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Editorial

Preconditioning: a balanced perspective

In 1986, Murry *et al* made a remarkable observation: they found that brief episodes of sublethal ischaemia in dogs protected or "preconditioned" the heart and reduced the infarct size caused by a subsequent more sustained period of coronary artery occlusion.[1] The ability of brief antecedent ischaemia to limit infarct size has since been described in every experimental model and species assessed to date. In addition, indirect evidence suggests that brief ischaemia may also protect the human heart: the improved in-hospital outcome recently described in patients experiencing angina before myocardial infarction may represent a clinical correlate with ischaemic preconditioning.[2]

The mechanisms responsible for this paradoxical protection remain unresolved, but experimental studies have demonstrated with certainty that the reduction of infarct size achieved with preconditioning is not simply due to favourable changes in myocardial oxygen supply (that is, not a consequence of increased collateral blood flow) or in haemodynamic indices of oxygen demand.[1 3] Rather, brief ischaemia seems to trigger an as yet unidentified endogenous subcellular adaptive response that acutely increases the tolerance of the myocytes to a subsequent ischaemic insult. For these reasons, ischaemic preconditioning has been hailed as "the most powerful experimental strategy known for protecting the functioning heart from infarction".[4] It is important to recognise, however, that preconditioning is not a panacea.

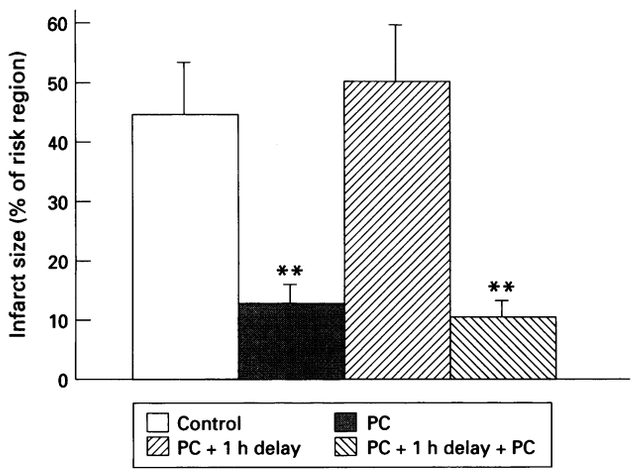

*Figure 1 Mean values of infarct size, expressed as a percentage of the myocardium at risk of infarction, after 90 minutes of sustained coronary artery occlusion in the rat model. Infarct size was significantly smaller in rats that received three 3-minute episodes of preconditioning ischaemia immediately before the sustained occlusion (PC) than in controls that received the sustained occlusion alone. This protective effect was lost when a 1 hour delay was imposed between the preconditioning regimen and the sustained occlusion (PC + 1h delay), but was re-established in rats that received a second series of three 3-minute occlusions after the 1 hour delay and immediately before the sustained occlusion (PC + 1h delay + PC). ** $P < 0{\cdot}01$ v control and PC + 1h delay. Adapted from reference 5.*

Infarct size reduction with preconditioning has temporal limitations

There are three important temporal limitations to achieving the profound myocardial salvage that is characteristic of ischaemic preconditioning. First, the sustained occlusion must follow soon (within minutes) after the brief ischaemic stimuli. For example, Li and colleagues found that in rats three 3-minute episodes of preconditioning ischaemia significantly reduced infarct size when the sustained occlusion was initiated 5 minutes after the last brief reperfusion, but if a 1 hour delay was imposed between the brief and sustained occlusions the protection was lost.[5] This temporal loss of protection could, however, be re-established. If, after the 1 hour delay, rats received a second series of brief occlusions before the sustained ischaemia, the hearts were again preconditioned (fig 1).[5] In addition, hearts preconditioned with brief ischaemia seem to show a resurgence of protection against prolonged ischaemia initiated 24 hours later—a distinct form of cardioprotection, termed the "second window",[6] which probably differs in its mechanism from acute ischaemic preconditioning.

Second, the heart becomes tolerant to numerous repeated preconditioning stimuli. Cohen *et al* reported that whereas one episode of brief ischaemia effectively preconditioned the rabbit heart, protection was attenuated when rabbits received 40–65 repeated 5-minute occlusions over 3–4 days before the sustained occlusion.[7] Once again, however, this tolerance could be reversed: in rabbits that received multiple brief occlusions followed by 3 days without intervention, and then underwent one brief occlusion before the sustained ischaemia, protection was re-established and the hearts were again effectively preconditioned.[7]

Finally, the protection achieved with preconditioning is lost with increasing durations of sustained coronary artery occlusion.[1 3] For example, in the canine model, preconditioning profoundly reduced infarct size caused by 40 minutes of occlusion, but was ineffective when the duration of the sustained ischaemic insult was extended to 3 hours.[1] These data illustrate a crucial concept: preconditioning does not prevent myocyte necrosis, but rather limits infarct size by slowing or delaying myocardial cell death.

