The energy metabolism in the right and left ventricles of human donor hearts across transplantation

Serban C. Stoicaa, Duwarakan K. Satchithanandaa, Carl Atkinsona, Paul A. Whitea, Andrew N. Redingtonb, Martin Goddarda, Terence Kealeyc, Stephen R. Largea,∗

aDepartment of Transplantation, Papworth Hospital, Cambridge CB3 8RE, UK
bDepartment of Cardiology, Hospital for Sick Children, Toronto, Canada MSG 1X8
∗Department of Clinical Biochemistry, University of Cambridge, Addenbrooke’s Hospital, Cambridge CB2 2QQ, UK

Received 3 September 2002; received in revised form 29 December 2002; accepted 7 January 2003

Abstract

Objective: Brain death appears to predominantly affect the right ventricle (RV) and right ventricular failure is a common complication of clinical cardiac transplantation. It is not clear to what extent myocardial energy stores are affected in the operative sequence. We aimed to describe the time-dependent variation in high energy phosphate (HEP) metabolism of the two ventricles, and the relationship with endothelial activation and postoperative functional recovery.

Methods: Fifty-two human donors had serial biopsies from the RV and the left ventricle (LV) at (1) initial evaluation, (2) after haemodynamic optimisation, (3) end of cold ischaemia, (4) end of warm ischaemia, (5) reperfusion, and (6) at 1 week postoperatively. HEP was measured by chemiluminescence in biopsies 1–5 and adhesion molecules (P-selectin, E-selectin, VCAM-1) and thrombomodulin were analysed by immunohistochemistry in biopsies 5–6. Seventeen donors and five recipients had RV intraoperative pressure–volume recordings by a conductance catheter. Six patients served as live controls.

Results: Brain death did not affect HEP metabolism quantitatively. There was no difference between the RV and LV at any time point, but significant time-dependent changes were observed. The RV was prone to HEP depletion at retrieval, with ATP/ADP falling from 3.89 to 3.13, but recovered during cold ischaemia. During warm ischaemia the ATP/ADP ratio fell by approximately 50%, from 5.48 for the RV and 4.26 for the LV, with partial recovery at reperfusion (P < 0.005). Hearts with impaired function in the recipient showed marked variations in HEP levels at reperfusion, and those organs with RV dysfunction failed to replenish their energy stores. However, these organs were not different from normally functioning allografts in terms of endothelial activation and clinical risk factors. There was poor correlation between pressure–volume and HEP data in either donor or recipient studies. Hearts followed-up with HEP and pressure–volume studies showed improvement in the recipient, despite functioning against a higher pulmonary vascular resistance.

Conclusions: HEP are preserved over a wide range of contractile performance in the donor heart, with no metabolic difference between the two ventricles. No correlation with endothelial activation was seen either. Preservation efforts should be directed to the vulnerable periods of implantation and reperfusion.

© 2003 Published by Elsevier Science B.V.

Keywords: Heart transplantation; Brain death; Endothelium; Energy stores; Myocardial contractility

1. Introduction

Donor heart failure is the commonest cause of early mortality and an important cause of post-transplant morbidity [1] despite careful assessment of the organ prior to harvesting. While clearly multifactorial, the process of brain death significantly affects myocardial performance and experimental studies have shown that the right ventricle (RV) is affected more than the left ventricle (LV). The mechanism of this injury and its subsequent modulation by ischaemia and reperfusion are unknown, but it is clear that endothelial dysfunction of the allograft plays an important role [2,3]. Furthermore, previous studies by our group [4,5] and others [6] have shown that assessment of myocardial energy stores at the time of procurement is of value in predicting postoperative function. The relationship between high energy phosphates (HEP) and myocardial function has not been examined, however, and the longitudinal changes
that may occur throughout the early course of transplantation are unexplored. The purpose of this study was to describe the time course of HEP levels in both ventricles from the time of procurement to 1 week postoperatively and to examine coincident changes in myocardial performance and expression of surface antigens by the endothelium. The effects of brain death per se were assessed by comparing these variables in hearts from brain-dead and non-brain-dead donors.

