1. Introduction

In cardiac surgery, pre-operative as well as induced ischaemia and reperfusion almost always occur during routine surgery, during aortic clamping and declamping, occlusion of single coronary arteries during off-pump coronary artery bypass grafting, transplantation, or operations for acute ischaemic states such as evolving myocardial infarction.

Cardiac surgeons have therefore, from the very beginning, paid much attention to the ischaemic and reperfusion phase.1 As early as 1972, Cooley et al.2 reported an extreme form of ischaemic damage, the ‘stone heart’ representing an early report about the detrimental effects of uncontrolled reperfusion after ischaemia. In 1973, Hultgren et al.3 described a consecutive series of patients with open coronary arteries undergoing various open cardiac operations to develop a 7% incidence of transmural myocardial infarction. The authors stated that ‘there is clearly an urgent need to further improve the protection of the heart during surgery’. The knowledge that the ischaemic period had to be as short as possible to avoid ischaemia resulted in the paradigm that ‘as long as the aortic clamp is applied, the hearts dies’.

Several years later, methods were developed to protect the heart during the ischaemic period by lowering the metabolic demand of the heart by the induction of electromechanical arrest and profound cardiac cooling with cold crystalloid cardioplegic solutions.4,5 Later, blood-containing cardioplegic solutions were described, which allowed the heart to be supplemented with oxygen during the cross-clamp period.6

The next important step was the recognition that not ischaemia per se, but rather reperfusion with ‘normal’ blood under ‘normal’ conditions produced further damage. This was termed ‘reperfusion injury’ and generated an intensive discussion if reperfusion injury does exit or if not all the changes seen upon reperfusion are only the sequelae of the ischaemia per se.7–10 Today, there is no doubt that reperfusion injury can paradoxically reduce the beneficial effects of myocardial reperfusion.9 Uncontrolled reperfusion cause additional injury to the heart after an ischaemic insult. In contrast, there are methods available to reduce or even avoid reperfusion injury by interventions used at the beginning of myocardial reperfusion.7,8,11

Despite a great number of mostly experimental but also clinical studies in the field of ischaemia/reperfusion injury, only very few strategies have been incorporated in the routine clinical practice. To the most part, this is due to the fact that very few experimentally developed reperfusion...
strategies have been shown to be clinically effective. Of all various strategies described to reduce reperfusion injury (post- and preconditioning, single-pharmacological interventions, etc.), the concept of controlling the conditions of reperfusion and the composition of the initial reperfusate has a solid experimental background, and clinical studies have shown benefits compared with ‘uncontrolled’ reperfusion. In addition, this concept has also successfully applied to other organs. Nevertheless, the usage of this knowledge in clinical practice is still very limited and the majority of cardiac surgeons do not use any reperfusion therapy at all. Therefore, this review will describe the background, the rationale, and current results with controlled reperfusion as an intraoperative intervention to reduce the sequelae of ischaemia/reperfusion injury and give two examples of the application of this approach in other tissue (skeletal muscle and whole body) where cardiac surgeons are often involved in the treatment for acute ischaemia (acute limb ischaemia after the placement of the intra-aortic balloon pump and whole body ischaemia during pre-, intra-, or post-operative resuscitation).

2. Overview of reperfusion strategies

Myocardial ischaemia results in anaerobic glycolysis for energy production, which cannot meet the energy demands of cardiac muscle. The accumulation of end products of anaerobic glycolysis results in cellular acidosis. The loss of high-energy phosphates results in intracellular calcium influx and ultimately in myofibrillar contracture.

This makes them susceptible to further injury during reperfusion with regular blood. There have been many reports describing the detrimental effects of normal blood reperfusion which will result in cell swelling, contracture of myofibrils, disruption of the sarcolemma, and the appearance of intramitochondrial calcium phosphate particles. Several potential mediators of reperfusion injury have been described including the oxygen paradox, the calcium paradox, the pH paradox, inflammation and opening of the mitochondrial permeability transition pore (PTP).

During myocardial ischaemia, the mitochondrial PTP channel remains closed. However, it opens within the first few minutes after myocardial reperfusion in response to mitochondrial calcium overload, oxidative stress, restoration of a physiological pH, and ATP depletion. Opening of this channel results in the collapse of the mitochondrial membrane potential and uncouples oxidative phosphorylation, resulting in ATP depletion and cell death.

