Improved right heart function after myocardial preservation with 2,3-butanedione 2-monoxime in a porcine model of allogenic heart transplantation

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Background: Right heart dysfunction is a major cause for early morbidity and mortality after heart transplantation. Experiments were designed to evaluate the influence of the calcium-desensitizing drug 2,3-butanedione 2-monoxime (BDM) on right heart function in a porcine model of heart transplantation.

Methods: Donor hearts of domestic pigs were arrested with BDM in Krebs solution (n = 7) and with BDM in Bretschneider’s histidine-tryptophan-ketoglutarate (HTK) solution (n = 6). There were 2 control groups: University of Wisconsin (UW, n = 6) and HTK (n = 6). An isovolumic model was used in which the right ventricular volume was precisely controlled in vivo with an intracavitary high-compliance balloon. After 4 hours of ischemia, hearts were transplanted into recipients. After 1 and 2 hours of reperfusion, the right ventricular balloon volume was increased in 10-mL increments until right ventricular failure occurred and the developed pressures were recorded.

Results: Maximal right ventricular developed pressures were significantly different after 2 hours of reperfusion (UW: 35 ± 13 mm Hg; HTK: 47 ± 8 mm Hg; Krebs+BDM: 49 ± 9 mm Hg; HTK+BDM: 50 ± 6 mm Hg; P = .04). Hearts subjected to BDM could be loaded with a significantly increased volume after 1 hour and after 2 hours (UW: 57 ± 10 mL vs HTK: 43 ± 8 mL vs Krebs+BDM: 70 ± 10 mL vs HTK+BDM: 67 ± 15 mL; P = .002). Postischemic right ventricular end-diastolic compliance was significantly increased in groups treated with BDM after 1 hour (P = .02) and after 2 hours (P = .039).

Conclusions: The drug BDM significantly improves right ventricular function in a heart transplantation model. The increase in volume load and developed right ventricular pressure achieved by BDM application would translate into a decreased risk of right ventricular failure after clinical transplantation.

Right ventricular (RV) dysfunction remains a common and potentially severe complication after heart and combined heart-lung transplantation.1,2 Ischemia-reperfusion injury, incurred during organ harvest, distant procurement, and subsequent reperfusion, is the principal underlying cause. In recent years, new drugs have experimentally shown the potential to further improve myocardial protection, which remains based on clinically well-proven crystalloid solutions. We3 previously obtained beneficial results from the application of a C1-esterase inhibitor during reperfusion in a porcine heart transplantation model. A promising substance that has been suggested as an additive to cardioplegic solutions is 2,3-butanedione 2-monoxime (BDM).4
BDM is an effective, fast-acting, and fully reversible inhibitor of cardiac and skeletal muscle contraction in man. The drug has been claimed to counteract ischemia-reperfusion injury when added to cardioplegic solutions in various isolated heart preparations in small animals with a focus on left ventricular (LV) functional recovery. Its use during initial reperfusion in a Langendorff preparation also gave improved myocardial function; however, the problem of systemic effects of the drug remains unaddressed. As a cardioplegic adjunct, no systemic effects have to be anticipated. For investigation of its additive protective properties, we used our porcine isovolumic right heart transplantation model, which allowed total control of RV function after transplantation under conditions of constant, controlled LV hemodynamics. Our study investigated whether the RV can benefit from the protective effects of BDM-enhanced cardioplegia under circum-stances mimicking the scenario of clinical heart transplantation. Therefore, we added BDM to the clinical standard heart preservation solution, Bretschneider’s HTK solution, and compared it with HTK alone as well as with an additional control group using another clinical standard heart preservation solution, University of Wisconsin (UW) solution. To test BDM independently without the various cardioprotective adjuncts included in HTK solution, we set up a group in which BDM was added to Krebs-Henseleit buffer, which is not cardioplegic itself.