2. Patients and methods

2.1. Donors and cardiac sampling

Fifty-two donor hearts were studied. Six were considered unsuitable using our current organ selection criteria [7] and were not subjected to longitudinal assessment. In the remaining 46 hearts, serial biopsies were obtained at the following time points:

1. on initial assessment in the donor (RV and LV),
2. before explantation (RV and LV),
3. at the end of cold ischaemic time (RV and LV),
4. at the end of warm ischaemic time (RV and LV),
5. after 10 min of reperfusion (RV and LV), and
6. at 1 week postoperatively (RV only).

The biopsies are named R1, L1, R2, L2, etc. Details of the patients are presented in Fig. 1. Seventeen donors and five corresponding recipients also had an RV assessment with the conductance catheter. Local donors were haemodynamically optimised before biopsy 2 according to previously described protocols, in which the cornerstone of management was the use of the pulmonary artery catheter and hormone resuscitation. A potential limitation of our study is that eight donor hearts were retrieved by other centres. However, they were managed according to principles of donor optimisation and acceptance was based on our clinical criteria [7]. The donor heart was arrested with 1 l of St Thomas’ 2 crystalloid solution and immersed in cold normal saline solution for transportation. Where heart–lung blocks were retrieved pneumoplegia was preceded by a prostacyclin infusion directly into the pulmonary artery over 10–15 min (details in Ref. [8]). The mean age of donors whose hearts were not used (n = 6) was 44.3 ± 5.0 years and the mean age of patients donating usable hearts (n = 46) was 37.9 ± 10.9 years. The mean ischaemic time for the transplants followed-up (n = 42) was 184.2 ± 28.5 min.

Our investigation protocol was approved by the Huntingdon Local Research Ethics Committee. The retrieval surgeon placed small purse strings on the anterior free wall of the RV and at the LV apex. Serial trucut transmural biopsies were obtained from these sites using a 16 G/9 cm Temno needle (Allegiance, IL) and the sutures were tied after the last intraoperative biopsy. The LV site was also used for venting and deairing after implantation. No complications related to this method of tissue sampling were recorded. The 1-week biopsy was obtained in standard fashion via the transjugular route from the RV endomyocardium and processed for histology only.

2.2. Recipients

Forty-three transplants were performed and 42 cases were followed-up: 29 hearts, using a biatrial cuff technique, and 13 heart–lung transplants (Fig. 1). The mean age of the heart recipients was 49.6 ± 7.9 years and the indications were idiopathic cardiomyopathy (n = 13), ischaemic cardiomyopathy (n = 12), valvulopathy (n = 3), and congenital heart disease (n = 1). The mean age of the heart–lung
2.4. Immunohistochemistry

In 22 randomly selected sets of transplanted hearts formalin-fixed paraffin-embedded sections were stained for P-selectin, E-selectin, VCAM-1 and thrombomodulin. An avidin–biotin complex technique was employed, automated with the Dako Techmate® 500 X–Y autostainer (Dako, Glostrup, Denmark). Appropriate positive controls were used and negative controls were performed by omission of the primary antibody. Sections were initially stained with haematoxylin and eosin and examined for their general histological appearance. To ensure that the endothelium is intact even in the absence of ischaemic changes all biopsies were stained with CD31, a marker of histological integrity. CD31 was also used to count all the vessels on individual sections. This assisted in evaluating P-selectin, the adhesion molecule with the highest expression in our study (for a description of the principle see Tanio et al. [12]). Sections were examined by two independent observers blinded to the patient’s identity and to the side and timing of the biopsy. For individual vessels (excluding microcapillaries) the staining for adhesion molecules was defined as present or absent. Results for adhesion molecules are reported as percentage of non-muscularised positive vessels. Thrombomodulin expression was scored on a scale of 0 (absent) to 5 (normal, uniform staining). We described the time course of endothelial activation in clinical cardiac transplantation elsewhere [13]. In this study R5 and R6 biopsies only were compared for expression of endothelial markers in recipients with and without allograft failure (Table 1).

