General Anesthesia

Myocardial protection by anesthetic agents against ischemia-reperfusion injury: an update for anesthesiologists

[La protection myocardique contre les lésions d'ischémie-reperfusion par des anesthésiques : une mise à jour pour les anesthésiologistes]

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Purpose: The aim of this review of the literature was to evaluate the effectiveness of anesthetics in protecting the heart against myocardial ischemia-reperfusion injury.

Source: Articles were obtained from the Medline database (1980-, search terms included heart, myocardium, coronary, ischemia, reperfusion injury, infarction, stunning, halothane, enflurane, desflurane, isoflurane, sevoflurane, opioid, morphine, fentanyl, alfentanil sufentanil, pentazocine, buprenorphine, barbiturate, thiopental, ketamine, propofol, preconditioning, neutrophil adhesion, free radical, antioxidant and calcium).

Principal findings: Protection by volatile anesthetics, morphine and propofol is relatively well investigated. It is generally agreed that these agents reduce the myocardial damage caused by ischemia and reperfusion. Other anesthetics which are often used in clinical practice, such as fentanyl, ketamine, barbiturates and benzodiazepines have been much less studied, and their potential as cardioprotectors is currently unknown. There are some proposed mechanisms for protection by anesthetic agents: ischemic preconditioning-like effect, interference in the neutrophil/platelet-endothelium interaction, blockade of Ca^{2+} overload to the cytosolic space and antioxidant-like effect. Different anesthetics appear to have different mechanisms by which protection is exerted. Clinical applicability of anesthetic agent-induced protection has yet to be explored.

Conclusion: There is increasing evidence of anesthetic agent-induced protection. At present, isoflurane, sevoflurane and morphine appear to be most promising as preconditioning-inducing agents. After the onset of ischemia, propofol could be selected to reduce ischemia-reperfusion injury. Future clinical application depends on the full elucidation of the underlying mechanisms and on clinical outcome trials.

Objectif : *Évaluer l'efficacité des anesthésiques dans la protection du cœur contre les lésions myocardiques d'ischémie-reperfusion.*

Source : *Des articles ont été obtenus de la base de données Medline (1980-, les mots clefs étant heart, myocardium, coronary, ischemia, reperfusion injury, infarction, stunning, halothane, enflurane, desflurane, isoflurane, sevoflurane, opioid, morphine, fentanyl, alfentanil sufentanil, pentazocine, buprenorphine, barbiturate, thiopental, ketamine, propofol, preconditioning, neutrophil adhesion, free radical, antioxidant et calcium).*

Constatations principales : *La protection par des anesthésiques volatils, morphine et propofol, est relativement bien explorée. On s'accorde généralement pour dire que ces agents réduisent les lésions myocardiques causées par l'ischémie et la reperfusion. D'autres anesthésiques utilisés souvent en clinique, comme le fentanyl, la kétamine, les barbituriques et les benzodiazépines, ont été moins étudiés et leur potentiel cardioprotecteur est actuellement inconnu. On propose certains mécanismes de protection par les anesthésiques : un effet qui s'apparente à un préconditionnement, une interférence dans l'interaction entre polynucléaires neutrophiles/plaquettes-endothélium, un blocage de la surcharge de Ca^{2+} à l'espace cytosolique et un effet du genre antioxydant. Différents anesthésiques semblent présenter des mécanismes différents par lesquels la protection s'exerce. L'applicabilité de la protection induite par les agents anesthésiques est encore à étudier.*

Conclusion : *Il y a de plus en plus d'évidence de la protection induite par les anesthésiques. Présentement, l'isoflurane, le sévoflurane et la morphine semblent les agents inducteurs de préconditionnement les plus prometteurs. Après le début de l'ischémie, le propofol peut être choisi pour réduire les lésions d'ischémie-reperfusion. L'application clinique future repose sur la mise en lumière complète des mécanismes sous-jacents et sur des essais relatifs aux avantages cliniques.*

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1. Introduction

Anesthesiologists frequently treat patients with ischemic heart disease, as the number of such patients presenting for surgery is increasing. Accordingly, anesthesiologists witness the frequent occurrence of perioperative myocardial ischemia. Because myocardial ischemia-reperfusion injury can lead to severe complications, measures to minimize myocardial damage have been an important target of research. Anesthetics may provide protection so, consequently, a better understanding of the role of anesthetics in the prevention of myocardial injury may provide anesthesiologists with strategies to improve outcome.

Since Freedman reported that enflurane improved postischemic functional recovery in 1985,[1] the protective effect of halogenated anesthetics against myocardial ischemia-reperfusion injury has been investigated in various animal models. There is accumulating evidence that halothane, isoflurane and sevoflurane possess such properties. The search for protection by opioids started only recently, when Schultz *et al.*[2] found that ischemic preconditioning, a powerful intervention to reduce ischemia-reperfusion injury, is mediated by opioid receptors. This study led to the discovery that exogenous morphine protects the heart against infarction.[3] Propofol is known to be a free radical scavenger[4,5] and has recently been shown to be protective.[6–10]

This review summarizes our current understanding of cardioprotection to evaluate possible protective effects in patients who are at risk of myocardial ischemia.

2. Methods

Articles examining whether an anesthetic agent provides myocardial protection against ischemia-reperfusion injury were obtained from the Medline database (1980-). Search terms included heart, myocardium, coronary, ischemia, reperfusion injury, infarction, stunning, halothane, enflurane, desflurane, isoflurane, sevoflurane, opioid, morphine, fentanyl, alfentanil, sufentanil, pentazocine, buprenorphine, barbiturate, thiopental, ketamine, propofol, midazolam, diazepam, preconditioning, neutrophil adhesion, free radical, antioxidant and calcium. The language of articles was limited to English. All articles of investigation obtained using the above-mentioned search strategy were included in this review article, except those already reviewed elsewhere.[11] Articles investigating the mechanisms of anesthetic-induced protection were searched using the same method. Recent and representative studies are cited in this narrative review.

The limitations of our search method include: 1) the article list may not be complete because: a) only one database, Medline, was used; b) the language of the articles was limited to English; and c) relevant articles written in English may have been missed in Medline;[12] 2) we did not take publication bias (negative results are less likely to be published) into consideration; 3) the process by which articles were selected was not strictly objective; and 4) the interpretation of previous findings and our conclusions remain subjective.

3. Ischemia-reperfusion injury

Myocardial ischemia initiates a range of cellular events, which are initially mild and become progressively damaging with increasing duration of ischemia. The damage caused during ischemia is called "ischemic injury". Although reperfusion means a termination of ischemia and is essential for the cell to survive and to restore normal function, it paradoxically causes damage to the cell; this injury is called "reperfusion injury". The term "ischemia-reperfusion injury" is also used to represent both types of damage together, as 1) it is not always easy to distinguish one from the other, and 2) ischemia is often accompanied by reperfusion and reperfusion cannot occur without prior ischemia.

We briefly address the mechanisms of ischemia-reperfusion injury. Further information is found elsewhere.[13–17] Ischemia precludes adequate oxygen supply, which rapidly results in depletion of adenosine triphosphate (ATP). This inhibits ATP-driven Na^+-K^+ pumps, increasing $[Na^+]_i$. $[H^+]_i$ is increased because of poor washout of metabolites and inhibition of mitochondrial oxidation of $NADH_2$. Increased $[H^+]_i$ enhances Na^+-H^+ exchange to retain normal pH_i, leading to increased $[Na^+]_i$, hence $[Ca^{2+}]_i$ is augmented via Na^+-Ca^{2+} exchange.[13,15] High $[Ca^{2+}]_i$ degrades proteins and phospholipids.[14,16] The production of free radicals is also enhanced after the onset of ischemia. They are known to be derived mainly from neutrophils and mitochondria.[13,16] The target components of free radicals involve lipids and proteins,[16] which constitutes the structure of the cell and enzymes.