Methods

Animal Preparation

In 25 Landrace pigs (27.0 ± 3.8 kg) anesthesia was induced with azaperone (5 mg/kg, given intramuscularly), atropine (0.5 mg total dose, given intramuscularly), and thiopental sodium (15 mg/kg, given intravenously). Animals were intubated and underwent mechanical ventilation with an inspired oxygen fraction of 0.5. All animals received humane care in compliance with the German animal protection legislation, the “Principles of Laboratory Animal Care,” and the “Guide for the Care and Use of Laboratory Animals” prepared by the Institute of Laboratory Animal Resources, National Research Council, and published by the National Academy Press, revised 1996.

Donor Heart Preparation

After median sternotomy the pericardium was opened and the superior and inferior venae cavae and the pulmonary artery were encircled with tapes. Heparin (300 IU/kg) was administered intravenously, and the cardioplegia cannula was secured with a purse-string suture in the ascending aorta. After a 10-minute stabilization phase, the inferior vena cava was transected, the left atrial appendage cut, and the ascending aorta crossclamped after ligation of the superior vena cava. Cardioplegic solution was then administered (1000 mL at a perfusion pressure of 40 mm Hg and a temperature of 4°C).

Four experimental groups were designed. Two control groups received either UW or Bretschneider’s HTK solution as the cardioplegic solution. One group received BDM in Krebs-Henseleit buffer (30 mmol/L), and one group received BDM in HTK solution (30 mmol/L). The solutions were of the following compositions:

- **UW solution (mmol/L):** 30 Na⁺, 125 K⁺, 5 MgSO₄, 25 phosphate, 100 lactobionate, 30 raffinose, 5 adenosine, 1 allopurinol, 3 glutathione, and 50 g/L pentastarch (ViaSpan, Belzer UW; DuPont Pharma, Bad Homburg, Germany).
- **HTK solution (mmol/L):** 15.0 NaCl, 9.0 KCl, 1.0 potassium hydrogen-2-ketoglutarate, 4.0 MgCl₂, 180.0 histidine, 2.0 tryptophan, 30.0 mannitol, and 0.015 CaCl₂ (Custodiol; Dr F. Köhler Chemie, Alsbach-Hähnlein, Germany).
- **Krebs+BDM (mmol/L):** 118.3 NaCl, 4.7 KCl, 1.2 MgSO₄, 1.2 KH₂PO₄, 25.0 NaHCO₃, 2.5 CaCl₂, and 11.1 glucose supplemented with 30 mmol/L BDM (Sigma Chemical Co, St Louis, Mo).
- **HTK+BDM:** 30 mmol/L BDM was added to HTK solution as described above.

Additionally, the hearts were topically cooled with slushed ice. After completion of cardioplegia, hearts were excised according to the standard technique and placed on slushed ice consisting of physiologic saline solution. So that RV volume could be precisely controlled in vivo, an isovolumic model was used in which RV volume was regulated with an intracavitary balloon. A high-compliance latex balloon, connected to a 2-cm–diameter polyurethane tube (Figure 1), was inserted into the RV through the transected pulmonary artery. The tricuspid valve was closed by a doubled running suture to prevent balloon herniation and thus provide absolute volume control in this model. So that the balloon could fill the entire RV cavity and conform maximally to its internal contour, the tricuspid valve chordae tendineae were cut. A 14-gauge cannula was inserted into the RV apex for drainage of thebesian venous blood. Measured amounts of saline solution were added or withdrawn through a port at the end of the tubing.
TABLE 1. Morphometric analysis

