Translation of remote ischaemic preconditioning into clinical practice

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Reduction of the burden of ischaemia-reperfusion injury is the aim of most treatments for cardiovascular and cerebrovascular disease. Although many strategies have proven benefit in the experimental arena, few have translated to clinical practice. Scientific and practical reasons might explain this finding, but the unpredictability of acute ischaemic syndromes is one of the biggest obstacles to timely application of novel treatments. Remote ischaemic preconditioning—which is a powerful innate mechanism of multiorgan protection that can be induced by transient occlusion of blood flow to a limb with a blood-pressure cuff—could be close to becoming a clinical technique. Several proof-of-principle and clinical trials have been reported, suggesting that the technique has remarkable promise. We examine the history, development, and present state of remote preconditioning in cardiovascular disease.

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Introduction

Acute myocardial infarction and stroke are the leading causes of death and morbidity worldwide.[1] Recognition that early restoration of blood flow (reperfusion) is crucial to reduce organ damage and improve outcomes prompted development of treatment with thrombolytic drugs for acute coronary syndromes and thrombotic stroke.[2] Recently, focus has shifted towards direct restoration of vessel patency by primary angioplasty for evolving myocardial infarction.[3] Although these strategies have undoubtedly had a major effect on outcome, we have come to understand that reperfusion itself is an important cause of end-organ damage. Starting almost immediately after restoration of blood flow, a cascade of adverse events leads to a vicious cycle of cell death and local and widespread inflammatory responses and injury, which increase the extent of infarction in otherwise viable tissue.[4]

Although ischaemia and reperfusion are rarely as longlasting or complete as are unpredictable clinical events, they are necessary elements in treatment of many cardiovascular diseases. Cross-clamping of the descending aorta before repair of abdominal aortic aneurysm, and of the ascending aorta before cardiac surgery (albeit modified by use of cardioplegia and cardiopulmonary bypass), are perhaps the most overt examples, but even brief local ischaemia (eg, during therapeutic balloon angioplasty) can be associated with measurable cardiac ischaemia-reperfusion injury, and similar injury occurs with several other procedures.

Many experimental techniques have been shown in the laboratory to modify profoundly the extent of reperfusion injury. Very few of these techniques have reached clinical practice, either because of poor effectiveness when applied to the complex biology of human disease, or, as in the case of local ischaemic preconditioning, because of difficulties in application of the stimulus when and where needed in patients at risk.[5–7] Almost 25 years after local ischaemic preconditioning—the most potent innate mechanism of protection that can be induced in our tissues—was first described, the idea was in danger of not becoming implemented in practice, rather than a potent clinical approach.[8] However, the discovery that we might be able to induce this protective state systemically with a remote stimulus might change all that.

We discuss remote ischaemic preconditioning as it pertains to cardiovascular disease, and the potential it has as a rapidly translatable clinical technique to modify predictable and unpredictable ischaemia-reperfusion syndromes.

Search strategy and selection criteria

We searched Web of Knowledge, Medline, and Embase using the search term "remote preconditioning", in combination with "ischaemic". We also searched the reference lists of articles identified by this search strategy and selected those that we judged relevant. Several review articles were included because they provide comprehensive overviews that are beyond the scope of this Review. The reference list was modified during the peer-review process on the basis of comments from reviewers.

What is preconditioning?

Murry and colleagues[8] first described myocardial preconditioning in a canine experimental model in 1986. They showed that exposure of the circumflex coronary artery territory to brief periods of ischaemia (four cycles of 5 min of ischaemia followed by reperfusion) before 40 min of complete ischaemia substantially reduced the extent of infarction after restoration of blood flow. This protective effect was lost if the myocardium was rendered ischaemic for 3 h, emphasising the need for early reperfusion, irrespective of the circumstances. Nonetheless, this important finding showed that the heart could be rendered resistant to a clinically relevant ischaemia-reperfusion insult. We have subsequently learned that the ability to undergo preconditioning is almost ubiquitous in tissues and is highly conserved across species. Although local preconditioning remains the most powerful innate

cytoprotective technique studied in experimental models, its triggers, subcellular mediators, and effector pathways show intraspecies and interspecies variability that should be taken into account when reviewing the vast amount of research about this subject.[9]