Injury after the onset of ischemia is further worsened when coronary vessels are damaged. Swollen endothelial cells prevent swift gas exchange. Malfunctioning endothelium and smooth muscle cannot provide vasodilatation when necessary. Neutrophils/platelets aggregating in the lumen hamper adequate coronary flow.[14,16] Neutrophils play a central role in the propagation of damage. They are attracted towards endothelial cells and subsequently migrate across the endothelium. They also release oxygen free radicals, cytokines and other proinflammatory substances. These substances harm the endothelium, vascular smooth muscle and myocardium.[17] A pathway for neutrophil sequestration is the specific interaction of adhesion molecules whose expression is promoted by ischemia-reperfusion. They are expressed on neutrophils and endothelial cells. Intercellular adhesion molecule-1 (ICAM-1), L-selectin and CD11b/CD18 are examples of these molecules. Treatment with monoclonal antibodies against adhesion molecules results in a significant reduction of ischemia-reperfusion injury.[17]

On reperfusion, $[H^+]$ outside the cell is abruptly reduced to normal levels because it is washed out. This results in an increase in $[Ca^{2+}]_i$ due to enhanced Na^+-H^+ and Na^+-Ca^{2+} exchange.[14,15] Reperfusion also results in a burst of free radical generation because oxygen is abundantly supplied.[14,16] Ca^{2+} and free radicals injure the heart further at reperfusion.[14,16] Damage of the vascular system is more prominent during reperfusion than ischemia.[16,17]

Ischemia-reperfusion injury has various clinical consequences. Infarction is one of the major events which we witness in clinical practice. Another consequence of ischemia-reperfusion injury is myocardial stunning. This phenomenon is defined as reversible myocardial dysfunction that persists after reperfusion.[14,18,19]

4. Ischemic preconditioning

There have been many reviews on ischemic preconditioning.[20–22] In brief, Murry *et al.* introduced this concept when they found, in a canine model, that four cycles of five-minute left circumflex coronary artery (Lcx) occlusion, in advance of 40-min Lcx occlusion, reduced infarct size by 75%. They named this protective effect ischemic preconditioning,[23] a phenomenon that has been extensively investigated because of its potency. Ischemic preconditioning can be observed in a wide range of preparations: from isolated cardiomyocytes and vascular endothelial cells to hearts *in situ* in various species.[20–22,24] Although ischemic preconditioning initially referred to the ability to mitigate infarction, the definition has been extended to employ other endpoints, such as stunning[18] and coronary abnormalities.[22] In humans, ischemic preconditioning enhanced postischemic contraction in ventricular trabeculae muscle, and survival rate of isolated cardiomyocytes.[24] Ischemic preconditioning elicited by two periods of three-minute aortic cross clamping prior to cardiopulmonary bypass for valve replacement reduced myocardial enzyme leakage, free radical formation and histological degeneration and enhanced contractility after weaning from cardiopulmonary bypass.[25,26] Szmagala *et al.* applied four-minute aortic cross clamping and six-minute reperfusion before coronary artery bypass grafting; troponin (T) from blood samples was reduced.[27]

Many investigators have investigated the mechanisms of ischemic preconditioning. The current understanding of the mechanisms as they relate to myocardial protection by anesthetic agents are reviewed. Further descriptions may be found elsewhere.[20–22] Preconditioning is a treatment "prior" to an ischemic event while ischemia-reperfusion injury is formed "during" and "after" an ischemic period. In principle, the signals generated by brief periods of ischemia in ischemic preconditioning must be passed to, and preserved in, intracellular component(s). This memory should be retrieved, at the time of ischemia, resulting in the forwarding of signals to the effector(s) that can mitigate ischemia-reperfusion damage. This basic hypothesis was used to elucidate the mechanisms of ischemic preconditioning; the search for cellular contributors has continued since then. Previous studies, by and large, led to identify the constituents of a series of cellular events in the heart (Figure). Ischemic stimuli cause the release of stress mediators from the heart, including adenosine, bradykinin, opioids, noradrenaline and free radicals.[20–22] They contribute as initiators, which pass signals to intracellular components, such as inhibitory guanine nucleotide binding proteins (G_i proteins) and protein kinase C (PKC).[20–22] Eventually,

ATP-sensitive K^+ channels (K_{ATP} channels) on the sarcolemma and mitochondria are activated.[20–22] Recent studies support the view that mitochondrial K_{ATP} channels play a greater role than sarcolemmal K_{ATP} channels.[21,22] It also seems certain that ischemic preconditioning depends upon stimulation of mitogen-activated protein kinases. Yet, it is still not clear where these kinases lie in the cascade of the signal transduction; it may be downstream of PKC.[20–22] The biggest concern of this signal transduction theory is that the events which link K_{ATP} channel opening and protection remain unknown. Blockade of the identified mediators involved in ischemic preconditioning reverses the protective effect. Conversely, treatments with chemicals that stimulate these mediators salvage the heart from the insult of ischemia-reperfusion.

5. Halogenated anesthetics

5.1 Evidence of protection by halogenated anesthetics

5.1.1 Summary of evidence before 1997

Halogenated anesthetics have been studied more extensively than any other anesthetics. In 1988, Warltier and colleagues demonstrated that pretreatment with halothane or isoflurane improved left ventricular systolic function after left anterior descending coronary artery (LAD) occlusion lasting 15 min. Segmental shortening of the myocardial wall in the LAD area recovered completely over five hours following reperfusion in anesthetic-pretreated dogs, while untreated dogs showed only 50% recovery.[28] This study triggered a dramatic proliferation of investigations regarding protection by anesthetics. The progress made by mid-1997 was reviewed by Ross and Foëx.[11] Briefly, a growing number of investigations have shown that halothane, enflurane and isoflurane enhance postischemic mechanical function at equianesthetic concentrations in various animal species, with only a few exceptions in isolated heart models. A few studies have reported an infarct-reducing effect of halogenated anesthetics.

5.1.2 Sevoflurane and desflurane

Further evidence for the protective properties of halogenated anesthetics was collected for halothane and isoflurane after 1997.[29–45] A new agent whose protection was demonstrated recently is sevoflurane. This anesthetic improves postischemic mechanical and coronary function, and reduces infarct size.[32,34,37,45–50] A beneficial effect of desflurane was suggested by a few groups.[34,49,51] Further investigations are needed to determine the effect of desflurane in other experimental settings.

5.1.3 Different approaches to assess myocardial injury

Several indices other than postischemic ventricular mechanical function have been examined to assess the degree of myocardial injury in the past few years. Reduction in infarct size has been well demonstrated with volatile anesthetics.[29–31,34,38,39,41,50] Results of infarction studies are more consistent than those of postischemic contractility. Kersten and coworkers administered isoflurane to dogs for one hour before 60-min LAD occlusion. The extent of infarction was reduced from 25% to 12%.[31] According to Cope *et al.*, preconditioning with halothane, enflurane or isoflurane diminished infarct size to less than one third in rabbits.[30]

There were a few infarction studies performed in the 1980s.[52–54] The infarct-reducing effect of halothane or isoflurane was inconclusive. These studies should be distinguished from those reported after 1995 because they differ largely in their experimental protocols. Previous studies employed longer duration of ischemia (6–48 hr) without reperfusion,[52,53] whereas the ischemic period was less than one hour followed by a reperfusion period in recent studies.[29–31,34,38,39,41,50]

Another approach to evaluate postischemic injury of the heart is to examine the coronary vasculature. Halothane, isoflurane and sevoflurane reduced the number of neutrophils sequestered in the coronary vasculature after ischemia.[32,37] A similar effect was also shown for platelets.[45,47] Reduced neutrophil/platelet entrapment by anesthetics was accompanied by enhancement of postischemic mechanical function.[37,47] However, the effects of halogenated anesthetics on neutrophil/platelet sequestration have been studied by a limited number of research groups.[32,37,45,47]

Novalija and coworkers measured coronary flow changes in response to endothelial-dependent and independent vasodilators. Sevoflurane preserved the reaction provoked by both types of vasodilators during the reperfusion period better than no treatment.[48]

5.2 Mechanisms of protection by halogenated anesthetics

Before 1997, mechanisms underlying the protection were suggested to be preservation of ATP, reduction in Ca^{2+} influx to the cell, inhibition of free radical formation, and activation of K_{ATP} channels.[11] Recently, progress in elucidating the mechanisms responsible for protection was achieved when investigators: 1) related ischemic preconditioning and anesthetic-induced protection and 2) examined the coronary system.

5.2.1 Preconditioning

Phenomena similar to ischemic preconditioning have been observed with halogenated anesthetic agents;

myocardial protection persists even though anesthetics were allowed to wash out. Because evidence indicated that halogenated anesthetics dilated the coronary arteries *via* K_{ATP} channels, known as a key constituent of the ischemic preconditioning pathway, Cason *et al.* hypothesized that halogenated anesthetics induced an ischemic preconditioning-like effect. They studied rabbit hearts *in situ* treated with five-minute coronary occlusion followed by reperfusion or 15-min isoflurane followed by washout before 30-min coronary occlusion. In the ischemic preconditioning group infarction was reduced by 74% compared to the non-pretreated group, and by 30% in the isoflurane group. Although less effective than ischemic preconditioning, isoflurane limited ischemic injury even though isoflurane was washed out, i.e., isoflurane had an ischemic preconditioning-like effect.[29] Cope *et al.* perfused isolated rabbit hearts with halothane, enflurane or isoflurane for five minutes and washed out the anesthetic for ten minutes before the onset of coronary occlusion. This treatment reduced infarction to the same extent as a single five-minute coronary occlusion.[30] One might doubt that anesthetics were eliminated completely from the heart tissue by washing for ten to 15 min, and suspect that the protection may not have been solely due to preconditioning. Kersten *et al.* used a longer washout period. Canine hearts *in situ* were protected to a similar degree when isoflurane was washed out for five or 30 min.[31] As studies to date unanimously conclude that halogenated anesthetics alleviate myocardial ischemic damage even when washed out,[29–31,38–40,44,46,48] it is generally considered that volatile anesthetics can "precondition" the heart.