On the basis of these pathophysiological findings, several strategies have been proposed to reduce reperfusion injury. These strategies have been used as a single intervention or as a multitargeted approach changing the composition of the reperfusate and the condition of reperfusion. The necessity, concentration, and interaction among the different components of each modification of the composition of the reperfusate and the conditions of reperfusion have been investigated experimentally before its clinical application, and details have been published previously.

2.1 Composition of the reperfusate

Hyperkalaemia of the initial reperfusate to induce asystole is based on studies showing that the O$_2$ demands of perfused dyskinesic muscle (4–8 mL/100 g/min) are ~55% of the contracting (beating, working) segment (7–12 mL/100 g/min) and are five-fold more when the same muscle segment is arrested and decompressed by total vented bypass (0.8–1.2 mL/100 g/min). Markedly reducing the ionic calcium (Ca$^{2+}$) of the reperfusate (<250 umol/L) and adding a calcium channel-blocking drug (e.g. diltiazem) have been shown to improve myocardial salvage more than using substrate-enriched blood cardioplegic solutions alone.

Hyperosmolarity and hyperglycaemia of the reperfusate are based on studies showing that markedly increased levels of osmolarity (>400 mOsm) and glucose (>400 mg/dL) improve the capacity of substrate-enriched blood cardioplegic solution to salvage myocardium after prolonged 4 h regional ischaemia.

The oxygen-derived free radicals generated during reperfusion represent one of the fundamental processes that produce damage. It is beyond the scope of this review to provide a comprehensive overview of the generation of free radicals, with their very rapid half-life produced from multiple sources and the potential beneficial effects of free radical scavengers during the initial reperfusion. There are conflicting reports in the literature concerning the effect of the administration of scavengers at the beginning of reperfusion. In addition, free radical scavengers are currently not available for clinical use, except for clinical trials.

Chemotactic factors of cardiac subcellular origin and activated complement fragments, such as C5a, are generated locally in ischaemic myocardium. This process activates circulating neutrophils, which accumulate and play an important role in initiating and sustaining reperfusion injury. Leucocytes and, in particular, neutrophils release large amounts of oxygen-free radicals in these circumstances. Leucocyte reduction can be achieved by leucocyte filters.

2.2 Conditions of reperfusion

Duration of the reperfusion has been found to be more important than the ‘dose’ of cardioplegic reperfusion. Continuing blood cardioplegic reperfusion until myocardial oxygen uptake reaches control levels enhances regional functional recovery after acute coronary occlusion.

Studies have indicated that failure to recover systolic shortening may be due to the needless expenditure of post-ischaemic oxygen delivery necessary to meet the relatively high O$_2$ demands of muscle with systolic bulging. Minimizing O$_2$ demands by left ventricular decompression with venting during blood cardioplegic reperfusion is essential to ensure immediate functional recovery and limit histological damage.

The role of gentle reperfusion in enhancing recovery can be inferred from the results of studies of global and regional ischaemia of brief duration (i.e. 30 min) in which gradual restoration of blood flow produced less oedema and functional loss than did sudden reperfusion. Okamoto et al. showed that early temporary, gentle reperfusion limits the post-ischaemic damage that occurs with sudden, complete revascularization (aortic unclamping without the control of reperfusion pressure or flow).
2.3 Concept of controlled reperfusion

On the basis of the earlier-mentioned studies, the concept of 'controlled reperfusion' evolved including the control of the composition of the reperfusate and the control of the condition of reperfusion. This strategy was tested experimentally after up to 6 h of regional ischaemia in canine hearts showing immediate recovery of contractile function, whereas normal, uncontrolled reperfusion failed to show any recovery after 2 h of regional ischaemia. In addition, clinical studies showed the advantages of controlled reperfusion in patients with acute coronary occlusion.

3. Clinical application and results using reperfusion strategies in cardiac surgery

3.1 Routine cardiac surgery

The use of blood cardioplegia and a terminal warm reperfusion has increased the safety of cardiac operations significantly. Surgical techniques of this method for myocardial protection have been described in detail elsewhere.

3.2 Acute coronary occlusions

There has been a significant increase in risk factors in patients undergoing emergency coronary artery bypass grafting over the last years owing to the fact that these patients are only referred for surgery if all attempts at percutaneous coronary intervention have failed. Therefore, improved methods of intraoperative protection are needed for these compromised patients. Over the last decade, knowledge has been gained in the pathophysiology of acute coronary occlusion on ischaemic and non-ischaemic (remote) myocardium that has evolved in a new surgical strategy for revascularization of patients with evolving myocardial infarctions, including the non-ischaemic as well as the ischaemic myocardium.