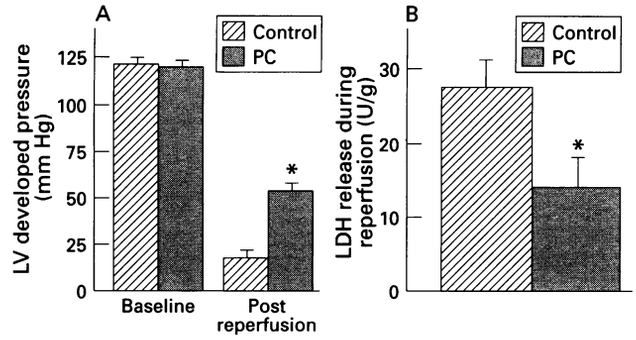

*Figure 2 (A) Mean values of left ventricular (LV) developed pressure measured at baseline and at 30 minutes after reflow, and (B) lactate dehydrogenase (LDH) released into the coronary effluent during reperfusion in isolated buffer-perfused rat hearts subjected to 30 minutes of sustained global ischaemia (control) and hearts preconditioned with four 5-minute episodes of global ischaemia before the sustained ischaemia (PC). Hearts were perfused at constant pressure. Preconditioning had a concomitant beneficial effect on recovery of global LV function and release of LDH. * $P < 0.05$ versus control. Adapted from reference 12.*

Preconditioning does not have an independent beneficial effect on acute recovery of contractile function

Though reduction of infarct size is the hallmark of preconditioning, many studies have sought to expand the definition of this phenomenon to encompass other deleterious sequelae of myocardial ischaemia/reperfusion. For example, in isolated heart preparations brief episodes of global ischaemia have been shown to enhance the recovery of left ventricular (LV) function after relief of a subsequent sustained period of global ischaemia.[8] As a result, enhanced recovery of LV function has become the "gold standard" of preconditioning in the isolated heart.

In contrast, in the in vivo setting, we and others found that preconditioning did not enhance segment shortening during the initial hours after relief of ischaemia in viable canine and porcine myocardium "stunned" by 15 minutes of sustained coronary artery occlusion.[9,10] Similarly, we found no significant difference between preconditioned dogs and controls in segment shortening of the viable but stunned peri-infarct tissue measured acutely after 1 hour of sustained coronary occlusion.[3,11]

How can this apparent discrepancy between the isolated heart models and the in vivo data be explained? There is increasing evidence that preconditioning the isolated heart both improves recovery of LV function and blunts the release of lactate dehydrogenase and creatine kinase (markers of cellular integrity) into the coronary effluent upon reflow (fig 2).[12] Thus the superior recovery of function seen after global ischaemia may simply reflect the well-established reduction in lethal cell injury seen with preconditioning. We therefore believe there is no discrepancy: preconditioning can result in an acute improvement in post-ischaemic global LV function, but this is a secondary consequence of "infarct size reduction" rather than an independent beneficial effect on viable but stunned myocardium.

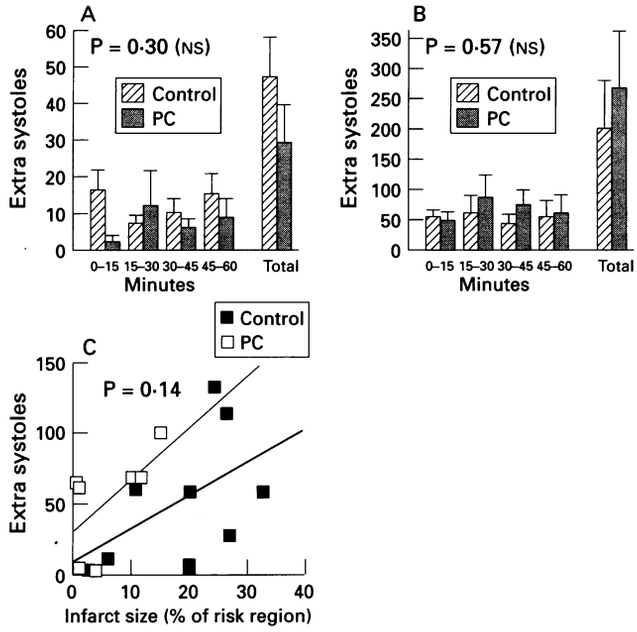

Figure 3 Number of extrasystoles per 15-minute interval measured (A) during 1 hour of sustained coronary artery occlusion and (B) during the initial hour of reperfusion in pentobarbitone anaesthetised control dogs that received no intervention before the sustained occlusion and dogs that received four 5-minute episodes of preconditioning ischaemia before the sustained occlusion (PC). Two-factor analysis of variance showed no significant difference in the number or distribution of extrasystoles between the groups. (C) Number of extrasystoles during the initial 15 minutes of reperfusion plotted as a function of infarct size (expressed as a percentage of the myocardium at risk of infarction) for all dogs in the protocol. Analysis of covariance indicates that for any infarct size, preconditioned dogs tended to have a greater number of extrasystoles upon reflow than did the controls. Adapted from reference 16.