These findings indicate that the signals for halogenated anesthetics, like for brief ischemia in ischemic preconditioning, are preserved in intracellular components that can mediate protection against forthcoming ischemia. The protection by halogenated anesthetics has been reversed by a selective adenosine A_1 receptor antagonist,[44] a G_i protein inhibitor,[41] PKC inhibitors,[30,42] and K_{ATP} channel blockers.[31,39,40,43,44,50,51] Contribution of the mitochondrial K_{ATP} channel appears certain,[39,40,51] but not the sarcolemmal K_{ATP} channel.[51] These observations strongly suggest that halogenated anesthetic agents provide protection *via* a mechanism similar to that of ischemic preconditioning. Hence, it can be assumed that the halogenated anesthetics stimulate adenosine receptors, followed by G_i proteins, PKC and K_{ATP} channels as shown in the Figure. As the protection by halogenated anesthetics is not accompanied by augmented release of adenosine,[30] it could be assumed that they stimulate adenosine receptors *via* a non-adenosine mechanism, or upregulate the adenosine receptor-G protein complex to promote the signal transduction downstream. However, there is no evidence to support this hypothesis.

A mechanism similar to that of ischemic preconditioning is very plausible. Nonetheless, there have been reports that suggest other mechanism(s). According to Schlack and coworkers, halogenated anesthetics have shown protection when they were administered only during ischemia[55] or reperfusion.[34,35,49] Protection was also observed with administration of these agents in a cardioplegic solution.[33] In addition, Yao *et al.* showed that isoflurane reduced stunning by a mechanism which is independent of the adenosine A_1 receptor.[56]

5.2.2 Coronary vasculature

According to recent reports, halothane, isoflurane and sevoflurane reduced the number of trapped neutrophils in the heart during reperfusion.[32,37] The postischemic expression of CD11b, which forms an integrin with CD18, was also suppressed by volatile anesthetics.[37] Kowalski *et al.* showed that neutrophil adhesion was attenuated even when sevoflurane was administered only during reperfusion.[32] Mobert *et al.* treated neutrophils and endothelial cells with volatile anesthetics, and measured the extent of activation in both cell types when they were pharmacologically stimulated. The anesthetic treatment decreased neutrophil adhesion on the endothelium and expression of CD11b, while the anesthetics did not affect endothelial cell activation *vis-à-vis* neutrophils.[57] These studies imply that halogenated anesthetics can act directly on neutrophils at the time of reperfusion. Volatile anesthetics also reduced platelet adhesion to the vascular wall after ischemia,[45,47] but sevoflurane failed to reduce the expression of glycoprotein IIb/IIIa, a platelet adhesion molecule involved in the platelet-endothelium interaction.[47]

Another possibility of coronary protection by halogenated anesthetics is *via* an ischemic preconditioning-like effect. Ischemic preconditioning is known to reduce ICAM-1 production and neutrophil entrapment, and to preserve the response to vasodilators.[22]

When ischemia is regional, one way to slow the progression of ischemic injury is to increase coronary collateral flow. Kersten *et al.* showed that sevoflurane selectively increased collateral flow to the ischemic area in dogs with chronic LAD stenosis. This effect was not reversed by glibenclamide, a non-selective K_{ATP} channel blocker.[58]

5.2.3 Energy metabolism

ATP is required by numerous ongoing chemical reactions and ion pumps to maintain cell integrity. The less

ATP consumed by contractile work, the less the myocardium suffers during ischemia. Because of their distinctive negative inotropic and vasodilative effect, it has long been thought that halogenated anesthetics might lessen myocardial injury by reducing ATP consumption during ischemia.[11] However, it is unlikely that protection derives solely from this negative inotropic effect, since the extent of protection has been shown to be independent of mechanical function during ischemia[30,43,48]

6. Opioids

6.1 Evidence of protection by opioids

Protection by opioid receptor agonists against ischemia-reperfusion injury has been demonstrated during the last several years. Contribution of endogenous opioid peptides to adaptation to hypoxia had been suggested[59] when Mayfield and D'Alecy, in 1994, examined whether exogenous opioid receptor agonists can increase tolerance to hypoxia. They found that D-Pen2-D-Pen5-enkephalin (DPDPE), a δ-opioid agonist, can extend survival time of mice under severe hypoxia.[60] Meanwhile the δ-agonist D-Ala2-D-Leu5-enkephalin (DADLE), which had been identified as a hibernation-inducing trigger in nature, showed protective effects in multi-organ preparations, including the heart, preserved for transplant.[61]

In 1996, Schultz and colleagues were the first to demonstrate that an opioid could attenuate ischemia-reperfusion damage in the heart. Morphine at the dose of 300 $\mu g \cdot kg^{-1}$ was given before LAD occlusion for 30 min in rats *in vivo*. Infarct area/area at risk was diminished from 54 to 12% by this treatment.[3] The infarct-reducing effect of morphine has been shown in hearts *in situ*, isolated hearts and cardiomyocytes.[62–64] Morphine also improved postischemic contractility.[65] Consequently, it is now well accepted that morphine provides protection against ischemia-reperfusion injury. Kato *et al.* demonstrated that fentanyl enhances postischemic mechanical function in isolated rat hearts.[66,67] However, this opioid did not show protection in isolated rabbit hearts according to Benedict *et al.*.[65] The discrepancy might be due to differences in species and/or fentanyl concentrations. Pentazocine and buprenorphine improved postischemic contractility in rabbits *in vitro*.[65,68] Overall, the effects of opioids other than morphine have not been sufficiently investigated to allow conclusions to be drawn.

The concomitant use of opioids, cardioplegia and hypothermia has been investigated. Morphine, pentazocine and buprenorphine improved mechanical function after global total ischemia for two hours at 34°C.[65,68] Opioids also elicited a protective effect in heart preservation at 4 or 10°C for four to18 hr;[68–70] this might be an indication of the usefulness of opioids in heart transplantation.

6.2 Mechanisms behind the protection by opioids

6.2.1 Preconditioning

The involvement of opioid receptors in ischemic preconditioning has been demonstrated in various animal species and humans.[2,62,63,71-77] Among opioid receptor subtypes, there is evidence that δ-opioid receptors are responsible for ischemic preconditioning in rats[71,75,78,79] and humans.[76] Although opioid receptors are more abundant in the central nervous system, they are also present in the heart.[74,76] Opioid receptor subtype distribution in the heart appears to differ between species. δ- and κ-, but not μ-opioid receptors are expressed in the rat heart.[74] In human atrium, δ- and μ- have been shown to be dominant compared to κ-receptors.[76] Naloxone blocked the effect of ischemic preconditioning in isolated hearts,[73] and cardiac myocytes.[63] Quaternary naloxone, which does not cross the blood-brain barrier, eliminated the protection by ischemic preconditioning in *in vivo* models.[72,73] Therefore it is suggested that it is in the heart itself that opioid receptors play a role in protection by ischemic preconditioning.

There is evidence that exogenous opioids precondition the heart. In the study by Liang *et al.* cardiac myocytes were treated with morphine followed by incubation in drug-free media, before ischemia was induced. The survival rate of morphine-treated cells was higher.[63] Yellon and colleagues treated isolated hearts with DADLE for two five-minute cycles, each of which was followed by five-minute drug-free perfusion. This treatment diminished infarct size.[75] A similar effect was observed in human trabeculae.[76]

Morphine and fentanyl are capable of binding to δ- and κ-receptors although they are preferentially μ-agonists.[80] It appears that the δ-opioid receptor subtype is responsible for opioid-induced protection.[64,67,75,76,78,81-86] Selective δ-[64,75,76,83,84] and δ_1-[81,82,85–87]agonists have shown protection. Conversely protection by morphine and fentanyl is abolished by δ-antagonists.[64,67,78,83] The role of κ-receptors remains controversial.[75,86,88,89] Whether μ-receptors contribute to cardioprotection in humans is not known. Other cellular mediators which pass signals in ischemic preconditioning (Figure) are also involved in the protection conferred by opioid agonists. The beneficial effects were eliminated by a G_i protein inhibitor,[81] a PKC inhibitor,[62,67,85,86] and a selective mitochondrial K_{ATP} channel blocker.[63,66,70,76,82,83,85,86]

The effect of "classic" or "early" ischemic preconditioning is transient and lost within 0.5–2 hr after brief