3.3 Non-ischaemic myocardium

Studies of the natural history of acute regional ischaemia after coronary occlusion have shown that acute occlusion of a coronary artery not only affects the ischaemic myocardium, but also causes structural, functional, and metabolic alterations in the remote and adjacent myocardium. The function of the remote myocardium is the principal determinant of early survival after an otherwise non-lethal coronary occlusion (i.e. 30% of the left ventricle at risk). Survival after acute coronary occlusion is determined by the infarct size and the capacity of the remote, non-ischaemic myocardium to support the systemic circulation. Cardiogenic shock or left ventricular power failure develops if >40% of the left ventricular muscle mass acutely loses its contractile properties or if there is insufficient remote myocardium to compensate for the acute loss of <40% of contractile mass. Therefore, unimpaired blood flow to remote muscle has to be provided by revascularization in patients with multivessel disease, and active resuscitation of the remote muscle is necessary during surgical revascularization.

3.2.2 Ischaemic myocardium

Normal blood reperfusion can reverse successfully the damage imposed by a 15 min coronary occlusion, but cannot prevent massive structural, biochemical, and functional changes (not present before the onset of reflow) after 40 min of regional ischaemia. Normal blood reperfusion after longer periods of ischaemia (6 h) produces such extensive transmural necrosis that muscle salvage is unlikely. Whether this myocardial reperfusion injury occurs after normal blood reperfusion depends on the severity of ischaemia. Short ischaemic periods or high collateral blood flow during longer ischaemic periods might preserve cellular regulatory mechanisms and prevent reperfusion injury, whereas normal blood reperfusion after prolonged severe ischaemia always produces additional damage. The surgical approach to reduce or avoid this additional damage during reperfusion and thus preserve ventricular function even after prolonged periods of ischaemia is based on a treatment of ischaemic tissue during the initial reperfusion phase, before normal blood reperfusion is allowed to occur.

The surgical strategy of controlled regional reperfusion incorporates each of the principles of modification (see preceding paragraphs) of the conditions of reperfusion (total heart decompression, gentle reperfusion pressure, regional cardioplegia, normothermia, prolonged reperfusion duration) and the composition of the reperfusate [oxygenation, cardioplegia (K+), hypocalcaemia, hyperosmolarity, oxygen free radical scavengers, hyperglycaemia, leucopenia, etc.].

3.3 Heart transplantation

In heart transplantation (HTX), long global ischaemia in often pre-injured organs can be expected with potentially severe sequelae of ischaemia–reperfusion injury. Despite significant innovations and refinements in many aspects of cardiac transplantation, myocardial protection strategies have not changed much in recent decades. In the field of transplant organ protection, the main area of interest is the ideal composition of hypothermic crystalloid storage solution. Recently, a perfusion system was developed to significantly reduce the duration of ischaemia by providing constant blood flow in a beating heart during transport (Organ Care System). The superiority of this perfusion device has still to be proved even though it is an innovative solution to reduce the ischaemic time significantly.

The earlier-mentioned principles of controlled reperfusion have been applied successfully to the field of HTX. The cardiac surgeon is in a unique position to 'treat' the ischaemic injury during the initial reperfusion phase and attempt to avoid or reduce reperfusion damage by controlling the conditions of reperfusion and the composition of the reperfusate. The group from UCLA could show that reperfusion with substrate-enriched, leucocyte-depleted blood cardioplegic solutions for the first 3 min of reperfusion and leucocyte-depleted blood for additional 7 min after prolonged hypothermic ischaemia resulted in shorter duration of inotropic support, decreased leakage of myocardial enzymes, and the prevention of ultrastructural damage.

Our group has successfully applied advanced techniques of myocardial protection during explantation, storage, and initial reperfusion to the most severe form of ischaemia–reperfusion damage in the transplant setting: orthotopic transplantation of pig hearts from non-heart-beating donors after 30 min of normothermic ischaemia without
donor pre-treatment. In this experimental model, unmodified reperfusion after crystalloid cardioplegia for myocardial protection resulted in ischaemic contracture (‘stone heart’) within a few minutes after the start of reperfusion. In contrast, controlled reperfusion with leucocyte-depleted, substrate- and HOE 642-enriched blood cardiopile in the same model resulted in markedly improved contractility, and the animals were successfully weaned from extracorporeal circulation. Haemodynamic measurements 24 h after HTX revealed no significant difference between the non-heart-beating donors and a control group transplanted from beating-heart donors. Post-mortem examination showed only minimal histological damage to the myocardium.