<table>
<thead>
<tr>
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<th>UW</th>
<th>HTK</th>
<th>Krebs+BDM</th>
<th>HTK+BDM</th>
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<tbody>
<tr>
<td>Donor weight (kg)</td>
<td>30.3 ± 3.3</td>
<td>24.1 ± 1.9</td>
<td>28.6 ± 4.6</td>
<td>25.6 ± 1.0</td>
</tr>
<tr>
<td>Recipient weight (kg)</td>
<td>42.3 ± 5.3</td>
<td>34.9 ± 2.7</td>
<td>38.6 ± 3.5</td>
<td>33.2 ± 2.2</td>
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<tr>
<td>Total heart weight (g)</td>
<td>119.1 ± 11.4</td>
<td>104.7 ± 11.4</td>
<td>135.7 ± 19.9</td>
<td>104.7 ± 6.4</td>
</tr>
<tr>
<td>RV free wall (g)</td>
<td>31.1 ± 3.2</td>
<td>27.9 ± 4.3</td>
<td>37.0 ± 7.9</td>
<td>29.2 ± 3.4</td>
</tr>
<tr>
<td>LV free wall (g)</td>
<td>54.4 ± 9.0</td>
<td>49.6 ± 4.7</td>
<td>67.3 ± 10.4</td>
<td>51.2 ± 4.4</td>
</tr>
<tr>
<td>Interventricular septum (g)</td>
<td>33.6 ± 10.8</td>
<td>27.2 ± 3.9</td>
<td>31.4 ± 6.2</td>
<td>24.3 ± 2.7</td>
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</table>

Mean values ± SD are given for University of Wisconsin (UW, n = 6), Bretschneider’s HTK (HTK, n = 6), BDM in Krebs solution (Krebs+BDM, n = 7), and BDM in Bretschneider’s HTK (HTK+BDM, n = 6) groups. Differences among groups were not statistically significant.

Recipient Preparation
In 25 Landrace pigs (37 ± 4.9 kg) anesthesia and ventilation were induced as described above. Mean animal weights did not differ significantly among groups (Table 1). The mean body weight of recipient animals was chosen 10 kg heavier than that of donor animals for technical ease of subsequent transplantation. Furthermore, pilot studies revealed superior hemodynamic stability in heavier recipients. Maintenance anesthesia was administered intravenously as a continuous infusion of thiopental sodium (5 mg · kg⁻¹ · h⁻¹) and fentanyl (1 µg · kg⁻¹ · h⁻¹). A carotid artery catheter was used for monitoring systemic arterial pressure. After median sternotomy and intravenous administration of a 300-IU/kg dose of heparin, total cardiopulmonary bypass was implemented via bicaval venous and bifemoral arterial cannulas. Animals were kept normothermic.

Transplantation
Total ischemic time of the hearts was 4 hours (including transplantation). After the recipient hearts were excised, donor hearts were transplanted with left atrial and aortic anastomoses created with 4-0 polypropylene running suture. After deairing, the aortic crossclamp was removed and the hearts were defibrillated and paced at a constant rate of 120 beats/min.

In this model, the RV was isolated from the circulation by complete drainage of systemic venous return and coronary sinus effluent to a pump-oxygenator. Oxygenated blood was returned to the systemic arterial circulation via the femoral arteries. The coronary arteries remained directly perfused from the ascending aorta.

Thirty minutes after the crossclamp was opened, an additional arterial line from the extracorporeal circulation circuit was inserted into the left atrium. By means of this technique, cardiac output from the left side of the heart could be controlled by pumping blood into the left atrium at a constant flow rate of 200 mL/min. Hence, a possible volume-related influence of LV function on RV performance could be avoided. Mean arterial pressure was kept constant at 60 mm Hg by adjustment of heart-lung machine flow. No drug therapy was necessary. Pump flow rates did not differ among groups.