Despite this caveat, the final common pathway (at least in acute resistance to ischaemia and reperfusion) is induction of a cascade of intracellular kinases and subsequent modification of mitochondrial function within the cell, via opening of ATP-sensitive potassium channels[10] and closure of mitochondrial permeability transition pores.[11] However, the precise nature (plasmalemmal or mitochondrial) and role of ATP-sensitive channel involvement remains to be fully characterised.[12,13] In 1993, Marber and colleagues[14] described a so-called second window of protection.[14] The classic protection induced by brief ischaemia wanes after a few hours (first window of protection), then recurs after 24–48 h, and can persist for up to 3–4 days (second window). Although classic and second window preconditioning share common underlying mechanisms, including triggers, transduction mechanisms, and effectors, downstream effects and cell properties differ.[15]

Experimental work has suggested that effects of preconditioning occur largely through modulation of responses to reperfusion,[16] and consequently the theory that the conditioning stimulus could be effective only when delivered before ischaemia was challenged. Local ischaemic postconditioning, in which reperfusion is interrupted by further brief periods of ischaemia, followed by continued reperfusion also induced cardioprotection in experimental models.[17] However, this system also needs direct local ischaemia of the target organ, which is technically difficult and has restricted use of this method of preconditioning and postconditioning in the clinical setting.

A systematic review of local ischaemic preconditioning in cardiac surgery has summarised data from 22 trials of 933 patients spanning 10 years.[18] Substantial reductions in ventricular arrhythmia, inotrope requirement, and length of intensive-care unit stay were reported. However, most of these studies were not powered for clinical endpoints, there were heterogeneity and bias with respect to several outcomes, and many were proof-of-concept studies. Furthermore, primary outcome measures were most often biomarkers of myocardial injury, and so positive effects were limited to patients in whom cardioplegia, as opposed to cross-clamp fibrillation, was used for cardioprotection. Continued evolution of cardioprotection techniques, recognition that some anaesthetics can induce cardioprotection, and changes in surgical methods have confounded the translation of these results to modern practice. Consequently, no consensus exists for the clinical use of this technically difficult intervention.

Five studies have assessed the effectiveness of local ischaemic postconditioning during primary angioplasty for acute myocardial infarction. In the first study, 30 patients were randomly assigned to local postconditioning at primary angioplasty for acute coronary syndrome. After restoration of vessel patency, reperfusion was interrupted by four cycles of 1 min of coronary reocclusion. This local postconditioning intervention reduced creatine kinase concentrations, suggesting reduced infarction.[19] This finding was confirmed in a second long-term follow-up study of 42 patients using nuclear techniques and echocardiography, which showed reduced infarction and improved left ventricular function after local postconditioning.[20] Results of other studies of local postconditioning have been consistent with these findings.[21–23]

An observational study that retrospectively assessed the number of balloon inflations during primary angioplasty for ST-elevation myocardial infarction showed that patients with four or more balloon inflations had reduced release of cardiac enzymes. These data further lent support to the notion of postconditioning.[24] Understandably, inclusion criteria for these studies were very specific, and this intervention is, of course, only possible in angioplasty reperfusion therapy.

The relevance of these biological effects to clinical effects also remains to be shown. Neither local preconditioning nor postconditioning has been studied with respect to the second window of protection, for practical and methodological reasons, and no large study has been undertaken of a local preconditioning or postconditioning strategy that was powered to show effectiveness on hard clinical endpoints. Thus, although these techniques have some potential as clinical methods, their use will probably be restricted by their inherent technical limitations. Furthermore, when delivered locally, the conditioning stimulus itself might be associated with local tissue injury, which has undoubtedly reduced the attractiveness of local preconditioning to practitioners. The situation might be different if the same extent of protection could be induced without the need to intervene directly with the target organ. Thus, a substantial amount of research exists about exercise-induced cardioprotection, which might have clinical use in predictable ischaemia, and various pharmacological interventions targeting preconditioning pathways are possible.[25,26] Tissue protection can also be achieved with a remote ischaemic stimulus.

What is remote ischaemic preconditioning?

Karen Przyklenk and colleagues[27] developed the idea of remote preconditioning in the early 1990s. They showed that brief ischaemia of the circumflex artery reduced subsequent infarction in the territory of the left anterior descending artery, a process termed intraorgan preconditioning.