The antiarrhythmic component of preconditioning is model dependent

A second surrogate endpoint in many preconditioning protocols has been the incidence of arrhythmias. In the rat model, there is overwhelming evidence that preconditioning can reduce the incidence of extrasystoles, ventricular tachycardia (VT), and reversible ventricular fibrillation (VF) induced by ischaemia and reperfusion.[5,13–15] Similar observations have been made in chloralose-anaesthetised dogs: Vegh *et al* found a lower incidence of VT and fatal VF during 25 minutes of sustained occlusion and upon reperfusion in preconditioned dogs compared with controls.[15]

In contrast, in our hands, preconditioning failed to attenuate the incidence of lethal VF in pentobarbitone-anaesthetised dogs. In addition, the number and distribution of extrasystoles, measured during 1 hour of sustained occlusion and the first hour of reperfusion, was not altered with preconditioning[16] and, in fact, when infarct size was included as a covariate, we found a trend toward a greater number of extrasystoles upon reperfusion in preconditioned dogs compared with controls (fig 3). Indeed, in the pig model, preconditioning had a significant proarrhythmic effect: the onset of VF was accelerated, fibrillation threshold was decreased, and action potential duration was shortened in preconditioned pigs compared with controls.[17] Thus in contrast to the consistent reports of infarct size reduction, the antiarrhythmic component of preconditioning is dependent upon the model and species—and perhaps even anaesthesia—that is used.

Preconditioning does not confer consistent protection to the coronary vasculature

Finally, a handful of studies have sought to determine whether the beneficial effects of preconditioning extend beyond the myocyte and protect the coronary vasculature from injury during ischaemia/reperfusion. For example, one indicator of damage to the coronary vessels is the well-documented progressive loss of coronary vasodilator

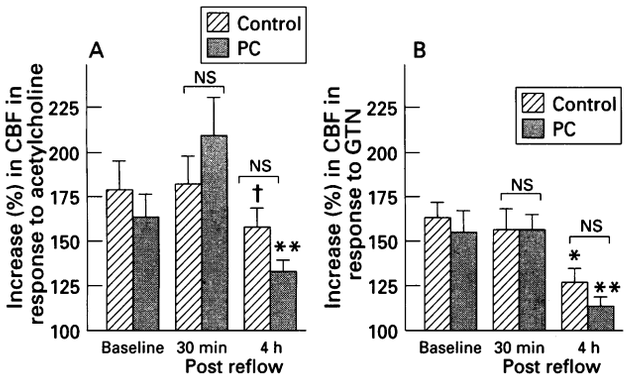

*Figure 4 Percentage increase in coronary blood flow (CBF) in response to submaximal intravenous doses of (A) the endothelium-dependent vasodilator acetylcholine and (B) the endothelium-independent vasodilator glyceryl trinitrate (GTN) assessed at baseline and at 30 minutes and 4 hours after reperfusion in control dogs subjected to 1 hour of sustained coronary artery occlusion and dogs that received four 5-minute episodes of preconditioning ischaemia before the sustained occlusion (PC). All dogs showed a significant deterioration in endothelium-dependent and endothelium-independent vasodilation at 4 hours after reperfusion when compared with baseline, with no significant difference between the control and PC groups. †P = 0·06; *P < 0·05; **P < 0·01 versus corresponding baseline value. Adapted from reference 18.*

reserve that occurs after relief of sustained ischaemia. We observed the expected loss of vasodilator reserve after 1 hour of sustained coronary occlusion in our in vivo canine model, but found no difference between preconditioned dogs and controls (fig 4).[18] Richard and colleagues, however, reported that preconditioning preserved endothelium-dependent relaxation in distal segments of rat coronary arteries excised after 20 minutes of in vivo sustained coronary occlusion.[19] We have insufficient data to determine whether this inconsistency represents a discrepancy between studies or a difference between models. Nonetheless, the results suggest that, as was the case with arrhythmias, preconditioning does not confer consistent protection on the coronary vasculature.

Implications

The seminal observation that preconditioning can, within the framework of specific temporal limitations, profoundly reduce infarct size has generated unprecedented enthusiasm and investigative effort in experimental myocardial ischaemia and infarction. Indeed, great progress has been made in defining the fundamental properties of this intriguing phenomenon. The fact that ischaemic preconditioning does not have an independent effect on contractile function, attenuates arrhythmias in some but not all models, and does not provide consistent protection to the coronary vasculature has two important implications. First, care must be taken in extrapolating general conclusions regarding the mechanisms, etc of ischaemic preconditioning exclusively from measurements of these surrogate endpoints of function, arrhythmias, and vasodilator reserve. Second, maintaining a balanced perspective of what preconditioning can and cannot do is essential in formulating a rational and productive approach to elucidating the cellular mechanisms responsible for this endogenous cardioprotection and, ultimately, exploiting this knowledge to develop "preconditioning-mimetics" to protect the human heart.

KARIN PRZYKLENK
ROBERT A KLONER

Heart Institute,
Good Samaritan Hospital
and
Department of Medicine,
Section of Cardiology,
University of Southern California,
Los Angeles, California,
USA

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