Experimental Protocol
After transplantation, hearts were reperfused for 1 hour, and thereafter the RV balloon volume was increased by 10-mL increments until RV failure occurred. The point of RV failure was defined as a decrease in RV developed pressure, which occurred with the final administered RV balloon volume increment. RV developed pressure was defined as $P_{\text{systole}} - P_{\text{diastole}} - P_{\text{compliance}}$. The recordings were repeated after 2 hours of reperfusion. Left heart hemodynamics were kept constant under the conditions described above. The latex balloon-tubing system may influence measured RV pressure by balloon compliance ($P_{\text{compliance}}$). Therefore, the compliance of the RV balloon and tubing system was evaluated before each experiment with RV balloon volumes from 0 to 100 mL. At maximum volume, a balloon compliance of 8 mm Hg was obtained. The compliance relationship was linear over the range of balloon volumes used. The compliance test was performed for each balloon used and yielded identical curves. As previously described, the result was used to correct measured values for developed RV pressure.12 RV developed pressure and rate of RV pressure rise (RV dp/dt) were measured via a pressure transducer (Micron miniature transducer MP 15, volume displacement: 9 × 10⁻⁵ mm³/mm Hg; Micron Instruments, Simi Valley, Calif) attached to a separate port at the polyurethane tubing on the RV balloon-tubing system. The RV cavity balloon volume was defined as the total volume in the balloon-tubing system minus the volume in the tubing. RV diastolic compliance was calculated from the RV balloon volume in milliliters and the end-diastolic RV pressure in millimeters of mercury. Systemic blood pressure was measured with a fluid-filled catheter in the right common carotid artery. Experiments were terminated 2 hours after the second hemodynamic measurement by an overdose of thiopental. The transplanted hearts were removed and the RV and LV free wall and the interventricular septum were separated and weighed.

Blood samples for measurement of serum lactate and troponin-T levels, creatine kinase (CK) activity, and CK-MB concentration were drawn at the following intervals: after insertion of a carotid artery catheter and after 10, 60, and 120 minutes of reperfusion. Samples after 10, 60, and 120 minutes were collected from the coronary sinus to obtain exact measurements of myocardial marker release. Lactate and troponin-T serum levels, CK activity, and the CK-MB concentration were determined by standard laboratory tests.

Statistical Analysis
Data are expressed as mean ± standard deviation. The paired Student $t$ test was used to determine statistical significance of intra-group comparisons. One-way analysis of variance (ANOVA) was used to compare means of groups. If a significant $F$ value was obtained, a Bonferroni post hoc analysis was used to identify differences among means while controlling for multiple comparisons.
Results
RV Balloon Volume
Results of heart morphometric analyses are summarized in Table 1. As illustrated in Figure 2, A, the RV could be loaded with significantly more volume in the Krebs+BDM and HTK+BDM groups than in the HTK control group after 1 hour of reperfusion before the onset of RV failure. Although RV load in the UW control group was less than that in BDM-treated groups, this difference was not statistically significant (41.7 ± 13.3 mL [UW] vs 31.7 ± 11.7 mL [HTK] vs 55.7 ± 14.0 mL [Krebs+BDM] vs 61.7 ± 9.8 mL HTK+BDM; P = .002). The same observations could be seen after 2 hours of reperfusion (56.7 ± 10.3 mL [UW] vs 43.3 ± 8.2 mL [HTK] vs 70.0 ± 10.0 mL [Krebs+BDM] vs 66.7 ± 15.1 mL [HTK+BDM]; P = .002; Figure 2, B). After reperfusion, RV volume, measured at maximum RV developed pressure, increased in all groups (1 hour vs 2 hours of reperfusion; P < .05).

RV Developed Pressure
There was no significant difference in maximum RV developed pressure among groups after 1 hour of reperfusion (P = .063; Figure 3, A). After 2 hours, maximum RV developed pressure was significantly higher in HTK-, Krebs+BDM-, and HTK+BDM-treated groups as compared with UW-treated control animals (P = .04; Figure 3, B).

RV Diastolic Pressure-volume Relationship
RV diastolic compliance was calculated at maximum RV balloon developed pressure before the onset of RV failure. Krebs+BDM and HTK+BDM animals revealed significantly higher RV diastolic compliance than did UW and HTK controls both after 1 hour (P = .02; Figure 4, A) and after 2 hours of reperfusion (P = .039; Figure 4, B). Whereas the Krebs+BDM group and the BDM+HTK group did not differ with respect to RV diastolic compliance after 1 hour of reperfusion, after 2 hours of reperfusion...
BDM+HTK animals showed markedly, although not significantly, higher values than Krebs+BDM animals.

