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Effect of ischaemia and reperfusion on the intracellular concentration of taurine and glutamine in the hearts of patients undergoing coronary artery surgery

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Abstract

Taurine and glutamine are the most abundant intracellular free amino acids in mammalian hearts where changes in their intracellular concentrations are likely to influence a number of cellular activities. In this study we investigated the effects of ischaemia and reperfusion on the intracellular concentrations of taurine and glutamine in the hearts of patients undergoing coronary artery bypass surgery using cold crystalloid or cold blood cardioplegic solutions. Ischaemic arrest (30 min), using cold crystalloid cardioplegic solution ($n = 19$), decreased the intracellular concentrations (μmol/g wet weight) of taurine (from 9.8 ± 0.8 to 7.7 ± 0.7, $P < 0.05$) and glutamine (8.7 ± 0.5 to 7.2 ± 0.6). After 20 min of normothermic reperfusion the fall in taurine and glutamine was maintained (7.5 ± 0.5 and 7.4 ± 0.7 for taurine and glutamine respectively). Myocardial ischaemic arrest with cold blood cardioplegic solution ($n = 16$) did not cause a significant fall in tissue taurine or glutamine. However, on reperfusion there was a marked fall in the intracellular concentrations of taurine (9.4 ± 0.5 to 6.5 ± 0.7) and glutamine (8.0 ± 0.7 to 5.8 ± 0.4). The fall in amino acids was associated with a fall in ATP and a rise in tissue lactate. This work demonstrates that irrespective of the cardioplegic solution used to arrest the heart, there is a marked fall in tissue taurine and glutamine which may influence the extent of recovery following surgery. The fall in taurine is largely due to efflux whereas changes in glutamine are due to both transport and metabolism. Ischaemia, hypothermia and changes in the transmembrane concentration gradients are the likely factors responsible for the changes in tissue amino acids.

Keywords: Taurine; Glutamine; ATP; Lactate; Troponin; Ischemia; Reperfusion; Coronary artery surgery

1. Introduction

The slowly metabolised non-protein β-amino acid taurine is present at high concentration in mammalian heart cells (ranging between 3–40 mM) but at a much lower concentration in the plasma (less than 0.1

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mM), thus creating a large concentration gradient across the sarcolemma [1,2]. This gradient is maintained by a Na^+-taurine symport using the Na^+ electrochemical gradient [1–3]. The concentration of taurine is raised in the blood of patients following acute myocardial infarction [4], unstable angina [5] and cardiac surgery [6,7]. Work on animal experimental models has provided direct evidence for a fall in taurine in heart cells during cardiac insults (e.g., Refs. [3,8–10]). A fall in tissue taurine will influence myocardial function as taurine affects cellular calcium homeostasis and taurine deficiency is associated with development of cardiomyopathy [11,12].

In contrast to taurine and until recently little was known about the role of intracellular glutamine and its transport in heart cells [13,14]. Glutamine is one of the principal free intracellular amino acids in mammalian heart cells, and it is important as a nitrogen donor for the biosynthesis of a number of compounds such as nucleotides and amino acids (see Ref. [14]). Furthermore, muscular glutamine has been shown to increase protein synthesis and decrease protein degradation and regulate glycogen metabolism [13–15]. Work on isolated Langendorff guinea-pig hearts has shown that intracellular glutamine levels are influenced by different experimental conditions and that its transport, which is Na^+-dependent and is faster than other amino acids, is largely responsible for these changes [8,14,16,17].

A fall in the intracellular concentration of both taurine and glutamine is seen in the hearts of patients undergoing coronary artery surgery during ischaemia using cold crystalloid cardioplegic solution [18]. This fall was attributed to a rise in intracellular Na^+ concentration ($[Na^+]_i$), induced by ischaemia and hypothermia, and the absence of the amino acids from the extracellular perfusate. Because taurine and glutamine have several important cellular functions, a fall in their intracellular concentration may influence the functional recovery of the heart following surgery. It is likely, however, that on reperfusion, these stores will be replenished. Alternatively, the use of cold blood cardioplegic solution (contains amino acids) to arrest the heart may prevent the fall in amino acids. We have therefore set out to study the changes in myocardial intracellular amino acids during ischaemia and on reperfusion, comparing two methods of myocardial preservation: antegrade cold crystalloid or blood cardioplegia. Preliminary data have been published elsewhere [19,20].