2.5. Pressure–volume loops recording

RV functional assessment was carried out in 17 donors with stable circulation, once catecholamine support was minimised and loading optimised. (A number of donor hearts were excluded from this component of the study by lack of consent from the recipient or by organ export to another centre.) Recipient studies were carried out after separation from cardiopulmonary bypass in five selected heart transplants who were sufficiently stable for an invasive study. Recipients of heart–lung blocks were excluded due to the unknown variables introduced by the donor lungs at reperfusion. All studies were performed in the expiratory phase. After the initial evaluation, dopamine at 5 mcg/kg/min was used to test the contractile reserve.

In essence, a 7Fr conductance catheter with incorporated solid-state pressure transducer (Millar Instruments Inc., Houston, Texas) was inserted via a small sheath in the RV outflow tract. Pressure–volume loops were recorded at steady state, during parallel conductance determination, and during preload variation [14]. Preload variation was achieved by transient inferior venacaval snaring over at least five consecutive cardiac cycles. All results are averaged from at least two measurements. The conductance signal was generated and processed in a Sigma-5 DF unit (Leycom, Leiden, The Netherlands). Parallel conductance was calculated from the intersection of end-systolic and end-diastolic points following injection of 7 ml of 10%
saline at steady-state conditions. Diastole was defined by the R wave of the ECG and systole was defined by the maximum pressure/volume for each cardiac cycle. The end-systolic pressure–volume relationship (ESPVR) was produced by linear regression of consecutive systolic points in families of pressure–volume loops obtained during caval occlusion. The preload recruitable stroke work (PRSW) slope was calculated from a plot of stroke work against end-diastolic volume for these individual loops.

2.6. Statistics

Results are generally expressed as mean ± standard deviation. However, the distribution for ATP/ADP was positively skewed and for EC was negatively skewed. After logarithmic transformation of ATP/ADP and 1-EC, a normal distribution was obtained and the t-test for paired and unpaired observations was applied as appropriate. HEP results are, therefore, expressed as geometric means with 95% confidence intervals. Discrete variables were compared with Fisher’s exact test for categorical variables. VCAM-1, vascular cell adhesion molecule 1.

3. Results

3.1. Brain-dead vs. live patients

A comparison was made between 23 hearts from brain-dead donors used for transplantation and six hearts from live patients (three dominos and three controls undergoing aortic valve replacement). There was no statistically significant difference between biopsies from brain-dead and live patients (Fig. 2).

3.2. Trend over time and RV vs. LV

Only hearts used for transplantation from brain-dead donors were included in this analysis (n = 41). The values for ATP/ADP and EC are shown in Figs. 3 and 4. There were no statistically significant differences between RV and LV at any time point, but important time-dependent changes were seen in both ventricles. Energy stores were generally preserved until the end of cold ischaemia. The RV, however, seems predisposed to a drop in HEP levels during the procurement operation with subsequent replenishment during cold storage.

An important decrease in energy stores for both RV and LV was observed during warm ischaemia. Between R3 and R4, for example, the ATP/ADP fell from a mean of 5.27–2.57 (P < 0.001) and the EC fell from 0.88 to 0.82 (P < 0.001). Length of warm ischaemic time did not correlate with percentage fall in energy stores between...
time points three and four ($r = 0.14, P = 0.24$ for ATP/ADP, and $r = 0.18, P = 0.14$ for EC). Similarly, warm ischaemic time did not correlate with percentage recovery of energy stores at reperfusion (between biopsies 4 and 5) in either the RV or the LV. After 10 min of reperfusion the energy stores recovered, albeit to below the levels recorded at the end of cold ischaemia.

3.3. Allograft failure

Of 42 patients with follow-up biopsy studies extending into the reperfusion period, 11 developed donor organ failure. There were five cases of RV and six cases of global failure, with one fatal case in each sub-group. The mean value of HEP in L4 and R4 was not significantly different between sub-groups (Fig. 5). Dysfunctional organs, however, showed marked variations at reperfusion and overall failed to replenish their energy stores. Patients with RV dysfunction possibly have lower mean ATP/ADP in R5 compared with R4 ($P = 0.25$). The clinical risk factors and the putative protective factors (e.g. aprotinin administration to the recipient) were examined to ensure that they were equally distributed between patients with and without allograft failure (Table 1). An accumulation of risk factors was observed in the allograft failure group. The average ischaemic time was longer (207 vs. 176 min, $P = 0.01$) and there was a trend towards donors being still on inotropes at the time of organ retrieval. The allograft failure group also contained two patients mechanically bridged to transplantation. When the expression of endothelial adhesion molecules and thrombomodulin was compared at reperfusion and at 1 week postoperatively, the two groups were not significantly different (Table 1).