In addition, there are data available showing that there may be a relation between suboptimal intra-operative myocardial protection and late allograft vasculopathy.

3.4 Pediatric cardiac operations

Similar strategies which have been successfully applied to reduce reperfusion injuries after an ischaemia insult can also be applied to limit the damage which occurs after reoxygenation of hypoxic tissue (reoxygenation injury).

We and others have demonstrated the existence of reoxygenation injury in the immature hypoxaemic heart. A abrupt re-introduction of molecular oxygen is followed by lipid peroxidation, enzyme release, endothelial and intra-cellular lesions, leading to membrane damage and reduced myocardial performance. Oxygen free radicals play an important role in this oxygen-related damage. In the past, cyanotic infants were placed on cardiopulmonary bypass with high pO2 levels and therefore exposed to potential reoxygenation injury. This may add to the subsequent intraoperative period of ischaemia (aortic clamping) which is necessary for cardiac repair, resulting in myocardial dysfunction. In experimental studies using immature cyanotic piglet hearts, Ihnken et al. were able to show that reduction in the bypass prime and in blood cardioplegia to normoxic levels (100 mmHg) can reduce lipid peroxidation and NO production and restore anti-oxidant reserve capacity during the initial period of re-introduction of molecular oxygen and will result in improved contractility in a pO2-dependent fashion. Interestingly, the complexity in preventing reoxygenation injury (as with reperfusion injury) has been clearly demonstrated by the findings that the deleterious effect of reoxygenation is only slightly reduced by a gradual increase in pO2 on cardiopulmonary bypass without blood cardioplegic arrest. However, it can be reduced or even prevented by controlling pO2 to normoxic (100 mmHg) or hypoxic levels (20–30 mmHg) when starting cardiopulmonary bypass and by subsequent reoxygenation with normoxic, substrate-enriched, hypocalaemic, alkalotic warm blood cardioplegic introduction (controlled reoxygenation).

In our paediatric cardiac programme, this is now the standard approach to these cyanotic infants.

4. Further application of controlled reperfusion strategies

4.1 Acute limb ischaemia

Acute limb ischaemia due to embolization is best treated by surgical interventions, such as embolectomy with a Fogarty catheter. Intraoperative thrombolysis may be added if necessary. However, other treatment options such as endovascular modalities (e.g. thrombolytic therapy, percutaneous mechanical thrombectomy, ultrasound-accelerated thrombolysis) are also available and are most often used in patients with arterial thrombosis.

Following revascularization and reperfusion of acute ischaemic limbs with normal blood, a post-reperfusion syndrome (post-ischaemic syndrome, compartment syndrome, crush syndrome, myonephropathic-metabolic syndrome) may develop and represents the most severe complication during the post-operative period. It is associated with an unexpected high morbidity and mortality rate. The causes for the high mortality rate after acute limb ischaemia are (i) high rate of co-morbidity in these mostly older patients, (ii) emergency treatment without adequate pre-operative preparation, and (iii) development of post-reperfusion syndrome.

When normal blood is allowed to reperfuse the ischaemic limb after revascularization, time is the single most important determinant confounding a successful outcome and preventing post-reperfusion syndrome.

Post-reperfusion syndrome, the result of ischaemia/reperfusion injury in skeletal muscle, is characterized by massive oedema often requiring fasciotomy, which increases fluid requirements and may cause shock. In addition, the washout of myoglobin, potassium, lactate, and micro-thrombi from the damaged skeletal muscle into the systemic circulation may cause renal failure, arrhythmias, and eventually death. The local complications of post-reperfusion syndrome may also result in amputation or severe dysfunction in the salvaged limbs.