2. Materials and methods

Patients ($n = 35$) undergoing elective coronary artery bypass surgery were randomised to one of two accepted techniques of myocardial protection: antegrade cold (4°C) crystalloid St Thomas' I cardioplegic solution (contents in mmol/l: 16 $MgCl_2 \cdot 6H_2O$, 2 $CaCl_2$, 20 KCl, 147 NaCl, 1.0 procaine HCl) or cold (4°C) blood cardioplegic solution containing blood and St Thomas' I cardioplegic solution (4 blood: 1 crystalloid) and adjusted to give a final K^+ concentration of 20 mM. Cardioplegic solution was administered as a 1-litre bolus at the start of the ischaemic period. Infusions were repeated at 30-min intervals or earlier if electrical activity returned. The study was approved by the hospital ethics committee and patients informed consent obtained.

Preoperative and intraoperative variables for the two groups were comparable (Table 1).

2.1. Collection of biopsies

Three myocardial biopsy specimens (4–14 mg wet weight) were taken from the apex of the left ventricle

Table 1
Patients' characteristics: preoperative and intraoperative variables of patients undergoing coronary artery bypass surgery using crystalloid and blood cardioplegia

	Crystalloid cardioplegia	Blood cardioplegia
Age (years)	62.4 ± 8.3	58.6 ± 5.9
Sex (M/F)	17/2	15/1
Diabetes	2	1
Hypertension		54
Previous MI	8	10
Ejection fraction (%)	64.4 ± 9.4	62.3 ± 9.4
Unstable angina class I	3	2
Unstable angina class II	3	3
Unstable angina class III	0	1
No. of grafts	2.92 ± 0.7	3.07 ± 0.8
CPB time (min)	94.4 ± 22.2	85.1 ± 17.2
Ischaemic time (min)	44.6 ± 12.2	41.0 ± 13.7
Inotrope requirements	3	2
Perioperative MI	1	1
Death	0	0

Values are mean ± S.D. CPB, cardiopulmonary bypass; MI, myocardial infarction.

using a 'Trucut' needle. The first biopsy was taken immediately (approx. 5 min) after institution of cardiopulmonary bypass. The second biopsy was taken after 30 min of ischaemia (30 min after aortic cross-clamping) and the third approximately 20 min after reperfusion and following the removal of cross clamp. In this study there was no difference in myocardial temperature during the collection of the second biopsy (19.9 ± 1 for crystalloid vs. 19.7 ± 1°C for blood cardioplegia group). In cases where the cross-clamping period exceeded 45 min, an extra biopsy specimen was also taken. Each specimen was immediately frozen in liquid nitrogen until processing for the analysis of amino acids and other metabolites.

2.2. Determination of amino acids, ATP and lactate in biopsy specimen

In the first half of this study, 16 patients were recruited for amino acid analysis only. The procedure followed to extract free amino acids was similar to that described previously [18,21]. In brief, tissues were thawed, weighed and homogenised in ice-cold HPLC water. A sample of the homogenate was taken for protein determination and the rest transferred to an Eppendorf tube using 400 μl HPLC water and centrifuged at $400 \times g$ for 10 min. The filtrate was deproteinised using Millipore 10K molecular weight cut-off limit ultrafiltration units for 15 min at $8500 \times g$.

