



Pathophysiology of Ischemia/Reperfusion-induced Myocardial Injury: What We Have Learned from Preconditioning and Postconditioning?

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ABSTRACT

Organ damage after reperfusion of previously viable ischemic tissues is defined as ischemia/reperfusion injury. The pathophysiology of ischemia/reperfusion injury involves cellular effect of ischemia, reactive oxygen species and inflammatory cascade. Protection against ischemia/reperfusion injury may be achieved by preconditioning or postconditioning. In this review, we discuss basic mechanisms involved in the pathophysiology of ischemia/reperfusion injury. Moreover, the pharmacology of preconditioning and postconditioning is delineated.

Keywords: Ischemia/reperfusion injury, Preconditioning, Postconditioning

Ischemia is the condition in which the organ is deprived from blood flow followed by inadequate oxygen and nutrient supply. Although restoration of blood flow to an ischemic organ is essential to prevent irreversible cellular injury, reperfusion per se may augment tissue injury in excess of that produced by ischemia alone. Reperfusion of the previously-ischemic myocardium is often followed by the detrimental changes in coronary arteries and myocardial tissues, which ultimately results in cardiac dysfunction, known as ischemia/reperfusion (I/R) injury. The I/R injury has been implicated in the pathology of peripheral vascular insufficiency [1], angina [2], myocardial infarction [3] and stroke [4]. Brief intermittent periods of ischemia followed by reperfusion at a time prior to prolonged ischemia, known as ischemic preconditioning, or immediately after a period of ischemia before the onset of reperfusion, known as ischemic postconditioning, have been shown to reduce I/R-induced myocardial injury [5,6]. The present review has discussed basic mechanisms involved in the pathophysiology of I/R injury and the pharmacology of preconditioning and postconditioning.

Pathophysiology of Ischemia/Reperfusion Injury

The important consequences of ischemic reperfusion are reversible contractile dysfunction known as myocardial stunning and impairment of blood flow at microvascular level known as no-reflow with neutrophil plugging and vasoconstriction. Myocardial stunning is the contractile dysfunction of heart that persists after reperfusion despite the absence of irreversible damage and despite restoration of normal or nearly normal coronary flow [7]. The impairment in re-synthesis of high energy phosphates, alteration in sympathetic responsiveness, damage to collagen matrix, leukocyte activation, transient calcium overload, decreased sensitivity of myofilaments to calcium and generation of oxygen free radicals have been implicated in the pathogenesis of prolonged contractile dysfunction in myocardial stunning [8]. The ischemic myocardium reduces its metabolic needs and tends to adopt itself to survive with minimal requirements by reducing its own contractility. Such state is referred to as hibernating myocardium in which unlike myocardial stunning, the contractility is restored immediately once the blood flow is restored. The mechanisms responsible for the development of myocardial hibernation in which the heart reduces the contractile function in proportion to reduced blood flow are vet to be identified. The calcium responsiveness in experimental myocardial hibernation has been noted to be reduced and this reduction has not been related to decreased calcium sensitivity. Another important event of prolonged postischemic reperfusion is no-reflow phenomenon in which no blood flow occurs through coronary blood vessels due to increased leukocyteendothelial cell adhesion, platelet-leukocyte aggregation, interstitial fluid accumulation and loss of endothe-



Fig. 1: Cellular and vascular effects of ischemia

lium-dependent vasorelaxation, which all together result in mechanical blood flow obstruction [9].

The cellular and vascular effects due to prolonged ischemia have been shown in Fig. 1. Ischemia reduces cellular oxidative phosphorylation and thus the synthesis of energy-rich phosphates is decreased, which alter the membrane ATP-dependent ionic pump function. This alteration favors the entry of calcium, sodium and water into cell, which ultimately leads to cellular swelling. The reduced mitochondrial oxidative phosphorylation results in loss of major source of ATP production for energy metabolism. A compensatory increase in anaerobic glycolysis for ATP production leads to accumulation of hydrogen ions and lactate, resulting in intracellular acidosis [10]. Moreover, ischemia promotes the expression of proinflammatory genes, leukocyte adhesion molecules, endothelins and thromboxane A2 [9,11], which all together may affect the integrity of coronary vascular endothelium. Polymorphonuclear leucocytes (PMNs) are mobilized from intravascular space to the interstitium during hypoxia, and such responses may contribute significantly to tissue damage during subsequent reperfusion [12,13]. The migration of PMNs through the endothelial barrier may disrupt such tissue barriers and create the potential for extravascular fluid leakage and oedema formation [14].