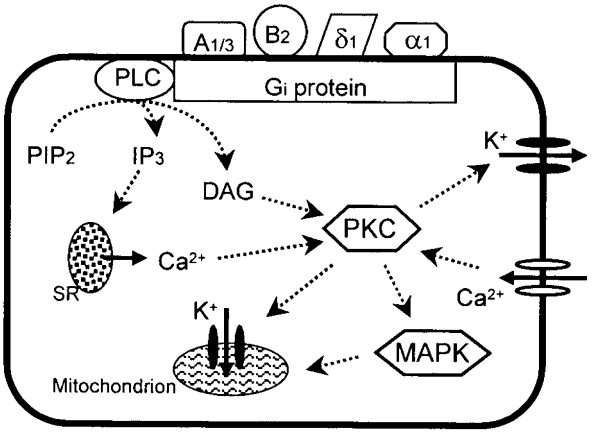

FIGURE Cellular mechanism of ischemic preconditioning. Activation of adenosine A_1 and A_3 ($A_{1/3}$), bradykinin$_2$ (B_2), δ_1-opioid (δ_1) and α_1-adrenergic (α_1) receptors stimulates phospholipase C (PLC) through inhibitory guanine nucleotide-binding proteins (G_i proteins). PLC produces diacylglycerol (DAG) and inositol 1,4,5-triphosphate (IP_3). Both DAG and IP_3 activate protein kinase C (PKC). PKC enhances opening of ATP-sensitive K^+ channels (K_{ATP} channels), probably via mitogen-activated protein kinases (MAPK). The K_{ATP} channel seems to be the end- effector for inducing protection, though the downstream process is still unknown. Recently mitochondrial K_{ATP} channels have been considered more important than sacrolemmal K_{ATP} channels. PIP_2 = phosphatidyl inositol; SR = sarcoplasmic reticulum.

ischemic episodes, but the protection exhibits a biphasic time course and returns after 24 hr.[20,22] This late protection is called "late", "delayed" or "second window" preconditioning. TAN-67, a selective δ_1-agonist, also provoked "late" preconditioning. The opioid showed protection one hour after its *iv* administration, but not after 12 hr. Then, the protective effect resumed after 24 hr and was not observed after 72 hr.[82] A κ-agonist also showed late preconditioning-like effect.[89]

6.2.2 Neutrophil adhesion and migration

The neutrophil hypothesis is much less convincing than the preconditioning hypothesis due to the lack of supporting evidence. There have been reports that morphine reduced plasma levels of ICAM-1, L-selectin and gp100^{MEL14} (equivalent to L-selectin in rats) during ischemia and reperfusion.[90,91] This observation was obtained when hearts were pretreated with morphine and also when morphine was administered only during reperfusion.[90] Morphine reduced adhesion molecules in a PKC and K_{ATP} channel-independent manner.[91] In this model it seems likely that morphine exerted a direct effect on reperfusion injury rather than mimicking ischemic preconditioning. Hofbauer *et al.* reported that remifentanil decreased neutrophil adhesion and transmigration, and ICAM-1 expression in a dose-dependent manner. Fentanyl also reduced migration of neutrophils.[92] By contrast, Szekely *et al.* reported that fentanyl, at higher concentration than that used by Hofbauer *et al.*, did not reduce postischemic neutrophil adhesion in the guinea pig heart.[93]

7. Propofol, ketamine, barbiturates and benzodiazepines

It has been shown that propofol decreases postischemic myocardial mechanical dysfunction, infarct size and histological degeneration.[6–10] This anesthetic has a chemical structure similar to that of phenol-based free radical scavengers such as vitamin E, and reduces free radicals.[4,5] Propofol attenuates $[Ca^{2+}]_i$.[94,95] Propofol also suppresses the activity of neutrophils.[96,97] These features suggest that propofol may directly intervene at the critical phase of reperfusion injury by reducing free radicals, Ca^{2+} influx and neutrophil activity, but does not act as a preconditioning-inducing agent. Indeed, protection was observed when the heart was treated with propofol solely during reperfusion.[7] Addition of glibenclamide, a K_{ATP} channel blocker, did not abolish the protection by propofol.[8]

Whether barbiturates protect the heart is still controversial, although these drugs were studied in the 1980s (Table I). Ketamine and benzodiazepines have hardly been investigated for myocardial protection (Table I). Whether these *iv* anesthetics interfere with neutrophil-endothelium interaction, Ca^{2+} influx and free radical production still needs to be elucidated (Table II). It seems unlikely ketamine has an ischemic preconditioning-like effect. This anesthetic blocked ischemic preconditioning,[98] reduced the production of inositol 1,4,5-triphosphate[99] and deactivated sarcolemmal K_{ATP} channels.[100] Thiamylal also inhibited sarcolemmal K_{ATP} channel activity.[101]

8. Comparisons between anesthetics

Several halogenated anesthetics, administered at the same minimum alveolar concentration (MAC) and in the same preparation, have been assessed for their protective effect in a number of investigations. Overall, there is little difference in the *extent* of protection among halogenated anesthetics. However, some studies have reported different protective properties between halogenated anesthetics. The results are not consistent with respect to which anesthetic agents are more protective than others. In addition, an explanation for the discrepancies between anesthetics is not readily available.[34,35,44,102]

There has been no evidence to suggest significant differences among *iv* anesthetics, such as propofol, ketamine and barbiturates.[30,103,104] Benedict *et al.* examined the effect of several opioids of equal potency on postischemic myocardial contractility. Morphine, buprenorphine and pentazocine improved postischemic function to a similar degree, while fentanyl did not.[65] According to Ross *et al.* postischemic contractility recovered to a similar degree in fentanyl- and propofol-anesthetized dogs.[105]

Cope *et al.* reported that infarct size was smaller when rabbits were anesthetized with halothane, enflurane or isoflurane compared to pentobarbital, ketamine/xylazine or propofol.[30] Because the volatile anesthetics caused blood pressure to be lower than the *iv* anesthetics, the question was raised whether the difference in blood pressure was a determinant of infarct size, but there was no correlation between the two parameters within the halothane-anesthetized group.[30] Haessler *et al.* reported that there was no significant difference in the extent of infarction between isoflurane, ketamine/xylazine and pentobarbital in rabbits *in vivo*. Yet, isoflurane showed a tendency toward smaller infarction.[103] Similarly, significant differences were not found between sevoflurane and propofol assessed by postischemic ventricular function.[8] However, it is not easy to establish the equipotency of doses for different types of anesthetics and, no clear justifications for choosing the dosage of anesthetics were presented in most studies. Hence the comparisons between anesthetics might not have been strictly valid. Meissner *et al.* compared desflurane and propofol at the concentrations that prevented movement at skin incision. The recovery from stunning was faster with desflurane.[106] However, in this study, the extent of ventricular wall dyskinesia seemed worse (although not statistically different) in the propofol-anesthetized group during the ischemic period; it is possible that the intensity of ischemic insult differed between the anesthetic groups.

9. Studies in humans

Because of obvious limitations, it has been difficult to provide evidence for anesthetic preconditioning in humans. In the study by Roscoe *et al.*, the protective effect of halothane and isoflurane was investigated in right atrial trabecular muscles obtained from patients undergoing coronary bypass grafting surgery. When the trabeculae were pretreated with isoflurane, the contractile force after the ischemic insult was improved. However, halothane did not show significant improvement.[44] The only study of opioids in humans is by Bell *et al.* DADLE administered to atrial trabeculae improved contraction.

Clinical studies have been conducted in patients undergoing coronary artery bypass graft surgery. Belhomme *et al.* assessed the effect of isoflurane on T I release. Preconditioning by isoflurane reduced T I release, but not to a statistically significant degree.[107] According to Tomai and coworkers, isoflurane suppressed the release of this enzyme in patients with compromised left ventricular function.[108] The same group employed postischemic mechanical function as another endpoint. Enflurane was administered immediately before starting cardiopulmonary bypass in coronary artery bypass graft surgery. Peak-systolic pressure in the ascending aorta was plotted against the end-diastolic area of the left ventricle. The postischemic change in the slope of this relationship was less pronounced in the enflurane group.[109] A limitation of these clinical studies is that the number of subjects was small. Taking into account patient variability, the number of subjects would have to be large for valid conclusions to be drawn.

10. Limitations of currently available data

All investigations of halogenated anesthetics cited in this review have demonstrated protection using anesthetic concentrations relevant to clinical practice, i.e., 0.5–2 MAC. Morphine diminished infarct area at a dose of 0.3 $mg \cdot kg^{-1}$ in rats *in vivo*;[3,78,91] this concentration is clinically relevant. Total plasma concentrations of morphine can reach 0.7 $\mu mol \cdot L^{-1}$ after 0.2 $mg \cdot kg^{-1}$ *iv* bolus injection for peripheral surgery.[110] When *iv* morphine was used for analgesia after breast reconstruction or operation on the spine, the plasma concentration was about 0.1 $\mu mol \cdot L^{-1}$.[111] Assuming morphine's protein binding to be 40%,[112] the concentrations used in some *in vitro* studies (0.3 $\mu mol \cdot L^{-1}$,[62] 0.1 $\mu mol \cdot L^{-1}$)[63] are close to the clinical range, but not others (100 $\mu mol \cdot L^{-1}$ [64], 1 $\mu mol \cdot L^{-1}$).[83] Fentanyl reached 0.07 $\mu mol \cdot L^{-1}$ and remained 0.03–0.04 $\mu mol \cdot L^{-1}$ after bolus dosage of 30 $\mu g \cdot kg^{-1}$ followed by 0.3 $\mu g \cdot kg^{-1} \cdot min^{-1}$ for cardiac surgery.[113] As fentanyl binds to plasma protein at the rate of 82%,[112] 0.47 $\mu mol \cdot L^{-1}$,[66,67] the concentration used in experimental studies is outside the clinical range. Propofol provides protection at 18–100 $\mu mol \cdot L^{-1}$ in rats,[6–9] while free plasma concentration during surgery is estimated to be less than 1 $\mu mol \cdot L^{-1}$.[112,114] Propofol failed to protect against reperfusion injury at a concentration of 4.5 $\mu mol \cdot L^{-1}$ in rats.[115] In the case of ketamine, neutrophil adhesion during reperfusion was inhibited at clinically relevant concentrations.[116,117] In summary, concentrations of *iv* anesthetics which protect the heart were not always within the therapeutic range for humans. However, it should be noted that pharmaco-

TABLE I Protection by different anesthetics

	Timing of administration		
	Preconditioning	*Preischemia/ Pre-, during ischemia*	*During and/or postischemia*
Halogenated	++ (5.2.1)	++ [41,43,109,118]	± (5.2.1)
Opioids	++ (5.2.1)	++ [3,62,64,70]	(10)
Propofol		++ (7)	(7)
Ketamine		[93]*1	
Barbiturates		± [93]*1 [119–121]	+ [120,122]
Benzodiazepines		[93]*1	

The effect is sorted according to the timing of anesthetic administration. + = supported by published studies; ++ = supported by published studies by multiple institutes; ± = published studies are conflicting; – = negated by published studies. Numbers in ( ) indicate sections in the text where these issues are discussed. *1 = the anesthetic was given pre-, during and postischemia.