3.4. Functional correlations

There was no correlation between cardiac index, as determined by the Swan–Ganz catheter in the donor
significant between sub-groups (\( P = 0.01 \)) and there was a trend towards improved ATP/ADP and \( \Delta P/\Delta t_{\text{max}} \) (Table 3). The ATP/ADP ratio at reperfusion was inversely correlated with \( \Delta P/\Delta t_{\text{max}} \) (\( r = -0.93, P = 0.02 \)), but positively correlated with PRSW (\( r = 0.90, P = 0.03 \)).

4. Discussion

This study showed that brain death does not affect energy metabolism of the heart in quantitative terms. Furthermore, no difference between the energy stores of the two ventricles was seen across transplantation. The biggest variations in HEP levels take place during implantation and post-reperfusion. Correlation of HEP levels with contractile function or endothelial activation was poor, showing that integrated allograft function is not captured by these variables.

4.1. Brain-dead vs. live patients

Brain death is not associated with energy stores depletion in the myocardium. This is in agreement with findings of other groups in animal and in human studies [15,16]. The fact that on the left side there was a trend towards lower energy stores in the live patients may be due to the reduced number of observations in this group (\( n = 6 \)). It is also recognised that patients with ventricular hypertrophy have reduced ATP levels in the sub-endocardium [17], and there were three such patients in the live group. Although animal experiments [18,19] and clinical observations show that the RV is more vulnerable in a brain-death environment, our results suggest that a quantitative deficiency in energy metabolism is not the underlying mechanism for this phenomenon.

4.2. Overall trend

Not only the energy stores in the RV and LV are comparable at the baseline, but also they tend to evolve in parallel across transplantation. The adenine nucleotides in the RV tend, however, to diminish during organ procurement but tend to recover during cold storage. This may be due to isolated episodes of pulmonary hypertension, at the moment of brain death or afterwards. During brain death, the left atrial pressure exceeds the pulmonary pressure, temporarily halting the entire pulmonary capillary blood flow [18,20]. The RV is physiologically not equipped to withstand such a formidable increase in afterload. Perhaps this initial injury contributes to the impaired RV contractile performance reported after brain death [19] and to the initial changes in RV HEP seen in our study. The missing biopsy 1 and 2 data in 20 cases may contribute to masking a statistically significant variation (Fig. 1). Whatever the mechanism of the initial RV HEP variation, at the end of
cold storage both ventricles have normal energy stores. Strictly, from a metabolic point of view, they seem equally well prepared for the subsequent strain imposed by warm ischaemia and reperfusion.

The fall in energy stores during organ implantation is a recognised phenomenon. It has been previously reported in liver transplantation, where there seems to be an inverse correlation between recovery of energy stores and length of warm ischaemic time [21]. We were unable to show a correlation between warm ischaemic time and the variation of HEP contents during either cardiac implantation or at reperfusion. Topical cooling of the heart during implantation has been used very selectively at our institution in recent years but the current results suggest that some form of protection may have to be reconsidered. Recovery of energy stores after reperfusion requires more than 10 min. This is in agreement with the results of others, which showed that HEP levels after 30 min of reperfusion are still below levels at the end of cold ischaemia [6]. Smolenski et al. did not comment on the postoperative cardiac function in the 20 patients studied.

### 4.3. Allograft failure

Compared to recipients who had an uneventful haemodynamic course, patients who went on to develop donor organ failure did not have lower HEP levels at the end of cold ischaemia (Table 1). Also, the HEP levels at time point 2, i.e. after optimisation of the donor circulation and before harvesting, were not statistically different (results not shown). Our findings are comparable to those of Starling et al. who demonstrated no relationship between HEP levels at the end of cold ischaemia and allograft function in the postoperative period [15]. Furthermore, in our study HEP levels before and after reperfusion are insignificantly different in patients with and without subsequent allograft dysfunction (Fig. 5).