In the second half of this study, a further 19 patients were initially recruited for ATP measurements only. However, after a series of control experiments we were able to use the same extract for amino acids determinations. Free amino acids levels determined in the ATP extract using this procedure were similar to those measured using the one described above. Subsequently, all amino acid data were pooled together for analysis. ATP extraction was carried out as described previously [21]. In brief, frozen biopsy specimens were crushed under liquid nitrogen and the resultant powder was extracted with perchloric acid. The extracts were centrifuged at $1500 \times g$ for 10 min at 4°C. The supernatant was neutralised and used for amino acid and ATP determinations. ATP content was measured using a bioluminescent assay described elsewhere [22]. In addition to amino acids and ATP measurements, we were also able to measure lactate content in some of the extracts. Lactate was measured using a plasma lactate determination kit from Sigma Diagnostics (Sigma, Poole, UK). It must be noted, however, that lactate concentrations reported here may not be a true reflection of the actual amounts found in the tissues. This is because the kit is normally used to measure a much more concentrated samples. In addition we were only able to measure lactate in five patients from the crystalloid group.

Amino acids were determined according to the Waters Pico-Tag method [23] and similar to that reported earlier [21]. Essentially, 100 μl of the ultrafiltrate (or extract) was dried using vacuum, centrifugation and cold trap on a Savant SV160 (Farmingdale, NY, USA). Free amino acids were derivatised using phenylisothiocyanate. The phenylisothiocarbamyl derivatised amino acids were separated by HPLC using a 30-cm Pico-Tag column (Millipore Corp., Milford, MA, USA) with two Waters delivery pumps (A and B) at a constant flow of 1 ml/min with the following gradient: 100% A for 13.5min, 97% A for 10.5 min, 94% A for 6 min, 91% A for 20 min, 66% A for 12.5 min and 0% A for 4 min. The solvents used were for A: 132 mM Na acetate, 470 ml/l triethylamine (pH 6.4) and 6% acetonitrile. Solvent B was 60% acetonitrile. Derivatised amino acids were detected at 254 nm (46°C) using a Waters 486 detector. Quantitative and qualitative analysis was carried out using amino acid standards (Sigma) and the acquired data was processed using the software Millenium 2000 supplied by Waters, Millipore (UK), Watford, Herts. Chemicals needed to derivatise amino acids and separate them were also obtained from Waters Millipore.

Amino acids concentrations were expressed per wet weight. In addition and in several biopsies, the concentrations were expressed per protein concentration (data not shown) and showed a similar trend to that expressed per wet weight. Protein determination was carried out according to the Lowry method [24] using a protein determination kit from Sigma. Bovine plasma albumin (Sigma) was used as a standard.

2.3. Amino acid determination in plasma

Blood from patients undergoing open heart surgery was collected and processed before the operation. Blood plasma was deproteinised using Millipore 10K

molecular mass cut-off limit ultrafiltration units for 15 min at 8500 × *g*. The procedure for amino acid determination was the same as described above.

2.4. Troponins T and I determination

Recent years have witnessed an increased use of myocardial troponins T and I as markers of myocardial injury [25,26]. These proteins are more sensitive and specific test for myocardial cell damage. Determination of blood concentration of troponins T and I was conducted prior to surgery and at 4, 12, 24 and 48 h postoperatively. The analysis was carried out using diagnostic kits provided by Boehringer Mannheim UK (Lewes, East Sussex) and Sanofi Diagnostics Pasteur (Guilford, Surrey, UK).

2.5. Data collection and analysis

Intergroup differences were analysed by ANOVA (Fisher's PLSD) using a Statview package provided on a Macintosh PC. Correlation matrix was calculated and the significance determined using Fisher's *r* to *z*. Level of statistical significance was taken at 95%.

3. Results

3.1. Plasma and muscle amino acids concentration

Table 2 shows the resting myocardial concentration for the amino acids taurine and glutamine in the two groups of patients. The intracellular concentration of amino acids in the first biopsy, collected immediately after institution of cardiopulmonary bypass, was taken as the free resting level. Although the intracellular concentration of taurine was slightly higher than glutamine, the plasma levels of glutamine were more than 10 fold higher than taurine (Table 2). Therefore the concentration gradient expressed as the muscle-to-plasma ratio shows taurine having a ratio of approximately 300 compared to 20 for glutamine (Table 2).