TABLE II Possible mechanisms by which an anesthetic agent confers protection

	Mechanisms			
	Preconditioning	*Neutrophil*	*Ca^{2+}*	*Free radicals*
Halogenated	++ (5.2.1)	+ (5.2.2)	++ [11]	+ [11]
Opioids	++ (6.2.1)	± (6.2.2)	± [123,124]	+ [125,126]
Propofol		++ (7)	++ (7)	++ (7)
Ketamine	– (7)	+ [93,127]	+ [128,129]	
Barbiturates		+ [93,96,130]	± [131,132]	+ [126,133]
Benzodiazepines		± [93,97,130]	+ [95,134]	

Different agents might have different mechanisms. Preconditioning = preconditioning-like effect in the heart; neutrophil = interference in neutrophil function; Ca^{2+} = blockade of Ca^{2+} overload in the myocyte; free radicals = antioxidant-like effect; + = supported by published studies; ++ = supported by published studies by multiple institutes; ± = published studies are conflicting; – = negated by published studies. Numbers in ( ) indicate sections in the text where these issues are discussed.

dynamics are largely species-dependent. It is, therefore, difficult to determine whether these *iv* anesthetics could be cardioprotective at the concentrations used in humans.

It is well demonstrated that anesthetics are protective when administered before ischemia. However, unlike cardiopulmonary bypass and transplantation, myocardial ischemia is not usually planned or expected in clinical practice, and in many cases patients can be treated only after myocardial ischemia is established. Are anesthetics effective when they are given during and/or after ischemia? As summarized in Table II, anesthetics have attributes which may contribute to protection when administered after the onset of ischemia, such as mitigation of Ca^{2+} overload, free radical production and neutrophil adhesion. Indeed, postischemic treatment with halogenated anesthetics[34,35,49,55] and propofol[7] had favourable effects in animals. Morphine has been tested for adhesion molecule levels in blood from patients with acute myocardial infarction. Intravenous administration of 3 mg morphine after the onset of myocardial infarction reduced the serum ICAM-1 level.[90] However it is of note that there have been reports which showed that halogenated anesthetics[43] and opioids[83] were not protective when administered after the onset of ischemia. Overall, it is still controversial whether anesthetics protect the heart when administered after the onset of ischemia.

11. Conclusion

An increasing number of investigations demonstrate that halogenated anesthetics, opioids and propofol protect against myocardial ischemia-reperfusion injury. The protective effect of ketamine, barbiturates and benzodiazepines remains to be elucidated. The mechanisms by which the protection is afforded may include an ischemic preconditioning-like effect, blockade of Ca^{2+} overflow to the cytosolic space, an antioxidant-like effect, and/or interference in the neutrophil/platelet-endothelium interaction. Halogenated anesthetics and opioids seem to be potent preconditioning-inducing agents. By contrast, propofol might provide protection via different mechanisms. Comparative investigations suggested that halogenated anesthetics might be more potent cardioprotectors than *iv* anesthetics. Isoflurane and a δ-opioid agonist showed protection in human hearts.

Currently halogenated anesthetics are arguably the most promising agents as cardioprotectors among anesthetics. Their beneficial effects against ischemia-reperfusion injury have been demonstrated better than for any other anesthetic. In addition halogenated anesthetics confer protection at clinically relevant concentrations and comparative studies support their use. Although the evidence is less supportive, morphine has also been shown to be protective at clinical concentrations. Hence, halogenated anesthetics and morphine might be good choices in anesthetizing patients at risk of myocardial ischemia. This does not necessarily indicate that other opioids and propofol are not effective, but that they have not been fully examined yet. The effect of halogenated anesthetics and morphine, when they are administered after the onset of ischemia, remains questionable. Being a distinct free radical scavenger, propofol might be able to mitigate injury during and/or after ischemia.

Further studies are needed to draw clear conclusions as to which anesthetic is most useful in protecting the myocardium. It is time for interested clinicians

to evaluate the effectiveness of anesthetics in the clinical setting.

References

1 *Freedman BM, Hamm DP, Everson CT, Wechsler AS, Christian CM II.* Enflurane enhances postischemic functional recovery in the isolated rat heart. Anesthesiology 1985; 62: 29–33.

2 *Schultz JEJ, Rose E, Yao Z, Gross GJ.* Evidence for involvement of opioid receptors in ischemic preconditioning in rat hearts. Am J Physiol 1995; 268: H2157–61.

3 *Schultz JEJ, Hsu AK, Gross GJ.* Morphine mimics the cardioprotective effect of ischemic preconditioning via a glibenclamide-sensitive mechanism in the rat heart. Circ Res 1996; 78: 1100–4.

4 *Murphy PG, Myers DS, Davies MJ, Webster NR, Jones JG.* The antioxidant potential of propofol (2,6- diisopropylphenol). Br J Anaesth 1992; 68: 613–8.

5 *Kahraman S, Demiryurek AT.* Propofol is a peroxynitrite scavenger. Anesth Analg 1997; 84: 1127–9.

6 *Ko SH, Yu CW, Lee SK, et al.* Propofol attenuates ischemia-reperfusion injury in the isolated rat heart. Anesth Analg 1997; 85: 719–24.

7 *Kokita N, Hara A, Abiko Y, Arakawa J, Hashizume H, Namiki A.* Propofol improves functional and metabolic recovery in ischemic reperfused isolated rat hearts. Anesth Analg 1998; 86: 252–8.

8 *Mathur S, Farhangkhgoee P, Karmazyn M.* Cardioprotective effects of propofol and sevoflurane in ischemic and reperfused rat hearts. Role of K_{ATP} channels and interaction with the sodium-hydrogen exchange inhibitor HOE 642 (cariporide). Anesthesiology 1999; 91: 1349–60.

9 *Javadov SA, Lim KHH, Kerr PM, Suleiman MS, Angelini GD, Halestrap AP.* Protection of hearts from reperfusion injury by propofol is associated with inhibition of the mitochondrial permeability transition. Cardiovasc Res 2000; 45: 360–9.

10 *Yoo KY, Yang SY, Lee J, et al.* Intracoronary propofol attenuates myocardial but not coronary endothelial dysfunction after brief ischaemia and reperfusion in dogs. Br J Anaesth 1999; 82: 90–6.

11 *Ross S, Foex P.* Protective effects of anaesthetics in reversible and irreversible ischaemia–reperfusion injury. Br J Anaesth 1999; 82: 622–32.

12 *Dickersin K, Scherer R, Lefebvre C.* Identifying relevant studies for systematic reviews. *In*: Chalmers I, Altman DG (Eds.). Systematic Reviews. London: BMJ Publishing Group, 1995: 17–36.

13 *Opie LH.* Cell death: myocardial infarction. *In*: Opie LH (Ed.). The Heart. Physiology, from Cell to Circulation, 3rd ed. Philadelphia: Lippincott-Raven Publishers, 1998: 543–61.

14 *Opie LH.* Myocardial reperfusion: new ischemic syndromes. *In*: Opie LH (Ed.). The Heart. Physiology, from Cell to Circulation, 3rd ed. Philadelphia: Lippincott-Ravens Publishers, 1998: 563–88.

15 *Opie LH.* Oxygen lack: ischemia and angina. *In*: Opie LH (Ed.). The Heart. Physiology, from Cell to Circulation, 3rd ed. Philadelphia: Lippincott-Ravens Publishers, 1998: 515–41.

16 *Maxwell SRJ, Lip GYH.* Reperfusion injury: a review of the pathophysiology, clinical manifestations and therapeutic options. Int J Cardiol 1997; 58: 95–117.

17 *Jordan JE, Zhao ZQ, Vinten-Johansen J.* The role of neutrophils in myocardial ischemia–reperfusion injury. Cardiovasc Res 1999; 43: 860–78.