Reperfusion with unmodified blood of skeletal muscle jeopardized by a preceding ischaemic period is the principle cause for increased morbidity and mortality after acute limb revascularization. Ischaemia itself results in the depletion of intracellular energy stores, thus making the tissue vulnerable to additional severe damage by normal blood reperfusion. However, it is of importance to note that skeletal muscle is relatively intact structurally and biochemically even after prolonged ischaemic periods. With the beginning of normal blood reperfusion, a cascade of subsequent injuries starts, including (i) activation and adhesion of leucocytes and platelets, (ii) generation of inflammatory mediators, (iii) calcium influx into cells, (iv) disruption of cellular membrane ion pumps, (v) generation of free radical oxygen, and (vi) finally cell death.

This ischaemia–reperfusion injury results in local as well as systemic complications. The local consequences are cell oedema and membrane rupture resulting in rhabdomyolysis, calcium influx into the cells, compartment syndrome, damage to the endothelial cells of the capillaries with subsequent increase in permeability, and disseminated intravascular coagulation. The systemic effects are due to a leakage of the myocyte content into the systemic circulation with subsequent acidosis, myoglobinuria, hyperkalaemia, and arrhythmias.

Experimentally, we and others have demonstrated that local and systemic complications after normal blood reperfusion following prolonged complete ischaemia can be reduced if the damaged skeletal muscle is managed carefully during the first phase (30 min) after restoration of perfusion by the strategy of controlled limb reperfusion.
Within the following years, we have developed the concept of ‘controlled limb reperfusion’ which includes the control of the conditions of reperfusion and the control of the composition of the reperfusate.

Several experimental and clinical reports indicate that sudden restoration of normal blood flow supply may cause severe oedema, rhabdomyolysis, and leakage of myocyte content into the plasma. In contrast, gradual reperfusion after ischaemia, rather than acute restoration of blood flow under systemic pressure, resulted in less severe manifestation of the reperfusion syndrome. Beneficial effects of neutrophil reduction by anti-adhesion antibodies or chemotherapeutic agents in diminishing the deleterious effect of ischaemia/reperfusion injury have also been published.

After the experimental development of the concept of controlled limb reperfusion, it was applied clinically by our group as well as by others in patients with severe, prolonged limb ischaemia. Surgical details of controlled limb reperfusion are described elsewhere. The data of these studies show superior results with controlled limb reperfusion compared with the conventional reperfusion strategy using unmodified blood as the initial reperfusate. In the future, further refinements will be added to the strategy of controlled limb reperfusion to improve the short-term (e.g. more potent free radical scavengers, angiogenesis factors) as well as long-term (e.g. stem cell therapy) outcome in this challenging patient group.

4.2 Cardiopulmonary resuscitation

After showing the beneficial effects of altering the initial reperfusion phase after a prolonged, acute ischaemic phase in many different organs (heart, skeletal muscle, kidney, lung, brain), the next step was to apply these strategies to the whole body, i.e. after cardiopulmonary resuscitation. The mortality from sudden death after cardiac arrest is 71%; most survivors sustain severe neurological damage that occurs in concert with impeded early and late recovery of other organs.

The physiological defect in sudden death is a whole-body ischaemia/reperfusion injury that follows current reperfusion methods that employ normal blood or uncontrolled reperfusion. The consequences initiate early and late organ dysfunction and lead to increased short- and long-term morbidity and mortality. An alternative to the conventional method of uncontrolled reperfusion is to vary the conditions and the compositions of whole-body reflow to employ controlled reperfusion.

The validity of our whole-body controlled reperfusion approach has been demonstrated from our recent preliminary data whereby complete neurological recovery occurs after 15 min of normothermic sudden cardiac arrest in pigs. To our knowledge, there are no previous reports of survival in swine following 15 min of untreated cardiac arrest.

Although reperfusion injury is the logical result of ischaemia followed by uncontrolled reperfusion, it is not a requirement, nor is it ‘irreversible’. There is an opportunity for intervention to prevent death if controlled reperfusion can be rapidly instituted rather than uncontrolled reperfusion. The need for controlled reperfusion mandates the use of cardiopulmonary bypass for the restoration of blood flow.

5. Conclusions

Modern reperfusion strategies have greatly influenced cardiac surgery in different areas. The ischaemic tolerance of various tissue subjected to intentional blood flow interruption during the surgical procedure can be significantly prolonged if (i) protection strategies are used that postpone ischaemic damage during this period, and (ii) reperfusion strategies are employed that reduce or avoid the injury seen after normal blood reperfusion (‘reperfusion injury’) by treating the ischaemic tissue for a period of time to enable these previously injured organs to be reperfused eventually with normal blood under ‘physiological’ conditions.

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References