Table 2
Concentration of amino acids in heart cells and in the plasma of patients undergoing coronary artery surgery using cold crystalloid ($n = 19$) and cold blood ($n = 16$) cardioplegia

Group		Taurine	Glutamine
Crystalloid	Muscle	9.76 ± 0.79	8.67 ± 0.50
	Plasma	0.043 ± 0.005	0.422 ± 0.016
	Muscle/plasma	290 ± 50	21 ± 1.5
Blood	Muscle	9.36 ± 0.53	8.04 ± 0.64
	Plasma	0.037 ± 0.006	0.444 ± 0.017
	Muscle/plasma	311 ± 40	19 ± 1.7

The concentration of amino acids (μmol/g wet weight) in ventricular biopsies collected immediately after institution of cardiopulmonary bypass is taken as resting level. Also shown is the concentration in the plasma (μmol/ml) and the amino acids gradient across the sarcolemma expressed as muscle/plasma ratio for each group.

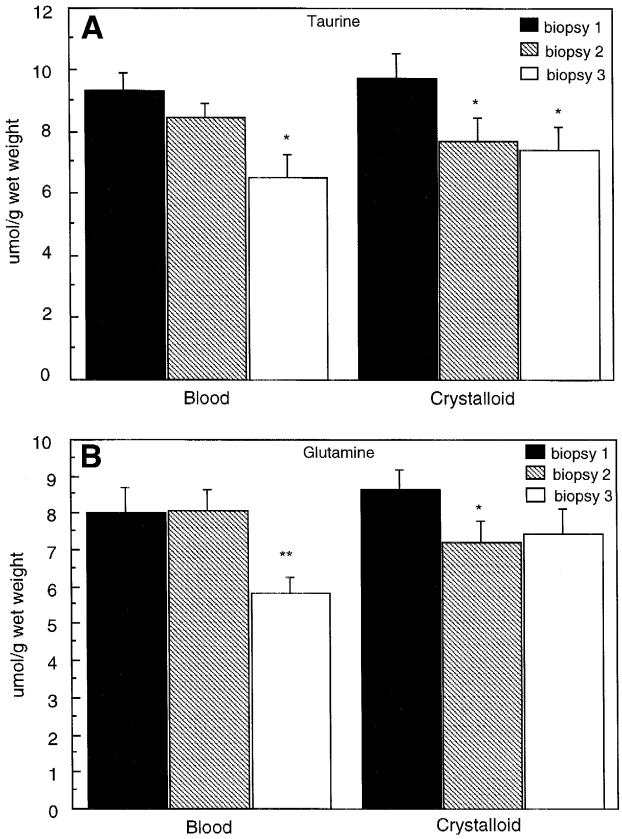

Fig. 1. The intracellular concentration of the amino acids taurine (A) and glutamine (B) in ventricular biopsies taken from patients during coronary artery bypass surgery using cold crystalloid ($n = 19$) or blood ($n = 16$) cardioplegia. Biopsy 1 was collected immediately after institution of cardiopulmonary bypass, biopsy 2 collected after 30 min of ischaemia and biopsy 3 collected 20 min after reperfusion. Values are mean ± S.E. * Significantly different from biopsy 1. ** Significantly different from biopsies 1 and 2.