18 *Bolli R, Marban E.* Molecular and cellular mechanisms of myocardial stunning. Physiol Rev 1999; 79: 609–34.

19 *Braunwald E, Kloner RA.* The stunned myocardium: prolonged, postischemic ventricular dysfunction. Circulation 1982; 66: 1146–9.

20 *Okubo S, Xi L, Bernardo NL, Yoshida K, Kukreja RC.* Myocardial preconditioning: basic concepts and potential mechanisms. Mol Cell Biochem 1999; 196: 3–12.

21 *Nakano A, Cohen MV, Downey JM.* Ischemic preconditioning. From basic mechanisms to clinical applications. Pharmacol Ther 2000; 86: 263–75.

22 *Rubino A, Yellon DM.* Ischaemic preconditioning of the vasculature: an overlooked phenomenon for protecting the heart? Trends Pharmacol Sci 2000; 21: 225–30.

23 *Murry CE, Jennings RB, Reimer KA.* Preconditioning with ischemia: a delay of lethal cell injury in ischemic myocardium. Circulation 1986; 74: 1124–36.

24 *Tomai F, Crea F, Chiariello L, Gioffre PA.* Ischemic preconditioning in humans. Models, mediators, and clinical relevance. Circulation 1999; 100: 559–63.

25 *Lu EX, Chen SX, Hu TH, Xui LM, Yuan MD.* Preconditioning enhances myocardial protection in patients undergoing open heart surgery. Thorac Cardiovasc Surg 1998; 46: 28–32.

26 *Li G, Chen S, Lu E, Li Y.* Ischemic preconditioning improves preservation with cold blood cardioplegia in valve replacement patients. Eur J Cardiothorac Surg 1999; 15: 653–7.

27 *Szmagala P, Morawski W, Krejca M, Gburek T, Bochenek A.* Evaluation of perioperative myocardial tissue damage in ischemically preconditioned human heart during aorto coronary bypass surgery. J Cardiovasc Surg (Torino) 1998; 39: 791–5.

28 *Warltier DC, Al Wathiqui MH, Kampine JP, Schmeling WT.* Recovery of contractile function of

stunned myocardium in chronically instrumented dogs is enhanced by halothane or isoflurane. Anesthesiology 1988; 69: 552–65.

29 *Cason BA, Gamperl AK, Slocum RE, Hickey RF.* Anesthetic-induced preconditioning. Previous administration of isoflurane decreases myocardial infarct size in rabbits. Anesthesiology 1997; 87: 1182–90.

30 *Cope DK, Impastato WK, Cohen MV, Downey JM.* Volatile anesthetics protect the ischemic rabbit myocardium from infarction. Anesthesiology 1997; 86: 699–709.

31 *Kersten JR, Schmeling TJ, Pagel PS, Gross GJ, Warltier DC.* Isoflurane mimics ischemic preconditioning via activation of K_{ATP} channels. Reduction of myocardial infarct size with an acute memory phase. Anesthesiology 1997; 87: 361–70.

32 *Kowalski C, Zahler S, Becker BF, et al.* Halothane, isoflurane, and sevoflurane reduce postischemic adhesion of neutrophils in the coronary system. Anesthesiology 1997; 86: 188–95.

33 *Lochner A, Genade S, Tromp E, Theron S, Trollip G.* Postcardioplegic myocardial recovery: effects of halothane, nifedipine, HOE 694, and quinacrine. Cardiovasc Drugs Ther 1998; 12: 267–77.

34 *Preckel B, Schlack W, Comfere T, Obal D, Barthel H, Thamer V.* Effects of enflurane, isoflurane, sevoflurane and desflurane on reperfusion injury after regional myocardial ischaemia in the rabbit heart in vivo. Br J Anaesth 1998; 81: 905–12.

35 *Preckel B, Schlack W, Thamer V.* Enflurane and isoflurane, but not halothane, protect against myocardial reperfusion injury after cardioplegic arrest with HTK solution in the isolated rat heart. Anesth Analg 1998; 87: 1221–7.

36 *Schlack W, Preckel B, Stunneck D, Thamer V.* Effects of halothane, enflurane, isoflurane, sevoflurane and desflurane on myocardial reperfusion injury in the isolated rat heart. Br J Anaesth 1998; 81: 913–9.

37 *Heindl B, Reichle FM, Zahler S, Conzen PF, Becker BF.* Sevoflurane and isoflurane protect the reperfused guinea pig heart by reducing postischemic adhesion of polymorphonuclear neutrophils. Anesthesiology 1999; 91: 521–30.

38 *Ismaeil MS, Tkachenko I, Gamperl AK, Hickey RF, Cason BA.* Mechanisms of isoflurane-induced myocardial preconditioning in rabbits. Anesthesiology 1999; 90: 812–21.

39 *Piriou V, Chiari P, Knezynski S, et al.* Prevention of isoflurane-induced preconditioning by 5- hydroxydecanoate and gadolinium. Possible involvement of mitochondrial adenosine triphosphate-sensitive potassium and stretch-activated channels. Anesthesiology 2000; 93: 756–64.

40 *Shimizu J, Sakamoto A, Ogawa R.* Activation of the adenosine triphosphate sensitive mitochondrial potassium channel is involved in the cardioprotective effect of isoflurane. J Nippon Med Sch 2001; 68: 238–45.

41 *Toller WG, Kersten JR, Gross ER, Pagel PS, Warltier DC.* Isoflurane preconditions myocardium against infarction via activation of inhibitory guanine nucleotide binding proteins. Anesthesiology 2000; 92: 1400–7.

42 *Toller WG, Montgomery MW, Pagel PS, Hettrick DA, Warltier DC, Kersten JR.* Isoflurane-enhanced recovery of canine stunned myocardium. Role for protein kinase C? Anesthesiology 1999; 91: 713–22.

43 *Coetzee JF, le Roux PJ, Genade S, Lochner A.* Reduction of postischemic contractile dysfunction of the isolated rat heart by sevoflurane: comparison with halothane. Anesth Analg 2000; 90: 1089–97.

44 *Roscoe AK, Christensen JD, Lynch C III.* Isoflurane, but not halothane, induces protection of human myocardium via adenosine A_1 receptors and adenosine triphosphate–sensitive potassium channels. Anesthesiology 2000; 92: 1692–701.

45 *Heindl B, Becker BF, Zahler S, Conzen PF.* Volatile anaesthetics reduce adhesion of blood platelets under low-flow conditions in the coronary system of isolated guinea pig hearts. Acta Anaesthesiol Scand 1998; 42: 995–1003.

46 *Novalija E, Stowe DF.* Prior preconditioning by ischemia or sevoflurane improves cardiac work per oxygen use in isolated guinea pig hearts after global ischemia. Adv Exp Med Biol 1998; 454: 533–42.

47 *Heindl B, Conzen PF, Becker BF.* The volatile anesthetic sevoflurane mitigates cardiodepressive effects of platelets in reperfused hearts. Basic Res Cardiol 1999; 94: 102–11.

48 *Novalija E, Fujita S, Kampine JP, Stowe DF.* Sevoflurane mimics ischemic preconditioning effects on coronary flow and nitric oxide release in isolated hearts. Anesthesiology 1999; 91: 701–12.

49 *Preckel B, Thamer V, Schlack W.* Beneficial effects of sevoflurane and desflurane against myocardial reperfusion injury after cardioplegic arrest. Can J Anesth 1999; 46: 1076–81.

50 *Toller WG, Kersten JR, Pagel PS, Hettrick DA, Warltier DC.* Sevoflurane reduces myocardial infarct size and decreases the time threshold for ischemic preconditioning in dogs. Anesthesiology 1999; 91: 1437–46.

51 *Toller WG, Gross ER, Kersten JR, Pagel PS, Gross GJ, Warltier DC.* Sarcolemmal and mitochondrial adenosine triphosphate–dependent potassium channels. Mechanism of desflurane-induced cardioprotection. Anesthesiology 2000; 92: 1731–9.

52 *Kissin I, Stanbridge R, Bishop SP, Reves JG.* Effect of halothane on myocardial infarct size in rats. Can Anaesth Soc J 1981; 28: 239–43.

53 *Davis RF, Sidi A.* Effect of isoflurane on the extent of myocardial necrosis and on systemic hemodynamics, regional myocardial blood flow, and regional myocardial metabolism in dogs after coronary artery occlusion. Anesth Analg 1989; 69: 575–86.

54 *Davis RF, DeBoer LWV, Rude RE, Lowenstein E, Maroko PR.* The effect of halothane anesthesia on myocardial necrosis, hemodynamic performance, and regional myocardial blood flow in dogs following coronary artery occlusion. Anesthesiology 1983; 59: 402–11.

55 *Schlack W, Hollmann M, Stunneck J, Thamer V.* Effect of halothane on myocardial reoxygenation injury in the isolated rat heart. Br J Anaesth 1996; 76: 860–7.

56 *Yao L, Kato R, Foex P.* Isoflurane-induced protection against myocardial stunning is independent of adenosine 1 (A_1) receptor in isolated rat heart. Br J Anaesth 2001; 87: 258–65.