The adenine nucleotide catabolism during ischemia leads to intracellular accumulation of hypoxanthine, which subsequently generates reactive oxygen species (ROS) upon reperfusion (Fig. 2). During ischemia, cellular ATP is degraded to form hypoxanthine. Under normal condition, hypoxanthine is oxidized by xanthine dehydrogenase to xanthine, but during ischemia, xanthine dehydrogenase is converted to xanthine oxidase. Unlike xanthine dehydrogenase, which uses nicotinamide adenine dinucleotide as its substrate, xanthine oxidase uses oxygen and therefore, during ischemia, is unable to catalyze the conversion of hypoxanthine to xanthine, resulting in a buildup of excess tissue levels of hypoxanthine. When oxygen is reintroduced during reperfusion, conversion of the excess hypoxanthine by xanthine oxidase results in the formation of ROS [15], including superoxide anions (O_2^-) hydroxyl radicals (OH⁻), hypochlorous acid (HOCl), hydrogen peroxide (H_2O_2), and peroxynitrite. ROS directly damage cellular membranes through lipid peroxidation [11]. Further, ROS stimulate leukocyte activation and chemotaxis by activating plasma membrane phospholipase A2 to form arachidonic acid, an important precursor for synthesis of eicosanoids such as thromboxane A2 and leukotriene B4. Moreover, ROS stimulate leukocyte adhesion molecule and cytokine gene expression *via* activation of transcription factors such as nuclear factor-kB (NF-kB) [11].

Multiple mechanisms have been postulated for the leukocyte-mediated tissue injury that occurs after ischemia/reperfusion. Microvascular occlusion [16], increased vascular permeability [17] and release of oxygen free radicals [18], cytotoxic enzyme [19] and inflammatory cytokines [20] have been demonstrated to contribute to leukocyte-induced tissue injury. I/Rinduced leukocyte activation has been noted to release ROS, proteases and elastases, which result in increased microvascular permeability, edema, thrombosis, and cell death [11,21,22]. Various signaling systems such as tumor necrosis factor- α (TNF-α) [23-25], Rho-kinase [26,27], NF-κB [28], janus kinase (JAK/STAT) [29,30], poly (ADP-ribose) polymerase (PARP) [31,32], p38 mitogen activated protein kinase (MAPK) [33], Caspases [34-35], interleukin-1 (IL-1) [36] and IL-6 [37] have been implicated in the pathophysiology of I/R injury. Further, polymorphonuclear leukocyte (PMN) [38,39] and factor associated with neutral sphingomyelinase activation (FAN) [40] have been noted to play a pivotal role in affected myocardium. Moreover, resident cardiac mast cells play a key role in I/R injury. Mast cells originate from pluripotent progenitor cells in bone marrow and are major players in the inflammation process [41,42]. Degranulation of mast cells releases various cytotoxic mediators, which have been noted to be involved in the pathophysiology of ischaemia/reperfusion injury [43,44].



Fig. 2: Formation of ROS in ischemia-reperfusion injury. ATP indicates adenosine triphosphate, AMP indicates adenosine monophosphate, ROS indicates reactive oxygen species

Preconditioning

In 1986, Murry and colleagues described an endogenous protective strategy in which multiple brief ischemic episodes in canine hearts limited infarct size from a subsequent sustained ischemic insult termed as ischemic preconditioning (IPC). The IPC has two phases of protection in which an early phase is lasting from few minutes to hours known as early preconditioning, and a late phase starts after 12 hours and lasts up to 3 days is referred to as delayed preconditioning [5,45,46].

Following the finding of IPC by Murry and colleagues, several studies have investigated the mechanisms involved in its organ protective effects. Determining the mechanisms by which IPC confers myocardial preservation may eventually lead to the development of therapies to reduce cardiomyocyte injury following cardiopulmonary bypass. These studies led to the discovery that preconditioning could be induced by pharmacological means [47,48] in which drugs are administered before the ischemic event, with an intervening washout period before ischemia known as true pharmacological preconditioning or without a washout period known as pharmacological pretreatment. Preconditioning may be triggered by substances like adenosine, bradykinin, NO, diazoxide, a mitochondrial ATP sensitive K⁺ channel (KATP) opener, PKC activators, opioids and prostaglandins [49]. In addition, anesthetics were investigated for their potential to precondition the heart before ischemia. All halogenated, volatile substances were found to be protective and their actions were comparable to that of ischemic preconditioning [50,51]. In consequence, this form of preconditioning was introduced into the clinical

setting. Indeed, studies in cardiac surgery confirmed the efficacy of this anesthetic preconditioning that postoperative Troponin I in preconditioned hearts was noted to be significantly reduced [51].

Short periods of ischemia in remote vessels or even distant organs protected the myocardium from injury induced by coronary artery ischemia/reperfusion. Thus, substances must have been released from the remote ischemic-reperfused tissue that protected the jeopard-ized myocardium [52]. The occlusion of circumflex artery has produced protection of myocardium supplied by left anterior descending coronary artery and this phenomenon is termed as intracardiac preconditioning [53]. Short occlusion of renal artery [54,55] abdominal aorta [56,57] and mesenteric artery [58-60] have been documented to protect myocardium against I/R-induced injury. This phenomenon is termed as remote preconditioning [61-62] or intra organ preconditioning [62] or preconditioning at a distant site [63].

Postconditioning

Brief intermittent episodes of ischemia and reperfusion, at the onset of reperfusion after a prolonged period of ischemia confer cardioprotection, a phenomenon is termed as ischemic postconditioning. This concept was first introduced by Zhao and colleagues [6].In a canine left anterior descending coronary artery ligation (LAD) model, they compared the protective effects of IPC to that of postconditioning. The brief ischemia and reperfusion of 30 seconds each after prolonged ischemia significantly reduced infarct size and endothelial dysfunction [6]. The word postconditioning was given since the stimulus (10-30 seconds for 3-6 times) is applied after a period of ischemia.