The notion was extended in a series of experiments in rodents, showing that, for example, transient ischaemia of the kidney[28] or small bowel[29] induced protection against subsequent myocardial infarction. There are now results showing that, in animals, transient ischaemia of a wide range of tissues induces a systemic effect with multiorgan protection (including the brain) against subsequent extended ischaemia-reperfusion injury.[30–37] This preconditioning at a distance, or remote ischaemic preconditioning, as it is more commonly known, recapitulates the amount of protection seen with local preconditioning,[38] seems to work by inducing similar intracellular kinases and changes in mitochondrial function,[35,39–41] and also has an early phase and second-window phase of protection.[31,42] However, recent results suggest that there are important mechanistic differences between local and remote preconditioning.[43]

The exact nature of signal transduction from remote tissue to target organ remains to be fully elucidated. Early results showing abrogation of the event by the ganglion blocker hexamethonium[29] implicated neural pathways in transfer of the preconditioning stimulus. This finding developed into the hypothesis that there is an afferent signal from the remote organ that stimulates the efferent limb of the reflex in distant tissues. In animal studies, adenosine, bradykinin, and calcitonin gene-related peptide are important mediators in the afferent loop of this reflex.[33,44,45] The efferent signal has not been well characterised, but local release of adenosine might be important for mediation of cardioprotection. Some of these results are species-specific, but a neural mechanism is important in human remote preconditioning.[46] Recent research has emphasised the necessity of intact neural pathways to the organ or tissue receiving the preconditioning trigger,[47] but not to the target organ.

Results of our own porcine study,[48] showing that remote ischaemic preconditioning (with a transient limb ischaemia stimulus) led to subsequent protection within a transplanted heart, excluded the need for intact afferent nerves to the heart, and lent support to the role of a circulating substance or group of substances in mediation of the protective effect. This finding concurs with the previous result that coronary effluent from an isolated heart that has been locally preconditioned protects against myocardial infarction when used to perfuse a naive heart in a Langendorff preparation.[49,50]

The nature of the circulating substance is unknown and might vary with species or stimulus, but it could function through opioid, endocannabinoid, or angiotensin-1 receptors and other G-protein-coupled receptors. Naloxone blocks the effects of protection from preconditioned coronary effluent,[51] and cardioprotection from mesenteric remote ischaemia is blocked by inhibition of the cannabinoid CB_2 receptor.[52] Losartan blocks the cardioprotective effect of renal remote preconditioning in rats,[32] and noradrenaline plays a pivotal part in the process.[53] Investigators have studied the role of nitric oxide in early remote conditioning using pharmacological inhibitors, with conflicting results; however, in delayed-phase conditioning, the evidence lends support to an important part for inducible nitric oxide synthase.[30,54–57]

Demonstration that transient ischaemia—eg, of intra-abdominal organs—could protect the heart against myocardial infarction was fundamental to development of the notion of remote preconditioning.[28,29] Clinical use of such a stimulus is almost as restricted as is local preconditioning of the target organ. However, in an experimental study in rabbits, myocardial protection was shown after low-flow ischaemia induced by femoral artery stenosis combined with electrical muscular stimulation of the gastrocnemius muscle, but not with femoral artery stenosis or electrical stimulation of the muscle alone.[58] This study has shown how a straightforward, clinically applicable stimulus can induce remote preconditioning—transient limb ischaemia by tourniquet or blood-pressure cuff.

In 2001, our group subsequently showed that four cycles of 5 min of ischaemia followed by 5 min of reperfusion of the arm protected against endothelial dysfunction induced by subsequent longlasting ischaemia in the other arm. In a second part of the study, a similar preconditioning stimulus to the hindlimb protected against myocardial infarction in pigs undergoing 40 min occlusion of the left anterior descending coronary artery.[59] Although transient limb ischaemia seems superficially similar to other remote preconditioning stimuli, data suggest that the signalling cascade and effectiveness vary dependent on species studied and type of stimulus applied. These differences have been carefully documented in a review by Hausenloy and Yellon.[60]

Remote preconditioning by limb ischaemia liberates one or more bloodborne effectors that circulate to have multiorgan protective effects.[61] In a Langendorff preparation, infarct size after coronary artery ligation and reperfusion was substantially reduced by remote preconditioning in vivo. This finding was associated with upregulation of mitogen-activated protein kinases P42 and P44, and subcellular redistribution of protein kinase C ε. Pretreatment with plasma and dialysate of plasma (obtained with 15 kDa cutoff dialysis membrane) from donor rabbits undergoing remote preconditioning similarly protected against infarction. The effectiveness of dialysate in this process was abrogated by passage through a C18 column, but eluate from this column provided the same amount of protection.