57 *Mobert J, Zahler S, Becker BF, Conzen PF.* Inhibition of neutrophil activation by volatile anesthetics decreases adhesion to cultured human endothelial cells. Anesthesiology 1999; 90: 1372–81.

58 *Kersten JR, Schmeling T, Tessmer J, Hettrick DA, Pagel PS, Warltier DC.* Sevoflurane selectively increases coronary collateral blood flow independent of K_{ATP} channels in vivo. Anesthesiology 1999; 90: 246–56.

59 *Mayfield KP, D'Alecy LG.* Role of endogenous opioid peptides in the acute adaptation to hypoxia. Brain Res 1992; 582: 226–31.

60 *Mayfield KP, D'Alecy LG.* Delta-1 opioid agonist acutely increases hypoxic tolerance. J Pharmacol Exp Ther 1994; 268: 683–8.

61 *Chien S, Oeltgen PR, Diana JN, Salley RK, Su TP.* Extension of tissue survival time in multiorgan block preparation with a delta opioid DADLE ([D-Ala^2, D-Leu^5]-enkephalin) Letter. J Thorac Cardiovasc Surg 1994; 107: 964–7.

62 *Miki T, Cohen MV, Downey JM.* Opioid receptor contributes to ischemic preconditioning through protein kinase C activation in rabbits. Mol Cell Biochem 1998; 186: 3–12.

63 *Liang BT, Gross GJ.* Direct preconditioning of cardiac myocytes via opioid receptors and K_{ATP} channels. Circ Res 1999; 84: 1396–400.

64 *Takasaki Y, Wolff RA, Chien GL, Van Winkle DM.* Met5-enkephalin protects isolated adult rabbit cardiomyocytes via δ-opioid receptors. Am J Physiol 1999; 277: H2442–50.

65 *Benedict PE, Benedict MB, Su TP, Bolling SF.* Opiate drugs and δ-receptor–mediated myocardial protection. Circulation 1999; 100(Suppl II): II-357–60.

66 *Kato R, Ross S, Foex P.* Fentanyl protects the heart against ischaemic injury via opioid receptors, adenosine A_1 receptors and K_{ATP} channel linked mechanisms in rats. Br J Anaesth 2000; 84: 204–14.

67 *Kato R, Foex P.* Fentanyl reduces infarction but not stunning via δ-opioid receptors and protein kinase C in rats. Br J Anaesth 2000; 84: 608–14.

68 *Schwartz CF, Georges AJ, Gallagher MA, Yu L, Kilgore KS, Bolling SF.* Delta opioid receptors and low temperature myocardial protection. Ann Thorac Surg 1999; 68: 2089–92.

69 *Bolling SF, Su TP, Childs KF, et al.* The use of hibernation induction triggers for cardiac transplant preservation. Transplantation 1997; 63: 326–9.

70 *Kevelaitis E, Peynet J, Mouas C, Launay JM, Menasche P.* Opening of potassium channels. The common cardioprotective link between preconditioning and natural hibernation? Circulation 1999; 99: 3079–85.

71 *Tsuchida A, Miura T, Tanno M, Nozawa Y, Kita H, Shimamoto K.* Time window for the contribution of the δ-opioid receptor to cardioprotection by ischemic preconditioning in the rat heart. Cardiovasc Drugs Ther 1998; 12: 365–73.

72 *Schultz JEJ, Hsu AK, Gross GJ.* Ischemic preconditioning is mediated by a peripheral opioid receptor mechanism in the intact rat heart. J Mol Cell Cardiol 1997; 29: 1355–62.

73 *Chien GL, Mohtadi K, Wolff RA, Van Winkle DM.* Naloxone blockade of myocardial ischemic preconditioning does not require central nervous system participation. Basic Res Cardiol 1999; 94: 136–43.

74 *Schultz JEJ, Gross GJ.* Opioids and cardioprotection. Pharmacol Ther 2001; 89: 123–37.

75 *Aitchison KA, Baxter GF, Awan MM, Smith RM, Yellon DM, Opie LH.* Opposing effects on infarction of delta and kappa opioid receptor activation in the isolated rat heart: implications for ischemic preconditioning. Bas Res Cardiol 2000; 95: 1–10; discussion 1.

76 *Bell SP, Sack MN, Patel A, Opie LH, Yellon DM.* Delta opioid receptor stimulation mimics ischemic preconditioning in human heart muscle. J Am Coll Cardiol 2000; 36: 2296–302.

77 *Tomai F, Crea F, Gaspardone A, et al.* Effects of naloxone on myocardial ischemic preconditioning in humans. J Am Coll Cardiol 1999; 33: 1863–9.

78 *Schultz JEJ, Hsu AK, Gross GJ.* Ischemic preconditioning and morphine-induced cardioprotection involve the delta (δ)-opioid receptor in the intact rat heart. J Mol Cell Cardiol 1997; 29: 2187–95.

79 *Schultz JEJ, Hsu AK, Gross GJ.* Ischemic preconditioning in the intact rat heart is mediated by δ_1- but

not μ- or κ-opioid receptors. Circulation 1998; 97: 1282–9.

80 *Jaffe JH, Martin WR*. Opioid analgesics and antagonists. *In*: Goodman Gilman A, Rall TW, Nies AS, Taylor P (Eds.). The Pharmacological Basis of Therapeutics, 8th ed. New York: Pergamon Press, 1990: 485–521.

81 *Schultz JEJ, Hsu AK, Nagase H, Gross GJ*. TAN-67, a δ_1-opioid receptor agonist, reduces infarct size via activation of $G_{i/o}$ proteins and K_{ATP} channels. Am J Physiol 1998; 274: H909–14.

82 *Fryer RM, Hsu AK, Eells JT, Nagase H, Gross GJ*. Opioid-induced second window of cardioprotection. Potential role of mitochondrial K_{ATP} channels. Circ Res 1999; 84: 846–51.

83 *McPherson BC, Yao Z*. Signal transduction of opioid-induced cardioprotection in ischemia–reperfusion. Anesthesiology 2001; 94: 1082–8.

84 *Bolling SF, Tramontini NL, Kilgore KS, Su TP, Oeltgen PR, Harlow HH*. Use of "natural" hibernation induction triggers for myocardial protection. Ann Thorac Surg 1997; 64: 623–7.

85 *Huh J, Gross GJ, Nagase H, Liang BT*. Protection of cardiac myocytes via δ_1-opioid receptors, protein kinase C, and mitochondrial K_{ATP} channels. Am J Physiol 2001; 280: H377–83.

86 *Wang GY, Wu S, Pei JM, Yu XC, Wong TM*. κ- but not δ-opioid receptors mediate effects of ischemic preconditioning on both infarct and arrhythmia in rats. Am J Physiol 2001; 280: H384–91.

87 *Fryer RM, Pratt PF, Hsu AK, Gross GJ*. Differential activation of extracellular signal regulated kinase isoforms in preconditioning and opioid-induced cardioprotection. J Pharmacol Exp Ther 2001; 296: 642–9.

88 *Wu S, Li HY, Wong TM*. Cardioprotection of preconditioning by metabolic inhibition in the rat ventricular myocyte. Involvement of κ-opioid receptor. Circ Res 1999; 84: 1388–95.

89 *Zhou JJ, Pei JM, Wang GY, et al*. Inducible HSP70 mediates delayed cardioprotection via U-50488H pretreatment in rat ventricular myocytes. Am J Physiol 2001; 281: H40–7.

90 *Wang TL, Chang H, Hung CR, Tseng YZ*. Attenuation of neutrophil and endothelial activation by intravenous morphine in patients with acute myocardial infarction. Am J Cardiol 1997; 80: 1532–5.

91 *Wang TL, Chang H, Hung CR, Tseng YZ*. Morphine preconditioning attenuates neutrophil activation in rat models of myocardial infarction. Cardiovasc Res 1998; 40: 557–63.

92 *Hofbauer R, Frass M, Gmeiner B, et al*. Effects of remifentanil on neutrophil adhesion, transmigration, and intercellular adhesion molecule expression. Acta Anaesthesiol Scand 2000; 44: 1232–7.

93 *Szekely A, Heindl B, Zahler S, Conzen PF, Becker BF*. Nonuniform behavior of intravenous anesthetics on postischemic adhesion of neutrophils in the guinea pig heart. Anesth Analg 2000; 90: 1293–300.

94 *Nakae Y, Fujita S, Namiki A*. Propofol inhibits Ca^{2+} transients but not contraction in intact beating guinea pig hearts. Anesth Analg 2000; 90: 1286–92.

95 *Buljubasic N, Marijic J, Berczi V, Supan DF, Kampine JP, Bosnjak ZJ*. Differential effects of etomidate, propofol, and midazolam on calcium and potassium channel currents in canine myocardial cells. Anesthesiology 1996; 85: 1092–9.

96 *Skoutelis A, Lianou P, Papageorgiou E, Kokkinis K, Alexopoulos K, Bassaris H*. Effects of propofol and thiopentone on polymorphonuclear leukocyte functions in vitro. Acta Anaesthesiol Scand 1994; 38: 858–62.