Fig. 3: Schematic representation of active and passive mechanisms of postconditioning

It has been proposed that passive and active phases are involved in cardioprotective mechanisms of postconditioning. The passive phase is initiated via stepwise reperfusion that reduces the delivery of oxygen radicals and mitochondrial Ca²⁺ overload. In active phase, the reperfusion injury salvage kinases (RISK) pathways which include PI3K, Akt and ERK-1/2 are activated by endogenous stimulators such as adenosine, opioids and some unidentified endogenous substances [64-66]. Postconditioning mediated activation of PI3 kinase, Akt and subsequently eNOS inhibit the opening of mitochondrial permeability transition pore (mPTP) to afford cardioprotection (Fig. 3) [67,68]. Further, postconditioning activates p70s6K through MEK 1/2 and ERK 1/2 signaling systems that initiates protein translation to mediate cardioprotection. It has been suggested that postconditioning mediated cardioprotection is likely produced via the ERK1/2 pathway rather than PI3 kinase/Akt pathways [69]. On the other hand, it has been noted that Akt and ERK activated during postconditioning do not protect myocardium from reperfusion injury [70]. These contradictory reports need future investigations.

Pharmacological agents like adenosine administered initially at reperfusion have been shown to be cardioprotective is known as pharmacological postconditioning [71-73]. The pharmacological postconditioning may reduce the effective dose of a cardioprotective drug. Inhalational anaesthetics such as isoflurane [74,75] and sevoflurane [76] given just before the onset of reperfusion reduce infarct size, termed as anaesthetic postconditioning. The administration of isoflurane and sevoflurane at the onset of reperfusion has decreased infarct size through activation of K_{ATP} channels and opening of mPTP [76,77].

Following to an established remote preconditioning, remote postconditioning has been demonstrated. The concept of remote postconditioning is not merely laboratory inquisitiveness, but it may have clinical application [78]. The occlusion of the renal artery immediately before the onset of reperfusion of a coronary artery significantly reduced myocardial infarct size [79]. One may have significant nervousness in applying cyclical angioplasty balloon inflations in the target coronary artery. An alternative approach is to stimulate postcon-

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ditioning via another part of body like leg, which is much more accessible than kidney [79]. A tourniquet could be applied to the leg during transport to the emergency room to set up a strong postconditioning strategy. The tourniquet could then be removed just before completion of the angioplasty procedure and in advance of the onset of reperfusion to afford cardioprotection in first minutes of reperfusion [80].

Preconditioning and Postconditioning: A Fleeting Look

Preconditioning and postconditioning have many features in common, but they have important differences too. The immediate difference between preconditioning and postconditioning is the impact of the timing of application. While preconditioning can stimulate adaptive changes in risk areas that increase the tissue tolerance to ischemia, postconditioning can have no such biochemical or molecular effect. In postconditioning, the trigger, mediator and effector released would be active during early reperfusion, but in preconditioning, the adaptive changes take place before or during ischemia. It has been reported that the degree of infarct size reduction was similar in postconditioning as well as preconditioning; however, these two interventions have not been additive [81]. In contrast, an additive effect of these two cardioprotective strategies in the in vivo rabbit model has been demonstrated [82]. It is unclear whether such additive effects are sensitive to duration of ischemia since Yang et al has employed 45 min ischemia and 3 hrs reperfusion. The existing data suggest that preconditioning and postconditioning express the same physiological and cellular aspects of protection. Both provide infarct size reduction, attenuation of endothelial dysfunction, reduction in neutrophil adherence to coronary vascular endothelium and tissue accumulation, reduction in superoxide anion generation in postischemic myocardium, reduction of reperfusion arrhythmias, similar engagement of survival kinases, inhibition of the permeability transition pore and reduction in apoptosis [83-85]. Adenosine, PI3 kinase and Akt pathways are commonly involved in the cardioprotecive effects of preconditioning and postconditioning. However, these protective strategies may differ since ERK1/2 has not been noted to be involved in preconditioning, but may be involved in postconditioning [82]. The involvement of effectors such as KATP channels and mPTP in both preconditioning and postconditioning would also suggest common pathways, but the time at which these pathways exert cardioprotection may differ. However, the classical differences between preconditioning and postconditioning are yet to be well clarified and distinguished.