The dialysate of remote preconditioning plasma from rabbits and volunteers was also tested in an isolated fresh cardiomyocyte model of simulated ischaemia and reperfusion. Necrosis in cardiomyocytes treated with dialysate was substantially lower than in control

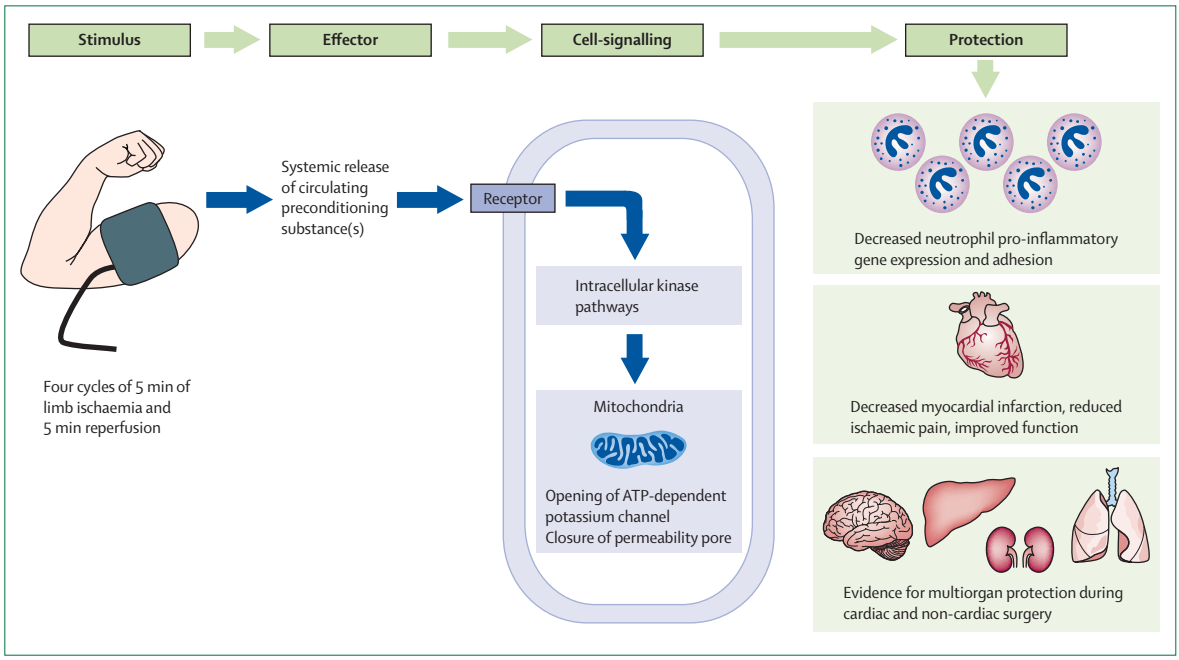

Figure: **Biological effects of remote ischaemic preconditioning**
Transient ischaemia of the arm liberates a circulating effector that induces remote cellular adaptation to a subsequent, extended, and potentially lethal period of ischaemia in remote tissues.

cardiomyocytes, and was similar to that of cells pretreated with classic preconditioning. This effect, mediated by remote preconditioning dialysate from rabbits, was blocked by pretreatment with the opiate receptor blocker naloxone. Thus, present knowledge suggests that transient limb ischaemia releases a low molecular weight (<15 kDa), hydrophobic, circulating factor that induces protection against myocardial ischaemia and reperfusion injury across species, is independent of local neurogenic activity, and needs opioid-receptor activation (figure).