RV end-diastolic pressure-volume curves for RV balloon volumes from 0 to 80 mL show significantly decreased diastolic pressures in BDM-treated groups after 1 hour (P = .02; Figure 5, A) and after 2 hours of reperfusion (P < .001; Figure 5, B).

**RV dP/dt**

The RV dP/dt, obtained at maximum RV developed pressure, increased in all groups between 1 and 2 hours of reperfusion except for the HTK+BDM group, which was already showing high values after 1 hour (Table 2). Differences among groups or between 1 hour and 2 hours of reperfusion were not statistically significant.

**Blood Parameters**

Serum CK activity and CK-MB concentration increased 10- to 100-fold during the experiment. Lactate levels increased 10-fold. No statistically significant differences among groups were observed with respect to serum CK activity, CK-MB concentration, or lactate levels. Troponin-T serum levels increased more than 100-fold from before surgery to 2 hours after reperfusion. Troponin-T levels after 2 hours of reperfusion were significantly lower in both BDM-treated groups than in the UW group (P = .02, Table 2).

**Discussion**

The addition of the calcium-desensitizing agent BDM to cardioplegic solutions resulted in a significant improvement of postischemic RV function in this study. Our protocol was designed to especially address the question of RV postischemic function, which remains a critical issue in clinical heart and heart-lung transplantation.

BDM is an effective inhibitor of skeletal and cardiac muscle contraction. Several mechanisms for its action have
been suggested. A direct modulation of cross-bridge interaction on the level of the intracellular contractile apparatus has been reported by a number of investigators in rat,\textsuperscript{13,14} guinea pig,\textsuperscript{15} and human cardiac muscle.\textsuperscript{16} Within this model, BDM exerts negative inotropic effects by reducing the number of force-generating cross-bridges by increasing the cross-bridge detachment rate as well as by reducing force generation per cross-bridge.\textsuperscript{15,16} Other studies focus on the effect of BDM on sarcoplasmic reticulum Ca\textsuperscript{2+} handling.\textsuperscript{17-19} This can be described as a Ca\textsuperscript{2+}-desensitizing effect and has been observed in both cardiac and skeletal skinned muscle fibers.\textsuperscript{20} Originally, BDM was described as a nucleophilic oxime with a phosphatase-like activity\textsuperscript{21} and has also been shown to inhibit actomyosin adenosinetriphosphatase.\textsuperscript{14,22,23} Yet, the exact effect of BDM on myocardial adenosinetriphosphatase activity and energy demand still needs to be determined. A cardioprotective effect of BDM has been questioned on theoretical grounds, because BDM leads to an increase in myocardial tension cost, that is, the ratio of adenosinetriphosphatase activity versus (developed) muscle tension,\textsuperscript{16} thus increasing adenosinetriphosphatase usage per developed tension.

In contrast to these theoretical considerations, there is strong evidence from various experimental settings that adjunctive application of BDM to cardioplegic solutions is beneficial.\textsuperscript{4,10,11} Contraction uncoupling during initial postischemic reperfusion by BDM has been equally effective when applied over a 20-minute period in a dosage of 20 mmol/L.\textsuperscript{9} The drug is fast acting and its effects are fully reversible within minutes.\textsuperscript{8,9} This is at variance to slow channel calcium antagonists like nifedipine or diltiazem, which may be disadvantageous in practical clinical terms because of an overhanging cardiodepressant effect.\textsuperscript{24} In our study, the use of BDM was restricted to the ischemic period to prevent systemic side effects during reperfusion. No discussion of BDM as a cardioplegic agent in a large animal model of transplantation has been published to date.

A number of investigators previously reported on improved parameters of LV function after cardioplegia with BDM in models of rat heart ischemia and reperfusion in a Langendorff preparation.\textsuperscript{6-8} Extrapolation of results from experiments in rat hearts related to calcium metabolism, however, is probably not feasible because calcium metabolism might differ between rats and other mammals.\textsuperscript{25} Therefore, results need to be corroborated in a species comparable with human calcium metabolism, with the pig as an acceptable model. Above that, the question of RV protection and recovery has not been addressed in those studies.