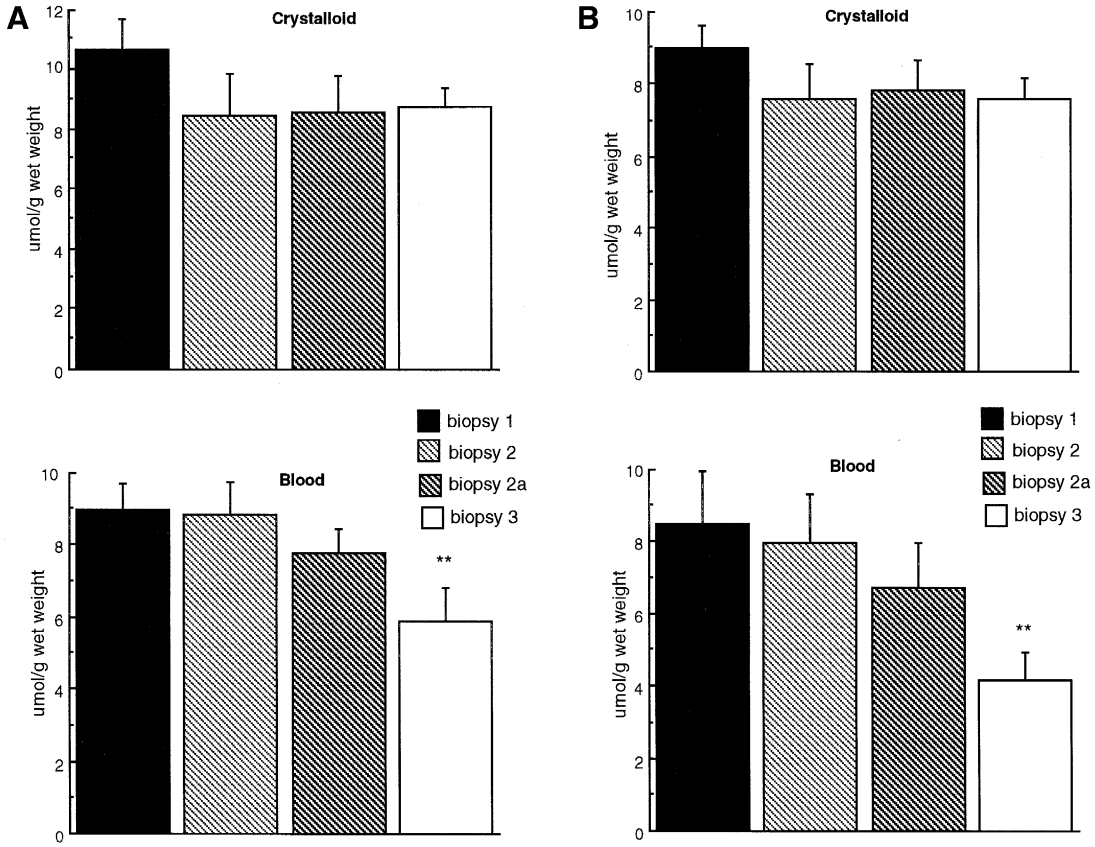

Fig. 2. The intracellular concentration of the amino acids taurine (A) and glutamine (B) in four ventricular biopsies taken from patients during coronary artery bypass surgery using cold crystalloid ($n = 6$) or blood ($n = 5$) cardioplegia when the ischaemic time exceeded 45 min. Biopsy 1 was collected immediately after institution of cardiopulmonary bypass, biopsies 2 and 2a collected after approximately 30 and 45 min of ischaemia, and biopsy 3 collected 20 min after reperfusion. Values are mean ± S.E. * * Significantly different from biopsies 1 and 2.

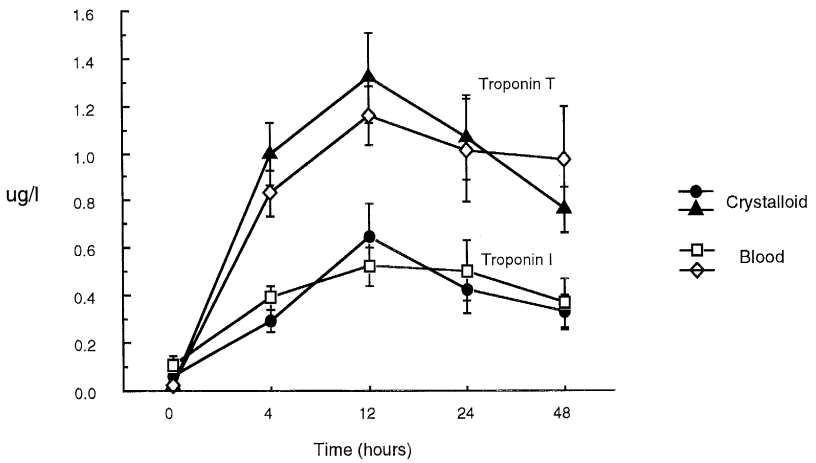

Fig. 3. Time-dependent release of troponin T and troponin I measured in the blood of patients before (0 time) and following coronary artery surgery using crystalloid or blood cardioplegia. Values are mean ± S.E. The release of troponins was similar for both crystalloid and blood cardioplegia groups.