Failure to replenish the energy stores at reperfusion may be predictive of donor organ failure, but this finding did not reach statistical significance due to the marked variability in HEP values at time point 5 (Fig. 5). However, this hypothesis deserves further testing. It would be particularly helpful for the operating surgeon to have a test which is predictive of donor organ failure, such that prophylactic treatments (e.g. inhaled nitric oxide) are started as early as in the operating room. Until such an instrument is available caution should be exercised so as to avoid an accumulation of clinical risk factors in individual cases. Given the increased inflammatory load of the donor heart in general, it was interesting to see that endothelial activation was not different either between the two groups (Table 1). Biopsies 5 and 6 are, however, snapshots in what is thought to be an intricate process of endothelial modulation post-reperfusion. This area requires further investigation. Perhaps donor organ failure, whether temporary or fatal, results from a

---

### Table 2

Pearson’s correlation coefficients between right ventricular HEP values and P/V parameters in brain-dead donors (n = 17) and recipients (n = 5) at time points 1 and 5, respectively.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Brain-dead donors (n = 17)</th>
<th>Recipients (n = 5)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean (SD)</td>
<td>ATP/ADP</td>
</tr>
<tr>
<td>ATP/ADP</td>
<td>4.23 (1.38)</td>
<td></td>
</tr>
<tr>
<td>EC</td>
<td>0.85 (0.03)</td>
<td></td>
</tr>
<tr>
<td>dP/dt_max (mmHg/s)</td>
<td>310.6 (66.4)</td>
<td>0.10</td>
</tr>
<tr>
<td>dP/dt_max (mmHg/s)</td>
<td>219 (98)</td>
<td>-0.79</td>
</tr>
<tr>
<td>ESPVR</td>
<td>0.43 (0.23)</td>
<td></td>
</tr>
<tr>
<td>EDPVR</td>
<td>0.067 (0.03)</td>
<td>0.49*</td>
</tr>
<tr>
<td>PRSW (g m)</td>
<td>15.7 (4.0)</td>
<td>0.25</td>
</tr>
<tr>
<td>Ea/Ees</td>
<td>0.95 (0.41)</td>
<td></td>
</tr>
</tbody>
</table>

*The mean (SD) column shows the absolute value and the adjacent two columns show correlation coefficients. ESPVR, end-systolic pressure-volume relationship; EDPVR, end-diastolic pressure-volume relationship; PRSW, preload recruitable stroke work; Ea, effective arterial elastance; Ees, end-systolic elastance.

* The only coefficients near the range of statistical significance are *P = 0.06, †P = 0.02, and ‡P = 0.03.

### Table 3

HEP and P/V parameters between time points 1 and 5 in five donor-recipients pairs.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Brain dead donor</th>
<th>Reperfusion</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATP/ADP</td>
<td>3.65 (1.35)</td>
<td>5.28 (2.77)</td>
<td>0.08</td>
</tr>
<tr>
<td>EC</td>
<td>0.86 (0.49)</td>
<td>0.86 (0.49)</td>
<td>0.93</td>
</tr>
<tr>
<td>dP/dt_max (mmHg/s)</td>
<td>315.2 (114.5)</td>
<td>683.2 (206.7)</td>
<td>0.01</td>
</tr>
<tr>
<td>dP/dt_max (mmHg/s)</td>
<td>189.6 (77.2)</td>
<td>453.4 (179.7)</td>
<td>0.08</td>
</tr>
<tr>
<td>ESPVR</td>
<td>0.41 (0.23)</td>
<td>0.52 (0.10)</td>
<td>0.44</td>
</tr>
<tr>
<td>EDPVR</td>
<td>0.076 (0.04)</td>
<td>0.08 (0.021)</td>
<td>0.89</td>
</tr>
<tr>
<td>PRSW (g m)</td>
<td>16.3 (5.9)</td>
<td>22.2 (5.4)</td>
<td>0.21</td>
</tr>
<tr>
<td>Ea/Ees</td>
<td>1.10 (0.57)</td>
<td>1.54 (0.59)</td>
<td>0.29</td>
</tr>
<tr>
<td>PVR (dyn s/cm5)</td>
<td>96.2 (10.2)</td>
<td>259.2 (59.5)</td>
<td>0.01</td>
</tr>
</tbody>
</table>