In addition to release of neurogenic and circulating factors, we have shown that the stimulus of transient limb ischaemia has other biological effects that might be relevant to its effectiveness. In healthy volunteers, the stimulus suppressed expression of proinflammatory genes in circulating leucocytes within 15 min, and still further at 24 h.[62] These genomic changes have profound functional effects. In another study of healthy volunteers,[63] undergoing daily remote preconditioning for 10 days, neutrophil adhesion was reduced on the first day, and persisted throughout the study period. Neutrophil phagocytotic capacity was unaffected early, but was reduced substantially by the tenth day. The biological significance of this finding remains to be seen, but does ask the question: what might be the downside of cells remaining in a chronically preconditioned state?

Philosophically, such a downside has to exist, otherwise human beings would presumably have evolved to remain intrinsically protected. Nonetheless, in a murine model we showed that the stimulus also modified gene expression in the heart, with upregulation of cardioprotective genes and suppression of proinflammatory genes.[64] Furthermore, in a porcine model, the stimulus, invoked by transient limb ischaemia, reduced coronary resistance and increased coronary blood flow,[65] and this effect can be detected in healthy volunteers.[66] Whether these vascular effects are related to the myocardial effects of remote preconditioning remains unclear.

These widespread generic effects might mean that this natural method is more physiologically inclusive than are those acting through only one mechanistic pathway. In addition to cytoprotective effects that are analogous to local preconditioning, the anti-inflammatory, gene expression, and physiological changes it induces might be important to the complex biology of human ischaemia-reperfusion injury. To test this suggestion before clinical application, we did a study in a porcine model investigating the effect of remote preconditioning to protect the heart and lungs during experimental cardiac surgery with bypass and aortic cross-clamping. We showed that remote preconditioning not only reduced myocardial injury, as assessed by serial troponin release, but was also associated with improved functional recovery and preserved lung function.[67]

Preconditioning, in which the remote stimulus is applied before onset of target organ ischaemia, is not possible when clinical presentation involves established ischaemia, such as during acute myocardial infarction or stroke. Analogous to local postconditioning, remote

postconditioning has also been studied in experimental models.[68] Thus, brief periods of kidney ischaemia reduce myocardial infarction when applied after onset of coronary ischaemia, and immediately before and during reperfusion. Dose and timing of limb ischaemia have been studied in man, and differences between lower and upper limb investigated. Remote postconditioning against postischaemia-reperfusion endothelial dysfunction can be achieved by two 5-min cycles of lower-limb ischaemia, but not by the same period of upper-limb ischaemia, although three cycles of upper-limb ischaemia do prevent endothelial ischaemia-reperfusion injury.[69] Lower-limb and upper-limb ischaemia both effectively reduce myocardial ischaemia-reperfusion injury, which has direct implications for clinical trials in acute myocardial infarction. We investigated this idea in an experimental setting, and showed that remote transient limb ischaemia applied after onset of cardiac ischaemia and before reperfusion effectively reduced myocardial infarction—a stimulus we have termed perconditioning.[70] This stimulus is also effective in postconditioning,[71] and thus studies have investigated evolving ischaemia-reperfusion syndromes.

Evolution of translational studies

As the experimental and preclinical data that lent support to the effectiveness of transient limb ischaemia to induce distant organ protection grew, the potential of this process as a clinical intervention became clear. Rightly or wrongly, its simplicity and, by comparison with a novel pharmaceutical, its restricted regulatory implications have facilitated a rapid translation to clinical trials. Although we are clearly some way from using this technique in routine clinical practice, an evolving amount of research exists showing its effectiveness (table).[72–77]

During conventional on-pump cardiac surgery, the heart and lungs are subjected to ischaemia and reperfusion, and the body is perfused at low pressure. Several techniques have evolved to protect the heart and other organs during this period, but myocardial injury remains an important predictor of adverse outcome, and the secondary inflammatory process contributes to postoperative morbidity. These adverse effects are amplified in children undergoing corrective cardiac surgery, in which aortic cross-clamp and cardiopulmonary bypass times tend to be long, and complete circulatory arrest continues to be used during complex procedures in some institutions.