3.2. Changes in taurine and glutamine during ischaemia and reperfusion

After 30 min ischaemia using cold crystalloid cardioplegic solution, there was a significant 21% fall in the intracellular concentration of taurine and 28% fall in glutamine (Fig. 1). After 20 min of normothermic reperfusion with blood, there was no further significant change in taurine or glutamine (Fig. 1).

In hearts arrested with cold blood cardioplegic solution, there was an insignificant 10% fall in taurine and no change in the intracellular concentration of glutamine (Fig. 1). However, on reperfusion there was a 40% fall in taurine (compared to biopsy 1) and a 54% fall in glutamine. On reperfusion, the final levels for taurine and glutamine in the crystalloid group were similar to those in the blood group (Fig. 1).

In several patients (six in the crystalloid group and five in the blood group), the ischaemic period was extended beyond 45 min and a second biopsy was collected at 45 min and analysed for amino acids. In these biopsy specimens, the changes in amino acids were not significantly different from 30-min biopsies (Fig. 2).

3.3. Changes in markers of myocardial injury

Table 3 shows that after 30 min of hypothermic ischaemia using crystalloid cardioplegic solution, there was a significant fall in ATP concentration. This fall was sustained after 20 min of normothermic reperfusion. In contrast to crystalloid cardioplegic solution, ischaemic arrest with blood cardioplegic solution resulted in a small insignificant fall in ATP. On reperfusion, however, there was a marked fall which was statistically significant when compared to the ATP levels in the preischaemic biopsies. The changes in tissue ATP were associated with changes in the opposite direction for lactate (Table 3). However, statistical significance was reached only in the blood cardioplegia group but not in the crystalloid cardioplegia group. It must be noted that we were only able to use extracts from five patients in the crystalloid cardioplegia group for lactate determination.

In this study we measured the release of myocardial troponins T and I as markers of reperfusion damage. Fig. 3 shows a time-dependent release of troponin T and troponin I before and at different periods after surgery. There was no significant difference in the release of troponins using cold crystalloid or cold blood cardioplegia.

Table 3
ATP and lactate concentrations measured in perchloric acid extracts of pulverised frozen ventricular biopsies taken from hearts arrested using cold crystalloid or cold blood cardioplegia

Metabolite	Biopsy no.	Crystalloid	Blood
ATP	1 (control)	3.0 ± 0.4	2.9 ± 0.5
	2 (ischaemia)	1.9 ± 0.3 *	2.5 ± 0.4
	3 (reperfusion)	2.0 ± 0.2	1.5 ± 0.3 *
Lactate	1 (control)	21 ± 4.0	20 ± 2.5
	2 (ischaemia)	32 ± 7.8	33 ± 6.5
	3 (reperfusion)	31 ± 7.6	39 ± 8.7 *

Biopsy 1 was collected immediately after institution of cardiopulmonary bypass, biopsy 2 collected after 30 min of ischaemia and biopsy 3 collected 20 min after reperfusion. Values are mean ± S.E. In the blood group, the same patients ($n = 10$) that were recruited for ATP measurements were also used for lactate determination. On the other hand, of the nine patients that were recruited for ATP measurements in the crystalloid group, only five were used for lactate determination. Lactate values may not necessarily reflect the exact tissue levels (see text).
* Significantly different from biopsy 1.

4. Discussion

In this work we present data showing the changes in the intracellular concentrations of the most abundant amino acids, taurine and glutamine, in the ventricles of patients undergoing coronary artery surgery. The changes were monitored after ischaemic arrest using cold crystalloid or blood cardioplegic solutions, and after 20 min of normothermic reperfusion. The changes in these amino acids and in ATP are not likely to be due to changes in the intracellular volume, as there were changes in the opposite direction for lactate, and when the concentrations of amino acids were expressed per protein content, similar trends were also seen.