Clinical Relevance of Preconditioning and Postconditioning

Numerous *in vitro* findings suggest that the human myocardium can be protected by ischemic preconditioning. In addition, the existence of this phenomenon *in vivo* has also been well demonstrated. Intermittent aortic cross-clamping before the sustained period of global

ischemia required for the insertion of coronary artery bypass grafts during cardiac surgery has been shown to provide cardioprotection [86]. The findings from many preclinical studies in which cardioprotection has been seen in healthy animal hearts might not be reproducible in the human myocardium since human ischemic heart disease is frequently associated with various disorders such as diabetes mellitus and its complications and left ventricular hypertrophy etc or with other contributing factors like older age. The presence of these conditions might interrupt with the protection induced by ischemic preconditioning [87,88]. The use of pharmacologic agents to target different components of the signaling pathway that mimic the protection induced by ischemic preconditioning, known as pharmacologic preconditioning, might enable this approach to be recognized as a clinical therapy [89,90]. Nicorandil has cardioprotective effects when given as an adjunctive therapy at the time of reperfusion in cardiac patients during surgery [91,92]. Other promising agents such as adenosine and inhibitor of Na⁺/H⁺ exchanger have been shown clinically to afford cardioprotection when given as an adjunct to reperfusion. Preclinical studies demonstrated that pharmacologic inhibition of Na⁺/H⁺ exchanger before myocardial ischemia could reduce infarct size through a reduction in myocardial calcium accumulation, to a level comparable to ischemic preconditioning [93,94]. Adenosine has been shown to be a great promising cardioprotective agent in different clinical settings of myocardial I/R [95].

The postconditioning has been shown to be effective in patients with coronary artery disease. The patients undergoing percutaneous coronary intervention were subjected to repeated balloon inflation (n = 10, 90 sec each) after angioplasty markedly reduced the magnitude of ST-segment elevation compared to controls [96,97]. The postconditioning with 4 cycles of 1 min reinflation followed by 1 min deflation of the angioplasty balloon in patients with total coronary artery occlusion showed reduced infarct size. Marked improvement in coronary blood flow has been noted in postconditioned patients [98]. Taken together, postconditioning would be a safe cardioprotective intervention to reduce reperfusion injury in patients with ischemic heart diseases.

CONCLUSION

The postconditioning and preconditioning strategies have the drawback of intermittent cross clamping of artery, which may be harmful in patients with severe artery disorders. A significant limitation of IPC has been the inability to apply this maneuver clinically except in situations where the ischemic event can be predicted. The advantages of postconditioning over preconditioning are that it can be initiated after an ischemic insult. Unlike preconditioning, postconditioning does not require initiation prior to the ischemic event. This aspect offers several interesting opportunities to cardiac surgeons. Postconditioning may be a safe and efficient cardioprotective strategy to reduce reperfusion injury in patients with ischemic heart diseases. However, further studies are warranted to investigate the cardioprotective role of postconditioning in broader patients of coronary artery disease with hypertension, hypercholesterolemia, obesity and diabetes. In addition, studies are obligatory to recognize the major signaling mechanisms involved in the cardioprotective effects of postconditioning, which may open a novel vista to use pharmacological interventions in the name of pharmacological preconditioning to limit lethal reperfusion injury during cardiac surgery.

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REFERENCES

- Muller V, losonczy G, vannay U, Fekete A, reusz G, Tulassay T and Szabo AJ. Sexual dimorphism in renal ischemia-reperfusion injury in rats: Possible role of endothelin. *Kidney International*. 2002; 62: 1364-71.
- Verma S, Paul WM, Fedak RD. Weisel MD. Butany J, Rao V, Maitland A, Li RK, Dhillon B, Yau TM. Fundamentals of Reperfusion Injury for the Clinical Cardiologist. *Circulation*, 2002; 105: 2332-6.
- McDonough JL, Arrell DK, Van Eyk JE. Troponin I Degradation and Covalent Complex Formation Accompanies Myocardial Ischemia/Reperfusion Injury. *Circ Res*.1999; 84: 9-20.
- Oliver CN, Starke-reed PE, Stadtman ER, Lidt GJ, Carneyt JM, and Floydt RA. Oxidative damage to brain proteins, loss of glutamine synthetase activity, and production of free radicals during ischemia/ reperfusion-induced injury to gerbil brain. *Neurobiology* 1990; 87: 5144-7.
- Murry CE, Jennings RB, Reimer KA. Preconditioning with ischemia: a delay of lethal cell injury in ischemic myocardium. *Circulation* 1986; 74: 1124–36.
- Zhao ZQ, Corvera JS, Halkos ME, Kerendi F, Corvera ME, Kerendi F, Wang NP, Guyton RA, Vinten-Johansen J. Inhibition of myocardial injury by ischemic postconditioning during reperfusion: comparison with ischemic preconditioning. *Am J Physiol Heart Circ Physiol* 2003; 285: H579-88.
- 7. Bolli R. Mechanism of myocardial "stunning." *Circulation*, 1990; 82:723-38.
- Ferrari R and Visioli O. Particular outcomes of myocardial ischaemia: Stunning and hibernation. *Pharmacol Res* 1995; 31: 235-41.
- Maxwell SR, Lip GY. Reperfusion injury: A review of the pathophysiology, clinical manifestations and therapeutic options. *Int J Cardiol* 1997; 58: 95-117.
- Buja LM. Myocardial ischemia and reperfusion injury. Cardiovase Pathol 2005; 14: 170-5.
- 11. Carden DL, Granger DN. Pathophysiology of ischemiareperfusion injury. J Pathol 2000; 190: 255-66.
- Collard CD, Park KA, Montalto MC, Alapati S, Buras JA, Stahl GL, Colgan SP. Neutrophil-derived glutamate regulates vascular endothelial barrier function. J Biol Chem 2002; 277: 14801-11.
- Eltzschig HK, Ibla JC, Furuta GT, Leonard MO, Jacobson KA, Enjyoji K, Robson SC, Colgan SP. Coordinated adenine nucleotide phosphohydrolysis and nucleoside signaling in posthypoxic endothelium: role of ectonucleotidases and adenosine A2B receptors. J Exp Med 2003; 198: 783-96.