The first study of remote preconditioning by transient limb ischaemia showed no effect on creatine kinase release in adults undergoing coronary bypass surgery.[78] However, these results cannot be considered definitive. The preconditioning stimulus consisted of only two cycles of 3 min of ischaemia and 2 min of reperfusion of the arm, and there were only four patients in both the control and preconditioning groups. In our experimental model of cardiac bypass surgery, we showed reduced myocardial injury and improved lung function after a clinically relevant period of cross-clamping and cardiopulmonary bypass.[67]

Consequently, we did a masked, randomised study of 37 children (20 control, 17 remotely preconditioned) having corrective cardiac surgery. Bypass times were similar between the groups and standard volatile agents (which have some intrinsic preconditioning effects) were used as anaesthesia. The results were remarkably similar to those of the preclinical study. Troponin I release was substantially lower in the group receiving remote preconditioning, and the postoperative inotrope score and airway resistance in this group were lower than they were in the control group.[72] This proof-of-principle study was not powered to report clinical endpoints, but on the basis of these results a large clinical endpoint study (including neurocognitive outcomes as a measure of neuroprotection) is underway and due to be completed in late 2009. An important part of this study is the assessment of genomic responses in control and preconditioned children, since investigators might be able to profile both adverse outcomes in the control group, and responses in children receiving preconditioning.

The protective effect of remote preconditioning by limb ischaemia has also been shown in a similar proof-of-principle study of 57 adults undergoing elective on-pump coronary bypass surgery.[73] The endpoint was troponin T release, which was significantly lower in the treatment group (n=27) than in the control group (n=30). Patients were not given inhalation anaesthesia, and either cross-clamp fibrillation or cardioplegia

	Patient group	Stimulus	Outcomes	N
Cheung (2006)[72]	Paediatric cardiac surgery	Upper-limb ischaemia (4 cycles of 5 min)	Reduced troponin; reduced inotrope score; reduced airway resistance	37
Hausenloy (2007)[73]	CABG	Upper-limb ischaemia (3 cycles of 5 min)	Reduced troponin	57
Ali (2007)[74]	AAA surgery	Lower-limb ischaemia (2 cycles of 10 min)	Reduced troponin I; reduced perioperative MI; preserved renal function	82
Hoole (2009)[75]	Elective coronary angioplasty	Upper-limb ischaemia (3 cycles of 5 min)	Reduced troponin I; reduced MACCE	242
Venugopal (2009)[76]	CABG (cold-blood cardioplegia)	Upper-limb ischaemia (3 cycles of 5 min)	Reduced troponin	45
Botker* (2009)[77]	Primary coronary angioplasty (STEMI)	Upper-limb ischaemia (3 cycles of 5 min)	Increased myocardial salvage; decreased infarct size at 1 month	333

N=Number of patients. CABG=coronary-artery bypass surgery. AAA=abdominal aortic aneurysm. MI=myocardial infarction. MACCE=major adverse cardiac and cerebral event. STEMI=ST-elevation myocardial infarction. *Presented as Featured Research abstract at Scientific Sessions of American College of Cardiology, March, 2009.

Table: **Studies with results showing potentially beneficial clinical outcomes of remote preconditioning**

were used during bypass. The same researchers have also reported another small and successful study in adult patients undergoing coronary-artery bypass grafting, but with cold-blood cardioplegia only, suggesting effectiveness with other cardioprotection techniques.[76]

Thus, remote preconditioning reduces myocardial injury in children and adults undergoing cardiac surgery. However, the clinical relevance of these findings is as yet undefined, and large studies are underway in both groups of patients to address this issue. A large clinical study in adult bypass surgery has completed recruitment and will report findings in the near future (Bonser R, University Hospitals Birmingham, personal communication).

In visceral organ transplantation, the donor organ can undergo substantial periods of ischaemia before reperfusion in the recipient. Reduction of the complications of ischaemia-reperfusion injury might therefore improve graft function and survival. Results of previous studies have suggested that local preconditioning is abrogated after brain death, although this finding might not be consistent. Thus, the biology of remote preconditioning and its effectiveness in this setting remain uncertain, although we have shown that remote preconditioning of a recipient animal leads to transferable protection against ischaemia-reperfusion injury in the heart taken from a brain-dead donor.[48] This idea has been translated into clinical trials, and studies are underway in renal and liver transplantation to test the potential of remote preconditioning to reduce graft injury and improve outcome.