*Expressed as mean (SD) and compared with the paired t-test. The recipient PVR (pulmonary vascular resistance) is the preoperative value obtained at right heart catheterisation.
more complex combination of intrinsic contractile dysfunction and variable loading conditions, particularly in the pulmonary bed. Finally, although marked variations in HEP levels at reperfusion may challenge the robustness of this assessment method, it is reassuring that other groups noticed the same phenomenon (see discussion in Ref. [15]).

4.4. Correlation with function

With the variables studied, a dissociation between energy metabolism and contractile function was demonstrated in the human donor heart. HEP levels in brain-dead donors did not correlate with the cardiac index measured by thermodilution or with RV load-independent parameters. The weak correlation between EDPVR and ATP/ADP in the 17 brain-dead donors is more likely to be a chance finding and did not even reach statistical significance (Table 2). Similarly, recipient correlations on a small number of patients (n = 5) are susceptible to error. Although two of the parameters reached statistical significance, it is counter-intuitive that ATP/ADP should be positively correlated with PRSW but reached statistical significance, it is counter-intuitive that ATP/ADP should be positively correlated with PRSW but inversely correlated with dP/dt. All biochemical and haemodynamic parameters improved after reperfusion, but only three of the variables were in the range of statistical significance (Table 3). This improvement took place despite implanting the heart in a circulation with a much higher pulmonary vascular resistance (259.2 vs. 96.2 dyn s/cm², P = 0.01). Since none of the five patients developed donor organ failure, perhaps removal of the heart from the brain-death environment was sufficient to ensure better function in these cases. This is a cautious conclusion based on small numbers. Some of the observed improvement may actually be due to the regular low dose of inotropic treatment used in three of the recipients at the time of recording.

Other groups analysed the metabolic state of the donor heart using 31P magnetic resonance spectroscopy [22]. A correlation was found between the energy content (expressed as creatine phosphate to ATP ratio) and the cardiac index at 1 week postoperatively, but not in the early postoperative period. (Spectroscopic methods are in addition limited in their ability to be repeated in the intraoperative period.) In a more general context, there is ongoing debate in areas of experimental and clinical cardiology on the relationship between HEP levels and contractile function [23–26]. The weight of evidence suggests that HEP levels are preserved over a wide range before systolic function is affected. In practical terms, as we have seen in this study, normal levels of adenine nucleotides in the cardiac tissue do not exclude early clinical dysfunction. In fact they tell us little about possible perturbations in ATP turnover or utilisation by the contractile apparatus. Recent studies show that HEP do not diffuse through the cytoplasm in homogenous fashion. On the contrary, the myocardial sarcoplasm is a sum of microdomains with restricted diffusion and preferential channelling of substrates to enzymes [27]. The efficiency of ATP utilisation is beyond the possibilities of current research. Taken together, these results suggest that the fate of the HEP adenine nucleotides at reperfusion remains incompletely described in transplantation, particularly in relation to clinical outcomes.

4.5. Usable vs. unusable hearts

Transplant surgeons would much appreciate a marker of intrinsic donor heart function, especially for borderline organs. In clinical studies reporting outcomes of donor hearts, a non-quantifiable variable is the amount of brain death-induced cardiac dysfunction. Earlier work from the Papworth group showed that hearts with an impaired birefringence index (a laboratory measure of biochemical and contractile function) have poor early and late clinical outcome [4]. These observations were made before introducing routine instrumentation of all donor hearts with a pulmonary flotation catheter in 1990. It is conceivable that a number of organs from our earlier reports were very borderline and/or were functioning in a sub-optimal haemodynamic milieu. Some may have been turned down by current Swan–Ganz catheter criteria [7], which could explain the magnitude of the initial birefringence observations. Subsequent work on rat myocardium submitted to warn ischaemia postulated that a low ATP/ADP ratio is the most likely explanation for a decrease in myocardial birefringence (Dr Darracott-Cankovic, unpublished observations). When the ATP/ADP index was measured in serial LV biopsies in clinical heart transplantation, a correlation with early mortality was observed, but a more detailed haemodynamic characterisation of recipients was not made in that study [5].