- Luscinskas FW, Ma S, Nusrat A, Parkos CA, Shaw SK. Leukocyte transendothelial migration: a junctional affair. Semin Immunol 2002; 14: 105-13.
- Charles D, Collard M.D, Simon G. Pathophysiology, Clinical Manifestations, and Prevention of Ischemia–Reperfusion Injury. Anesthesiology 2001; 94: 1133–8.
- Zoppo DGJ, Schmid-Schonbein GW, Mori E, Copeland BR, Chang CM. Polymorphonuclear leukocytes occlude capillaries following middle cerebral artery occlusion and reperfusion in baboons. Stroke 1991; 22: 1276-83.
- Bjork J, Hedqvist P, Arfors KE. Increase in vascular permeability induced by leukotriene B4 and the role of polymorphonuclear leukocytes. Inflammation 1982; 6: 189-200.
- Fujita MSH, Wakabayashi Y, Morita I. Cell adhesion molecule mediates endothelial cell injury caused by activated neutrophils. Keio Journal of Medicine 1996; 45: 207-12.
- 19. Weiss SJ: Tissue destruction by neutrophils. New Engl J Med 1989; 320: 365-76.
- Chamoun F, Burne M, O'donnell M, Rabb H. Pathophysiologic role of selectins and their ligands in ischemia reperfusion injury. Frontiers in Bioscience 2000; 5: E103-9.
- Panes J, Perry M, Granger DN. Leukocyte endothelial cell adhesion: Avenues for therapeutic intervention. Brit J Pharmacol 1999; 126: 537-50.
- Balakumar P, Singh AP, Singh M. Rodent models of heart failure. J Pharmacol Toxicol Meth 2007; 56: 1-10.
- Wang M, Tsai BM, Crisostomo PR, Meldrum DR. Tumor necrosis factor receptor-1 signaling resistance in the female myocardium during ischemia. Circulation 2006; 114: I-282-9.
- Zhang C, Xu X, Potter BJ, Wang W, Kuo L, Michael L, Bagby GJ, Chilian WM. TNF-α contributes to endothelial dysfunction in ischemia/reperfusion injury. Arterioscler Thromb Vasc Biol 2006; 26: 475-80.
- Balakumar P, Singh M. Anti-TNF-α therapy in heart failure: Future directions. Basic Clin Pharmacol Toxicol 2006; 99: 391-7.
- Takemoto M, Sun J, Hiroki J, Shimokawa H, Liao JK. Rhokinase mediates hypoxia-induced down-regulation of endothelial nitric oxide synthase. Circulation 2002; 106:57–62
- Balakumar P, Singh M. Differential role of Rho-kinase in pathological and physiological cardiac hypertrophy in rats. Pharmacology 2006; 78: 91-7.
- Squadrito F, Altavilla D, Squadrito G, Saitta A, Deodato B, Arlotta M, Minutoli L, Quartarone C, Ferlito M, Caputi AP. Tacrolimus Limits Polymorphonuclear Leucocyte Accumulation and Protects Against Myocardial Ischaemia-Reperfusion Injury. J Mol Cell Cardiol 2000; 32: 429-40.
- Bolli R, Dawn B, Xuan YT. Emerging role of the JAK-STAT pathway as a mechanism of protection against ischemia/reperfusion injury. J Mol Cell Cardiol 2001; 33: 1893-6.
- Mascareno E, El-Shafei M, Maulik N, Sato M, Guo Y, Das DK, Siddiqui MA. JAK/STAT signaling is associated with cardiac dysfunction during ischemia and reperfusion. Circulation 2001; 104, 325-9.
- Pieper AA, Walles T, Wei G, Clements EE, Verma A, Snyder SH and Zweier JL. Myocardial postischemic injury is reduced by poly ADPripose polymerase-1 gene disruption. Mol Med 2000; 6: 271–82.
- Pacher P, Szabo C. Role of Poly(ADP-ribose) polymerase -1 (PARP-1) in Cardiovascular Diseases: The Therapeutic Potential of PARP Inhibitors Cardiovasc Drug Rev 2007; 25, 3.
- Wang M, Tsai BM, Turrentine MW, Mahomed Y, Brown JW, Meldrum DR. p38 mitogen activated protein kinase mediates both death signaling and functional depression in the heart. Ann Thorac Surg 2005; 80: 2235-41.
- Pomerantz BJ, Reznikov LL, Harken AH and Dinarello CA. Inhibition of caspase 1 reduces human myocardial ischemic dysfunction via inhibition of IL-18 and IL-1β. Proc Natl Acad Sci 2001; 98: 2871–6.