4.1. Ischaemic arrest with cold cardioplegic solutions

Myocardial arrest using crystalloid cardioplegic solution but not blood cardioplegic solution, renders the heart ischaemic as evidenced by a fall in ATP and

an apparent increase in tissue lactate content (Table 3). Work on experimental models has shown that myocardial ischaemia results in a fall in adenine nucleotides accompanied with cellular acidosis. These changes will reduce the Na-pump activity and will activate the Na^+/H^+ exchanger to influx Na^+, resulting in an accumulation of $[Na^+]_i$ [27–30]. As hypothermia inhibits the Na-pump, a further rise in $[Na^+]_i$ is expected during hypothermic ischaemia [29].

Ischaemic arrest with crystalloid cardioplegic solution was associated with a fall in the intracellular concentrations of taurine and glutamine (Fig. 1). The fall in taurine is due to transport because taurine is a non-protein amino acid and is very slowly metabolised [27,31]. The transport of taurine is Na^+-dependent as shown by work on sarcolemmal vesicles, isolated heart cells and hearts [1,2]. The high sarcolemmal distribution ratio for taurine during control (preischaemic) conditions suggests an uptake requiring energy. The metabolic energy needed to influx taurine is 14.7 kJ mol^{-1} (calculated using the equation $RT\ln[AA]_i/[AA]_o$ where $R = 8.314$ J K^{-1} mol^{-1} is the gas constant, $T = 310°C$ is the absolute temperature and $[AA]_i$ and $[AA]_o$ are intracellular and extracellular concentrations respectively). The available Na^+ electrochemical energy at approximately 14.5 kJ mol^{-1} [3] is just enough for maintaining taurine gradient, assuming a stoichiometry of 1:1.The settings of the transporter can be reversed, to efflux taurine, by the absence of the extracellular amino acid, membrane depolarisation and a rise in $[Na^+]_i$. These conditions are likely to occur during arrest with cold crystalloid cardioplegic solution. This cardioplegia does not contain amino acids and the combined hypothermia and ischaemia will depolarise the membrane and induce a rise in $[Na^+]_i$.

Unlike taurine, changes in the intracellular concentration of glutamine is likely to be the net result of protein breakdown and synthesis and metabolism as well as transport. Measurements of branched-chain amino acids do not support significant changes in protein synthesis or breakdown during the duration of ischaemia and reperfusion (Suleiman et al., unpublished data). Work on rat hearts has provided evidence for the absence of glutamine synthetase (thus inability of heart cells to synthesise glutamine) and the presence of glutaminase (Rennie et al., personal communication). Ischaemic heart cells are known to utilise glutamate for energy production [32] and a fall in tissue glutamate does occur in the hearts of patients undergoing coronary artery bypass surgery ([18]; Suleiman et al., unpublished observations). It is possible therefore that the conversion of glutamine to glutamate provides the much-needed substrate for the Krebs cycle for energy production during ischaemia. In addition to metabolism, the transport of glutamine, which is both very fast and Na^+-dependent [14], may also contribute to the observed fall in the amino acid. The transmembrane distribution ratio for glutamine during control (preischaemic) conditions, suggests an uptake requiring energy. The metabolic energy needed to influx glutamine is 7.72 kJ mol^{-1}. The available Na^+ electrochemical energy at approximately 14.5 kJ mol^{-1} is more than sufficient for maintaining the glutamine gradient, assuming a stoichiometry of 1:1 for glutamine and Na^+. Like taurine, the settings of the transporter can be reversed to efflux glutamine, by the absence of the extracellular amino acid, membrane depolarisation and a rise in $[Na^+]_i$.

Unlike crystalloid cardioplegia, 30 min arrest with blood cardioplegia provokes a small but insignificant fall in tissue ATP, taurine and glutamine. The small fall in tissue taurine and glutamine was surprising, because blood contains the amino acids. The small fall in tissue ATP is likely to be due to the fact that the heart is rendered hypoxic during cold ischaemic arrest with blood. Myocardial uptake of haemoglobin-bound oxygen is greatly reduced with hypothermic blood cardioplegia [33].

