPost, although not all studies have had positive results (Table 1). A large

multicenter randomized clinical trial is now underway in Denmark (DANAMI-3) to investigate the effects of IPost on clinical outcomes in PPCI patients (Clinicaltrials.gov identifier NCT01435408).

Remote ischemic conditioning. IPost requires an invasive therapeutic intervention applied directly to the heart. However, the heart can be protected against acute IRI from a distance, by applying one or more cycles of brief, nonlethal ischemia and reperfusion to another organ or tissue, a phenomenon that has been termed remote ischemic conditioning (RIC) (68, 69). In the clinical setting, RIC has been achieved noninvasively by simply inflating and deflating a blood pressure cuff placed on the upper arm to induce three 5-minute cycles of ischemia and reperfusion (70, 71). This therapeutic approach has been reported to be beneficial in patients undergoing cardiac surgery (72–75) and in patients undergoing elective PCI (76). Most recently, Botker et al. (77) demonstrated that RIC applied by a paramedic to patients with STEMI in transit to the PPCI center improved myocardial salvage compared with control patients (Table 1). Again, the patients that benefited most from this therapeutic intervention were those presenting with an anterior STEMI and an occluded coronary artery (77). Whether RIC can actually improve clinical outcomes following PPCI is currently unknown.

Therapeutic hyperoxemia and hypothermia. Two other mechanical interventions that have been reported in animal studies to be beneficial against myocardial reperfusion injury include therapeutic hyperoxemia (78) and hypothermia (79). Hyperbaric oxygen reduces MI size by decreasing tissue edema, reducing formation of lipid peroxide radicals, altering nitric oxide synthase expression, and inhibiting leukocyte adherence and plugging in the microcirculation. Lowering myocardial temperature during ischemia to 32°C–33°C can limit MI size in experimental studies by reducing metabolic demand, reducing the inflammatory response, decreasing platelet aggregation, and increasing myocardial efficiency. Proof-of-concept clinical studies in PPCI patients have demonstrated these therapeutic interventions to be potentially promising (Table 1).

Pharmacologic agents for preventing myocardial reperfusion injury. Elucidation of the mechanistic pathways underlying IPost have resulted in the identification of a number of new targets to prevent myocardial reperfusion injury. These include pharmacologic modulators of the reperfusion injury salvage kinase prosurvival pathway, such as adenosine, atrial natriuretic peptide, atorvastatin, erythropoietin, exenatide, delcasertib, and GIK (Table 2). Other agents are known to preserve mitochondrial function during acute IRI, such as cyclosporin A, sodium nitrite, and TRO40303 (Table 2). However, the results of clinical studies investigating these agents have been mixed, with the most promising pharmacologic agents being cyclosporin A (80) and exenatide (ref. 81 and Table 2).

Cardiac MRI for assessing acute myocardial IRI

Recent advances in cardiac magnetic resonance (CMR) imaging have made it possible to retrospectively assess several key features of acute myocardial IRI in patients with STEMI who have undergone PPCI treatment. Performing a CMR scan in the first week following PPCI allows the detection and the quantification of several important prognostic imaging biomarkers including MI size, MVO, and intramyocardial hemorrhage (82). There is also the potential to measure myocardial salvage, with the AAR delineated by T2-weighted imaging of myocardial edema (83–85). However, this technique for measuring the AAR has its limitations and fur-

ther study is required to validate its use (86–88). A repeat scan performed 4 to 6 months later provides an assessment of final MI size and post-MI LV remodeling in PPCI patients. Therefore, the availability of CMR allows the assessment of the efficacy of therapeutic strategies for preventing acute myocardial IRI and provides robust surrogate endpoints of cardioprotection for future clinical studies. However, it must be appreciated that surrogate endpoints to assess the efficacy of therapeutic interventions should be used to inform the design of larger, multicenter, randomized clinical trials with hard clinical endpoints.

Improving translation of cardioprotection

The research field of cardioprotection has been plagued by the large number of failed attempts to translate promising therapeutic strategies for preventing lethal myocardial reperfusion injury discovered in the basic science laboratory into the clinical setting. The reasons for this failure have been discussed in detail elsewhere (59, 60, 89, 90) and can be summarized as the use of inappropriate experimental animal models, the clinical testing of inconclusive therapies, and poor clinical trial design. Many experimental MI models fail to represent the clinical setting of an MI in terms of comorbidities (such as age, diabetes, dyslipidemia, and hypertension) and concomitant medication, the presence of which may interfere with the therapeutic cardioprotective intervention (91). The therapeutic intervention should confer conclusive cardioprotection in all experimental animal models tested before being investigated in the clinical setting. The National Heart, Lung, and Blood Institute (NHLBI) formed the CAESAR Consortium, a network of research centers in the United States (92) in which small and large animal models of experimental MI are used to investigate any promising cardioprotective strategies using an approach akin to a multicenter, randomized, controlled clinical trial. Finally, in terms of improving clinical study design, several recent strategy documents (60, 92) have suggested the following guidelines: only patients with anterior STEMI with complete occlusion of coronary flow and no collaterals should be included; the therapeutic intervention should be administered prior to myocardial reperfusion; and relevant endpoints pertinent to acute cardioprotection should be used.

Conclusions

Acute myocardial IRI is the major cause of the detrimental effects of CHD on the myocardium. This form of myocardial injury is characterized in STEMI patients who present with acute myocardial ischemia, in whom treatment priority is timely and effective myocardial reperfusion using either thrombolytic therapy or PPCI. Although improvements in myocardial reperfusion continue to take place in terms of new antiplatelet and antithrombotic agents, there is still no effective therapeutic strategy for preventing myocardial reperfusion injury. However, this is an active area of ongoing research, with the recent discovery of several mechanical and pharmacologic adjuncts to PPCI for preventing myocardial reperfusion injury. Multicenter, randomized clinical trials are now underway to investigate whether these emerging therapeutic strategies for reducing acute myocardial IRI can improve clinical outcomes in patients with CHD.

Acknowledgments

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