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- 35. Taki J, Higuchi T, Kawashima A, Fukuoka M, Kayano D, Tait JF, Matsunari I, Nakajima K, Kinuya1 S, and Strauss HW. Effect of postconditioning on myocardial 99mTc annexin-V uptake: Comparison with ischemic preconditioning and caspase inhibitor treatment. J Nucl Med 2007; 48:1301–07.
- Suzuki K, Murtuza B, Smolenski RT, Sammut IA, Suzuki N, Kaneda Y, Yacoub MS. Overexpression of interleukin-1 receptor antagonist provides cardioprotection against ischemiareperfusion injury associated with reduction in apoptosis. Circulation 2001; 104: I-308-13.
- Vries DK, Schaapherder AFM. Pelt JV and Lindeman JHN. IL-6 mediates early ischemia–reperfusion injury in man. Vasc Pharmacol, 2006; 453: E139.
- Springer TA. Traffic signals for lymphocyte recirculation and leukocyte emigration: the multistep paradigm. Cell 1994; 76: 301-14.
- Kong T, Eltzschig HK, Karhausen J, Colgan SP, Shelley CS. Leukocyte adhesion during hypoxia is mediated by HIF-1dependent induction of beta 2 integrin gene expression. Proc Natl Acad Sci 2004; 101: 10440–5.
- O'Brien NW, Gellings NM, Guo M, Barlow SB, Glembotski CC, Sabbadini RA. Factor associated with neutral sphingomyelinase activation and its role in cardiac cell death. Circulation 2003; 92: 589-91.
- Singh AP, Singh M, Balakumar P. Effect of mast cell stabilizers in hyperhomocysteinemia-induced cardiac hypertrophy in rats. J Cardiovasc Pharmacol 2008; 51: 596-604.
- Balakumar P, Singh AP, Ganti SS, Krishan P, Ramasamy S, Singh M. Resident cardiac mast cells: Are they the major culprit in the pathogenesis of cardiac hypertrophy? Basic Clin Pharmacol Toxicol 2008; 102: 5–9.
- Parikh V, Singh M. Possible role of adrenergic component and cardiac mast cell degranulation in preconditioning-induced cardioprotection. Pharmacol Res 1999; 40, 2.
- Kaur H, Parikh V, Sharma A, Singh M. Effect of amiloride a Na/H exchange inhibitor on Cardioprotective effect of ischaemic preconditioning: Possible involvement of resident cardiac mast cells. Pharmacol Res 1997; 36: 2.
- Yellon DM, Baxter GF. A "second window of protection" or delayed preconditioning phenomenon: future horizons for myocardial protection? J Mol Cell Cardiol 1995; 27: 1023-34.
- Bolli R. The late phase of preconditioning. Circ Res 2000; 87: 972–83.
- Teoh LK, Grant R, Hulf JA, Pugsley WB, Yellon DM. The effect of preconditioning [ischemic and pharmacological] on myocardial necrosis following coronary artery bypass graft surgery. Cardiovasc Res 2002; 53: 175–80.
- 48. Kevelaitis E, Oubenaissa A, Mouas C, Peynet J, Menasche P. Ischemic preconditioning with opening of mitochondrial adenosine triphosphate- sensitive potassium channels or Na/H exchange inhibition: which is the best protective strategy for heart transplants? J Thorac Cardiovasc Surg 2001; 121: 155-62.
- Post H, Heusch G. Ischemic preconditioning. Experimental facts and clinical perspective. Minerva Cardioangiol 2002; 50: 569-605.
- 50. Vaage J, Valen G. Preconditioning and cardiac surgery. Ann Thorac Surg 2003; 75: S709-14.
- Chiari P, Bouvet F, Piriou V. Anaesthetic-induced myocardial preconditioning: fundamental basis and clinical implications. Ann Fr Anesth Reanim 2005; 24: 383-96.
- Przyklenk K, Darling CE, Dickson EW, Whittaker P. Cardioprotection 'outside the box' the evolving paradigm of remote preconditioning. Basic Res Cardiol 2003; 98: 149-57.
- Przyklenk K, Baurer B, Ovize M, Kloner RA and Whittaker P. Regional ischemic "preconditioning" protects remote virgin myocardium from subsequent sustained coronary occlusion. Circulation 1993; 87: 893-9.
- Mc Clahan TB, Nao, BS, Wolke LJ, Martin BS, Metz TE and Gallagher, KP. Brief renal occlusion and reperfusion reduces myocardial infarct size in rabbits. FASEB 1993; 7: A118-23.