97 *Galley HF, Dubbels AM, Webster NR*. The effect of midazolam and propofol on interleukin-8 from human polymorphonuclear leukocytes. Anesth Analg 1998; 86: 1289–93.

98 *Mullenheim J, Frassdorf J, Preckel B, Thamer V, Schlack W*. Ketamine, but not S(+)-ketamine, blocks ischemic preconditioning in rabbit hearts in vivo. Anesthesiology 2001; 94: 630–6.

99 *Kudoh A, Matsuki A*. Ketamine inhibits inositol 1,4,5-trisphosphate production depending on the extracellular Ca^{2+} concentration in neonatal rat cardiomyocytes. Anesth Analg 1999; 89: 1417–22.

100 *Ko SH, Lee SK, Han YJ, et al*. Blockade of myocardial ATP-sensitive potassium channels by ketamine. Anesthesiology 1997; 87: 68–74.

101 *Tsutsumi Y, Oshita S, Kitahata H, Kuroda Y, Kawano T, Nakaya Y*. Blockade of adenosine triphosphate–sensitive potassium channels by thiamylal in rat ventricular myocytes. Anesthesiology 2000; 92: 1154–9.

102 *Conradie S, Coetzee A, Coetzee J*. Anesthetic modulation of myocardial ischemia and reperfusion injury in pigs: comparison between halothane and sevoflurane. Can J Anesth 1999; 46: 71–81.

103 *Haessler R, Kuzume K, Chien GL, Wolff RA, Davis RF, Van Winkle DM*. Anaesthetics alter the magnitude of infarct limitation by ischaemic preconditioning. Cardiovasc Res 1994; 28: 1574–80.

104 *Tamaki F, Oguchi T, Kashimoto S, Nonaka A, Kumazawa T*. Effects of propofol on ischemia and reperfusion in the isolated rat heart compared with thiamylal. Jpn Heart J 2001; 42: 193–206.

105 *Ross S, Munoz H, Piriou V, Ryder WA, Foex P*. A comparison of the effects of fentanyl and propofol on left ventricular contractility during myocardial stunning.

Acta Anaesthesiol Scand 1998; 42: 23–31.

106 *Meissner A, Weber TP, Van Aken H, Zbieranek K, Rolf N.* Recovery from myocardial stunning is faster with desflurane compared with propofol in chronically instrumented dogs. Anesth Analg 2000; 91: 1333–8.

107 *Belhomme D, Peynet J, Louzy M, Launay JM, Kitakaze M, Menasche P.* Evidence for preconditioning by isoflurane in coronary artery bypass graft surgery. Circulation 1999; 100(Suppl II): II-340–4.

108 *Tomai F, De Paulis R, Penta de Peppo A, et al.* Beneficial impact of isoflurane during coronary bypass surgery on troponin I release. G Ital Cardiol 1999; 29: 1007–14.

109 *Penta de Peppo A, Polisca P, Tomai F, et al.* Recovery of LV contractility in man is enhanced by preischemic administration of enflurane. Ann Thorac Surg 1999; 68: 112–8.

110 *Chauvin M, Sandouk P, Scherrmann JM, Farinotti R, Strumza P, Duvaldestin P.* Morphine pharmacokinetics in renal failure. Anesthesiology of analgesic efficacy of oxycodone and morphine in postoperative intravenous patient-controlled analgesia. Acta Anaesthesiol Scand 1998; 42: 576–80.

111 *Silvast M, Rosenberg P, Seppala T, Svartling N, Pitkanen M.* Comparison of analgesic efficacy of oxycodone and morphine in postoperative intravenous patient-controlled analgesia. Anesthesiology 1987; 66: 327–31.

112 *Hull CJ.* Pharmacokinetics and pharamacodynamics. *In*: Prys-Roberts C, Brown BRJ (Eds.). International Practice of Anaesthesia. Oxford: Butterworth-Heinemann, 1996: 1/13/1-1/17.

113 *Sprigge JS, Wynands JE, Whalley DG, et al.* Fentanyl infusion anesthesia for aortocoronary bypass surgery: plasma levels and hemodynamic response. Anesth Analg 1982; 61: 972–8.

114 *Shafer A, Doze VA, Shafer SL, White PF.* Pharmacokinetics and pharmacodynamics of propofol infusions during general anesthesia. Anesthesiology 1988; 69: 348–56.

115 *Ebel D, Schlack W, Comfere T, Preckel B, Thamer V.* Effect of propofol on reperfusion injury after regional ischaemia in the isolated rat heart. Br J Anaesth 1999; 83: 903–8.

116 *Szekely A, Heindl B, Zahler S, Conzen PF, Becker BF.* S(+)-ketamine, but not R(-)-ketamine, reduces postischemic adherence of neutrophils in the coronary system of isolated guinea pig hearts. Anesth Analg 1999; 88: 1017–24.

117 *Idvall J, Ahlgren I, Aronsen KF, Stenberg P.* Ketamine infusions: pharmacokinetics and clinical effects. Br J Anaesth 1979; 51: 1167–73.

118 *Li F, Hayes JK, Wong KC, Szakacs J.* Administration of sevoflurane and isoflurane prior to prolonged global ischemia improves heart function in isolated rat heart. Acta Anaesthesiol Sinica 2000; 38: 113–21.

119 *Komai H, Berkoff HA, Rusy BF.* Protection of ischemic rat heart by cardioplegic doses of pentobarbital. J Surg Res 1981; 30: 42–6.

120 *Ruigrok TJC, Slade AM, van der Meer P, et al.* Different effects of thiopental in severe hypoxia, total ischemia, and low-flow ischemia in rat heart muscle. Anesthesiology 1985; 63: 172–8.

121 *Preuss KC, Gross GJ, Brooks HL, Warltier DC.* Time course of recovery of "stunned" myocardium following variable periods of ischemia in conscious and anesthetized dogs. Am Heart J 1987; 114: 696–703.

122 *Sinclair DM, de Moes D, Boink AB, Ruigrok TJ.* A protective effect of thiopentone on hypoxic heart muscle. J Mol Cell Cardiol 1980; 12: 225–7.

123 *Kanaya N, Zakhary DR, Murray PA, Damron DS.* Differential effects of fentanyl and morphine on intracellular Ca^{2+} transients and contraction in rat ventricular myocytes. Anesthesiology 1998; 89: 1532–42.

124 *Ela C, Barg J, Vogel Z, Hasin Y, Eilam Y.* Distinct components of morphine effects on cardiac myocytes are mediated by the κ and δ opioid receptors. J Mol Cell Cardiol 1997; 29: 711–20.

125 *Kanesaki T, Saeki M, Ooi Y, et al.* Morphine prevents peroxynitrite-induced death of human neuroblastoma SH-SY5Y cells through a direct scavenging action. Eur J Pharmacol 1999; 372: 319–24.

126 *Kang MY, Tsuchiya M, Packer L, Manabe M.* In vitro study on antioxidant potential of various drugs used in the perioperative period. Acta Anaesthesiol Scand 1998; 42: 4–12.

127 *Weigand MA, Schmidt H, Zhao Q, Plaschke K, Martin E, Bardenheuer HJ.* Ketamine modulates the stimulated adhesion molecule expression on human neutrophils in vitro. Anesth Analg 2000; 90: 206–12.

128 *Kanaya N, Murray PA, Damron DS.* Propofol and ketamine only inhibit intracellular Ca^{2+} transients and contraction in rat ventricular myocytes at supraclinical concentrations. Anesthesiology 1998; 88: 781–91.

129 *Kunst G, Martin E, Graf BM, Hagl S, Vahl CF.* Actions of ketamine and its isomers on contractility and calcium transients in human myocardium. Anesthesiology 1999; 90: 1363–71.

130 *Nishina K, Akamatsu H, Mikawa K, et al.* The inhibitory effects of thiopental, midazolam, and ketamine on human neutrophil functions. Anesth Analg 1998; 86: 159–65.

131 *Sakai F, Hiraoka M, Amaha K.* Comparative actions of propofol and thiopentone on cell membranes of isolated guineapig ventricular myocytes. Br J Anaesth 1996; 77: 508–16.

132 *Martynyuk AE, Morey TE, Raatikainen MJP, Seubert CN, Dennis DM.* Ionic mechanisms mediating the differential effects of methohexital and thiopental on action potential duration in guinea pig and rabbit isolated ventricular myocytes. Anesthesiology 1999; 90: 156–64.

133 *Almaas R, Saugstad OD, Pleasure D, Rootwelt T.* Effect of barbiturates on hydroxyl radicals, lipid peroxidation, and hypoxic cell death in human NT2-N neurons. Anesthesiology 2000; 92: 764–74.

134 *Nonaka A, Kashimoto S, Imamura M, Furuya A, Kumazawa T.* Mechanism of the negative inotropic effect of midazolam and diazepam in cultured foetal mouse cardiac myocytes. Eur J Anaesthesiol 1997; 14: 481–7.