Non-cardiac surgery is associated with ischaemia-reperfusion syndromes. In vascular surgery, this association is not only a result of the operation itself, but also because many patients have widespread atherosclerosis, and postoperative myocardial infarction is an important complication in this setting. The ability of remote preconditioning to protect the heart and kidneys has been investigated in a clinical trial of patients undergoing elective aortic aneurysm repair.[74] Remote preconditioning was induced by intermittent femoral artery occlusion and reperfusion in 82 patients (41 controls) studied in Cambridge, UK. The results were remarkable. Not only was postoperative incidence of myocardial infarction reduced (11 of 41 *vs* two of 41, $p<0\cdot002$), but there was also a reduction in loss of renal function, and treated patients had a reduced length of critical-care stay.

Investigators have also reported on use of remote preconditioning before elective coronary intervention in 242 patients.[75] Remote preconditioning was associated with reduced pain during the procedure, reduced electrocardiographic evidence of ischaemia, and reduced troponin release. Perhaps the most intriguing finding was a reduction in major adverse cardiac and cerebral events at 6 months. This result suggests that the systemic effects of preconditioning, perhaps to reduce the proinflammatory effects of even a short-lived sentinel event, might have longlasting secondary beneficial effects. Clearly, however, this finding will need to be substantiated in larger studies. A review of ClinicalTrials.gov in March, 2009, showed that 12 registered studies used remote preconditioning by limb ischaemia, including studies to assess hepatic and neural protection.

In view of our experimental data showing effectiveness of remote perconditioning, we tested the hypothesis that remote limb ischaemia could reduce myocardial reperfusion injury during evolving ST-elevation myocardial infarction. Patients were enrolled and randomly assigned to intermittent limb ischaemia or control, both of which were received in the ambulance during transfer for angioplasty. The primary endpoint was myocardial salvage assessed by nuclear scintigraphy. The study was completed after recruitment of 333 patients, and the results will soon be formally reported. Preliminary results were presented at the American College of Cardiology in March, 2009.[77] In summary, we reported a significant increase in myocardial salvage and infarct size in patients with occlusion of the left anterior descending artery. Our results establish the theory that postischaemic intervention is feasible in man. This finding has clear implications for testing of remote perconditioning in stroke, for example.

Future potential for therapeutic applications

Remote conditioning is easy to deliver through a straightforward procedure such as intermittent ischaemia of the upper or lower limb, induced by inflation of a blood-pressure cuff. It has no known adverse risks, is cheap, and readily applicable. Early proof-of-principle clinical trials using this procedure in the setting of predictable ischaemia-reperfusion syndromes have had promising results, and other studies are underway to define the potential clinical use of remote preconditioning in a range of situations targeting the heart, brain, kidney, and liver. Since remote conditioning can be applied effectively during the ischaemic phase and early into reperfusion, limb ischaemia can be administered after onset of target-organ ischaemia, such as during myocardial infarction or stroke, while patients are being transported for reperfusion therapy. In many ways, the outlook for this clinical method seems promising.

However, the understandable enthusiasm generated by these studies should be tempered with the need to show real and tangible clinical benefit.[7,79] The exact dose of ischaemia needed and the role of late-phase remote conditioning remain to be fully defined. Furthermore, the effect of age, drugs, or coexisting disease on responses induced by remote conditioning is poorly understood. Although no adverse effects are known, why are we not preconditioned all of the time? A

downside could yet be discovered. In the modern era, any novel intervention should undergo rigorous clinical testing in large appropriately powered trials in multiple centres, with a focus on clinical outcome measures. Although remote preconditioning has been rapidly translated from experimental discovery to encouraging small-scale proof-of-principle human studies using surrogate endpoints, the next phase will be the most challenging. Large-scale, probably multicentre, studies powered to show hard clinical outcomes will be needed to change practice. But where will financial support for these trials come from? In some ways, the simplicity of the technique might be its downfall; its non-pharmacological nature will preclude sponsorship from the pharmaceutical industry. Nonetheless, the results of these studies will ultimately decide whether remote preconditioning is used in a clinical setting for cytoprotection. For the time being, we believe that the future is encouraging.

Contributors

RKK was responsible for the search and review of published work, and report drafting and revision. TTN was responsible for report revision. ANR was responsible for review of reports and report drafting and revision.

Conflicts of interest

We declare that we have no conflicts of interest.

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