- 55. Pell TJ, Baxter GF, Yellon DM, Drew GM. Renal ischemia preconditioning myocardium role of adenosine receptors and ATP-sensitive potassium chennals. Am J Physiol 1998; 275: H1542-7.
- Weinbrenner C, Nelles M, Herzog, N, Sarvary L, Strasser RH. Remote preconditioning by intrarenal occlusion of the aorta protects the heart from infarction: a newly identified non-neuronal but PKC-dependent pathway. Cardiovasc Res 2002; 55: 590-601.
- Singh M, Sharma A. Mechanism of cardioprotective effect of remote aortic preconditioning. In pathophysiology of cardiovascular diseases.2004; Dhalla NS, Angel RA and Pierce, GN (eds) Kluswer Academic Publishers, Boston, 275-85.
- Gho BC, Schoemaker RG, Vanden Deol MA, Dunker DJ, Verdouw PD. Myocardial protection by brief ischemia in non cardiac tissue. Circulation 1996; 84: 2193-200.
- Liem DA, Verdouw PD, Ploeg H, Kazim S, Duncker DJ. Sites of action adenosine in interorgan preconditioning of the heart. Am J Physiol Heart Circ Physiol 2002; 283: H29-37.
- 60. Singh D, Chopra K. Evidence of the role of angiotensin AT(1) receptors in remote renal preconditioning of myocardium. Meth Find Exp Clin Pharmacol 2004; 26, 117-22.
- Heusch G, Schulz R. Remote preconditioning J Mol Cell Cardiol 2002; 34: 1279 81.
- Wang Y, Xu H, Mizogychi K, Oe M, Mata H. Intestinal ischemia induces rate preconditioning against myocardial infarction. A role of inducible nitric oxide synthase. Cardiovasc Res 2001; 49, 391-8.
- 63. Schoemaker RG, Van Heijuingan CL. Bradykinin mediates cardiac preconditioning at a distance. Circulation 2000; 278: H1571-6.
- 64. Tsang A, Hausenloy DJ, Mocanu MM, Yellon DM. Postconditioning: a form of "modified reperfusion" protects the myocardium by activating the phosphatidylinositol 3 kinase-Akt pathway. Circ Res 2004; 95: 230-2.
- Yang XM, Krieg T, Cui L, Downey JM, Cohen MV. NECA and bradykinin at reperfusion reduce infarction in rabbit hearts by signaling through PI3K, ERK, and NO. J Mol Cell Cardiol 2004; 36: 411-21.
- Morrison RR, Xing Lin Tan, Catherine Ledent, S. Jamal Mustafa, and Polly A. Hofmann Targeted deletion of A2A adenosine receptors attenuates the protective effects of myocardial postconditioning. Am J Physiol Heart Circ Physiol 2007; 293: H2523-9.
- 67. Gross ER, Gross GJ. Ligand triggers of classical preconditioning and postconditioning. Cardiovas Res 2006; 70: 212-21.
- Zhao ZQ, Vinten-Johansen J. Postconditioning: Reduction of reperfusion-induced injury. Cardiovasc Res 2006; 70: 200-11.
- Darling CE, Jiang R, Maynard M, Whittaker P, Vinten-Johansen J, Przyklenk K. Postconditioning via stuttering reperfusion limits myocardial infarct size in rabbit hearts: role of ERK1/2. Am J Physiol Heart Circ Physiol 2005; 289: H1618-26.
- Schwartz LM, Lagranha CJ. Ischemic postconditioning during reperfusion activates Akt and ERK without protecting against lethal myocardial ischemia-reperfusion injury in pigs. Am J Physiol Heart Circ Physiol 2006; 290: H1011-8.
- Kin H, Zhao ZQ, Wang NP, Halkos ME, Kerendi F, Guyton RA, Vinten-Johansen J. Pharmacological enhancement of postconditioning (PEP-C) increases myocardial salvage after acute myocardial infarction. Circulation 2005; 112(Suppl II): II-309.
- Yang XM, Philipp S, Downey JM, Cohen MV. Postconditioning's protection is not dependent on circulating blood factors or cells but involves adenosine receptors and requires PI3-kinase and guanylyl cyclase activation. Basic Res Cardiol 2005; 100: 57–63.
- Mykytenko J, Kerendi F, Reeves JG, Kin H, Zatta AJ, Jiang R, Guyton RA, Vinten-Johansen J, Zhao ZQ. Long-term inhibition of myocardial infarction by postconditioning during reperfusion. Basic Res Cardiol 2007; 102: 90–100.
- Lucchinetti E, da Silva R, Pasch T, Schaub MC, Zaugg M. Anaesthetic preconditioning but not postconditioning prevents early

activation of the deleterious cardiac remodeling programme: evidence of opposing genomic responses in cardioprotection by pre and postconditioning. Br J Anaesth 2005: 1–13.