4.2. Reperfusion with normothermic blood

The fall in tissue taurine and glutamine during ischaemic arrest with cold crystalloid cardioplegic solution was sustained after 20 min of normothermic reperfusion (Fig. 1). On the other hand, reperfusion following cold blood cardioplegic solution resulted in a marked fall in the amino acids (Fig. 1). The sustained fall in amino acids in the crystalloid group or the marked fall in the blood group is surprising, as the amino acids are present in the blood. As explained earlier (see Section 4.1), the changes in the intracellular concentrations of taurine and glutamine are related to transport and metabolism. It is known that early after reperfusion, heart cells are further

loaded with Na^+ (resulting from a further fall in ATP) but most importantly they are loaded with Ca^{2+} in exchange for Na^+, a process associated with cellular damage (e.g., increased energy consumption and reduced energy production (see Ref. [30]). That an increase in intracellular Na^+ has occurred is indicated by the fall in ATP and the rise in lactate (Table 3). The fall in taurine is another marker of a rise in $[Na^+]_i$, as the changes in taurine are largely due to transport (see above). On the other hand the sustained fall (or marked fall in the blood group) in glutamine on reperfusion may have resulted from both Na^+-dependent efflux and its utilisation for energy production.

A surprising observation obtained in this study is the finding that even though blood cardioplegic solution maintained the levels of ATP and amino acids, on reperfusion there was a marked fall in these substrates (Fig. 1 and Table 3). This is in contrast to the changes seen on reperfusion following arrest with crystalloid cardioplegic solution. A likely explanation for this effect is that blood cardioplegic solution contains a much reduced concentration of magnesium when compared to crystalloid cardioplegic solution (see Section 2). The availability of magnesium during the onset of reperfusion is likely to inhibit the Na/Ca exchanger (see [27]) and therefore reduce Ca^{2+} overload. This in turn will help preserve ATP as seen in the crystalloid group but not in the blood group. It can also be argued that blood cardioplegic solution contains oxygen which is a source of free radicals. Availability of oxygen may provide a substrate for generating free radicals at an early stage of reperfusion when using blood cardioplegic solution. Free radical-induced damage will result in Ca^{2+} overload and increased utilisation of ATP.

Although postischaemic metabolic derangement has been documented in animal models, little data is available showing this in humans during cardiac surgery (see [32]). In this work we provide evidence showing that early after reperfusion, there is metabolic derangement as seen by a sustained fall in the intracellular concentrations of ATP and the amino acids. The similar substrate derangement seen after ischaemia and reperfusion using crystalloid or blood cardioplegic solution was associated with similar myocardial injury as measured by the postoperative release of troponins T and I (Fig. 3).

4.3. Clinical implications

This work suggests that heart cells utilise intracellular taurine and glutamine to oppose a rise in $[Na^+]_i$ provoked by ischaemia/reperfusion (see above). This is important because a rise in $[Na^+]_i$ can lead to Ca^{2+} overload and cellular damage [28,30]. However, the loss of taurine and glutamine is likely to affect recovery following cardiac surgery. Taurine has several important roles which include membrane stabilisation, Ca^{2+} mobilisation, $[Na^+]_i$ regulation, regulation of phosphorylation of channels and transporters (see Section 1). It is also implicated in the maintenance of normal cardiac cellular function, as illustrated by the fact that its depletion is associated with the development of cardiomyopathy and that its presence affects the consequences of ischaemia, hypoxia and the calcium paradox. As for the fall in glutamine, the effects are many since glutamine is a nitrogen donor for the biosynthesis of a number of important compounds such as nucleotides and amino acids and muscular glutamine has been shown to increase protein synthesis, decrease protein degradation and regulate glycogen metabolism [14].

4.4. General conclusion

Following ischaemia and reperfusion during coronary artery bypass surgery, there is a fall in the intracellular concentrations of taurine, glutamine and ATP irrespective of the cardioplegic solution used to arrest the heart. The fall in amino acids is closely associated with metabolic stress as seen by a fall in ATP and a rise in lactate; conditions known to induce a rise in $[Na^+]_i$. This in turn is likely to activate a Na^+-dependent efflux of both taurine and glutamine. In addition, glutamine can be utilised for energy production. As these are important amino acids, strategies to replenish them must be formulated which must take into account better understanding of their transport.

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