Our current study group contained six patients whose hearts were turned down from donation and only two of those were rejected on functional grounds. We feel that any donor hearts excluded from donation by structural disease (e.g. ventricular hypertrophy, palpable coronary artery disease etc.) should also be excluded from comparisons of function. At time point 1 the mean ATP/ADP in the two hearts unsuitable on functional grounds was lower than in used organs (3.28 vs. 4.14 in the LV, and 2.75 vs. 3.89 in the RV). With such a small number of observations it is impossible to judge how good HEP levels are at detecting unsuitable organs. The two poorly functioning hearts were also failing on conventional Swan–Ganz haemodynamic criteria and on the basis of inotropic load too. This suggests that HEP levels may become a sensitive marker of cardiac function only when the heart is failing clinically, despite resuscitative efforts. This notion is in agreement with recent experimental reports, in which the reduced afterload and coronary perfusion were shown to be critically involved in brain death-induced cardiac dysfunction [28]. From the clinicians standpoint this implies that, within the practice of donor optimisation, a finer index of intrinsic cardiac function will have to be identified. Although HEP have
been a critical endpoint in myocardial protection research, the cumulative injury sustained in transplantation and the observed variability in HEP levels make this tool appropriate for research purposes only. In summary, we have shown that the energy metabolism of the two ventricles evolves in parallel across transplantation. There appears to be no immediate relationship between energy stores, endothelial activation and contractile function. The most vulnerable periods appear to be warm ischaemia and reperfusion and this is perhaps where most preservation efforts should be directed in future studies.

Acknowledgements

Mr S.C.S. and Dr D.K.S. were supported by a grant from the Garfield Weston Research Foundation. Dr Darracott-Cankovic kindly provided us with previously unpublished observations. We are grateful to Linda Sharples and Susan Charman at the MRC Biostatistics Unit in Cambridge for performing the statistical analysis.

References


Appendix A. Conference discussion

Dr A. Murday (Glasgow, United Kingdom): Could you comment on strategies to improve that circumstance, in other words, to improve the high-energy phosphate load after reperfusion, for example, substrate-enhanced reperfusion during that time.

Dr Stoica: There is a multitude of strategies, and my view is that every
little helps. There has been, for example, a study of donor blood cardioplegia from Italy and that showed a reduction in the incidence of right ventricular failure post transplant. Another randomized study from Montreal showed that modifying reperfusion is also beneficial. This can be done with leukocyte filters, Trasylol, hotshots, substrate enhancement, as you suggested. But the number of randomized clinical experiments in this area is actually very small, so there’s a lot of work to be done.

**Dr M. Grimm (Vienna, Austria):** Did you see any effect of the cause of brain deaths on energy loads? Or maybe the patient committing suicide is different from a patient dying from a cerebrovascular event with the history of hypertension. Did you see any effect of the underlying cause of brain death?

**Dr Stoica:** We did not look at that particular issue. And I know there is some evidence to suggest that the cause of brain death has an impact on recipient outcome. We hold the view that whatever the cause of brain death, the physiological derangement can be so profound, and the subsequent injury with ischemia and reperfusion is so complex, that analyzing this variable in isolation doesn’t take us any further. And we never like to say that we rule out a brain death donor from head trauma on these grounds alone.

**Dr S. Hagl (Heidelberg, Germany):** Dr. Stoica, I think in that context it is very important to answer the question whether all these patients underwent your specific pre-explant protocol?

**Dr Stoica:** They did.

**Dr Hagl:** Because I think that’s the most important probably, that you start from a relatively, let’s say, stable level.

**Dr Stoica:** Yes.

**Dr Hagl:** And though the differences between the events prior to that might be less than in other collectives and other cohorts. Do you agree or not?

**Dr Stoica:** Well, that’s certainly possible. But this physiological study describes what happens in an average set of hearts subjected to our protocol of optimization. So it’s difficult for me to speculate on the particular cause of brain death. I don’t know the answer to that.