- Feng J, Fischer G, Lucchinetti E, Zhu M, Bestmann L, Jegger D, Arras M, Pasch T, Perriard JC, Schaub MC, Zaugg M. Infarctremodeled myocardium is receptive to protection by isoflurane postconditioning. Anesthesiology 2006; 104:1004-14.
- Obal D, Dettwiler S, Favoccia C, Scharbatke H, Preckel B, Schlack W. The influence of mitochondrial KATP-channels in the cardioprotection of preconditioning and postconditioning by sevoflurance in the rat in vivo. Anesth Analg 2005; 101: 1252-60.
- Feng J, Lucchinetti E, Ahuja P, Pasch T, Perriard J-C, Zaugg M. Isoflurane postconditioning prevents opening of the mitochondrial permeability transition pore through inhibition of glycogen synthase kinase 3β. Anesthesiol 2005; 103: 987–95.
- Andreka G, Vertesaljai M, Szantho G, Font G, Piroth Z, Fontos G, Juhasz ED, Szekely L, Szelid Z, Turner MS, Ashrafian H, Frenneaux MP, Andreka P. Remote ischemic postconditioning protects the heart during acute myocardial infarction of pigs. Heart 2007; 93: 749-52.
- Kerendi F, Kin H, Halkos ME, Jiang R, Zatta AJ, Zhao ZQ, Guyton RA, Vinten-Johansen J. Remote postconditioning: brief renal ischemia and reperfusion applied before coronary artery reperfusion reduces myocardial infarct size via endogenous activation of adenosine receptors. Basic Res Cardiol 2005; 100: 404-12.
- Kin H, Zhao ZQ, Sun HY, Wang NP, Corvera JS, Halkos ME, Kerendi F, Guyton RA, Vinten-Johansen J.. Postconditioning attenuates myocardial ischemia-reperfusion injury by inhibiting events in the early minutes of reperfusion. Cardiovasc Res 2004; 62: 74-85.
- Halkos ME, Kerendi F, Corvera JS, Wang NP, Kin H, Payne CS, Sun HY, Guyton RA, Vinten-Johansen J, Zhao ZQ. Myocardial protection with postconditioning is not enhanced by ischemic preconditioning. Ann Thorac Surg 2004; 78: 961-9.
- Yang XM, Proctor JB, Cui L, Krieg T, Downey JM, Cohen MV. Multiple, brief coronary occlusions during early reperfusion protect rabbit hearts by targeting cell signaling pathways. J Am Coll Cardiol 2004b; 44: 1103-10.
- Argaud L, Gateau-Roesch O, Raisky O, Loufouat J, Robert D, Ovize M. Postconditioning inhibits mitochondrial permeability transition. Circulation 2005; 111: 194-7.
- Crisostomo PR, Wairiuko GM, Wang M, Tsai BM, Morrell ED, Meldrum DR, Preconditioning Versus Postconditioning: Mechanisms and Therapeutic Potentials. J Am Coll Surg 2006; 202: 797-812.
- Hausenloy DJ, Yellon DM. Survival kinases in ischemic preconditioning and postconditioning. Cardiovasc Res 2006; 70: 240-53.
- Jenkins DP, Steare SE, Yellon DM. Preconditioning the human myocardium: recent advances and aspirations for the development of a new means of cardioprotection in clinical practice. Cardiovasc Drugs Ther 1995; 9: 739-47.

- Schulman D, Latchman DS, Yellon DM. Effect of aging on the ability of preconditioning to protect rat hearts from ischemia– reperfusion injury. Am J Physiol Heart Circ Physiol 2001; 281: H1630-6.
- Tsang A, Hausenloy DJ, Mocanu MM, Carr RD, Yellon DM. Preconditioning the diabetic heart: the importance of Akt phosphorylation. Diabetes 2005; 54: 2360-4.
- Ramzy D, Rao V, Weisel RD. Clinical applicability of preconditioning and postconditioning: The cardiothoracic surgeons's view. Cardiovas Res 2007; 70: 174-80.
- Vinten-Johansen J, Zhao ZQ, Jiang R, Zatta AJ, Dobson GP. Preconditioning and postconditioning: innate cardioprotection from ischemia-reperfusion injury. J Appl Physiol 2007;103: 1441-8.
- 91. Ono H, Osanai T, Ishizaka H, Hanada H, Kamada T, Onodera H, Fujita N, Sasaki S, Matsunaga T, Okumura K. Nicorandil improves cardiac function and clinical outcome in patients with acute myocardial infarction undergoing primary percutaneous coronary intervention: role of inhibitory effect on reactive oxygen species formation. Am Heart J 2004; 148: E15.
- Kloner RA, Rezkalla SH. Preconditioning, postconditioning and their application to clinical cardiology. Cardiovas Res 2007; 70: 297-307.
- Avkiran M and Marber MS. Na+/H+ exchange inhibitors for cardioprotective therapy: progress, problems and prospects. J Am Coll Cardiol 2002; 39: 747–53.
- 94. Yellon DM and Hausenloy DJ. Realizing the clinical potential of ischemic preconditioning and postconditioning. Nature 2005; 2: 11.
- Quintana M, Kahan T, Hjemdahl P. Pharmacological prevention of reperfusion injury in acute myocardial infarction. A potential role for adenosine as a therapeutic agent. Am J Cardiovasc Drugs 2004; 4: 159–67.
- Laskey WK. Brief repetitive balloon occlusions enhance reperfusion during percutaneous coronary intervention for acute myocardial infarction: a pilot study. Catheter Cardiovasc Interven 2005; 65: 361-7.
- 97. Thibault H, Piot C, Ovize M. Postconditioning in man. Heart Fail Rev 2007; 12: 245–248.
- Staat P, Rioufol G, Piot C, Cottin Y, Cung TT, L'Huillier I, Aupetit JF, Bonnefoy E, Finet G, André-Fouët X, Ovize M. Postconditioning the human heart. Circulation 2005; 112: 2143-8.

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