The isovolumic model selected for this study enables control of the effects of confounding variables for right heart function and compensation mechanisms in right heart failure. Although it does not exactly mimic the clinical situation, the complexity of RV hemodynamics and associated determinants provided the rationale for its use. The isovolumic preparation used in this study permitted precise control of RV volume. Because left heart hemodynamics influence maximum RV developed pressure,\textsuperscript{26} observations in this study were made under conditions of controlled, constant left heart hemodynamics, with constant left heart output and systolic LV (and hence, aortic) pressure. Thus, the present model allowed optimal control of potential variables that may influence data obtained on RV function. Pulmonary vascular resistance is a very inconsistent parameter in models of orthotopic heart transplantation. This problem was avoided in the isovolumic right heart preparation used in this study. During measurements the RV is subjected to increasing balloon volumes, and thereby increasing afterload, inasmuch as the volume is not ejected. However, one

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**TABLE 2. RV dP/dt and CK-MB and troponin-T blood levels**

<table>
<thead>
<tr>
<th></th>
<th>UW</th>
<th>HTK</th>
<th>Krebs+BDM</th>
<th>HTK+BDM</th>
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</thead>
<tbody>
<tr>
<td>RV dP/dt (mm Hg/s²)</td>
<td>1: 4041 ± 1675</td>
<td>1: 4920 ± 731</td>
<td>1: 5370 ± 1581</td>
<td>1: 5440 ± 2039</td>
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<tr>
<td></td>
<td>2: 4385 ± 1382</td>
<td>2: 7464 ± 3540</td>
<td>2: 5862 ± 1288</td>
<td>2: 4789 ± 657</td>
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<tr>
<td>Plasma CK-MB concentration (µg/L)</td>
<td>1: 6.3 ± 3.4</td>
<td>1: 6.5 ± 2.3</td>
<td>1: 5.0 ± 1.3</td>
<td>1: 6.0 ± 2.7</td>
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<td></td>
<td>2: 53.4 ± 25.1</td>
<td>2: 44.3 ± 7.2</td>
<td>2: 66.1 ± 23.9</td>
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<tr>
<td></td>
<td>3: 93.1 ± 26.8</td>
<td>3: 92.1 ± 34.0</td>
<td>3: 113.3 ± 33.7</td>
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<td></td>
<td>4: 98.4 ± 25.7</td>
<td>4: 121.0 ± 47.2</td>
<td>4: 134.7 ± 49.2</td>
<td>4: 93.8 ± 25.4</td>
</tr>
<tr>
<td>Plasma troponin-T levels (µg/L)</td>
<td>1: 0.05 ± 0.09</td>
<td>1: 0.2 ± 0.4</td>
<td>1: 0.04 ± 0.09</td>
<td>1: 0.01 ± 0.0</td>
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<td></td>
<td>2: 4.3 ± 4.3*</td>
<td>2: 1.4 ± 0.7</td>
<td>2: 1.2 ± 0.6</td>
<td>2: 1.0 ± 0.3</td>
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<td></td>
<td>3: 10.3 ± 5.6*</td>
<td>3: 5.7 ± 3.0</td>
<td>3: 5.8 ± 3.5</td>
<td>3: 4.2 ± 2.1</td>
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<tr>
<td></td>
<td>4: 11.7 ± 6.1*</td>
<td>4: 6.4 ± 2.8</td>
<td>4: 5.3 ± 2.6</td>
<td>4: 5.1 ± 1.6</td>
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*Statistically significant difference between UW and BDM-treated groups (P = .049, P = .05, and P = .02, respectively, ANOVA).
limitation of the model is the condition of RV afterload during the remainder of the reperfusion period. With the balloon empty, effective RV afterload is low. Additionally, LV output was restricted to 200 mL/min and LV afterload was adjusted to 60 mm Hg. Thus, the total load for the transplanted heart was relatively low. Although this might be different from the clinical situation of early reperfusion after heart transplantation, it provided the advantage that no inotropic support was necessary.

Previous studies have shown that improved RV function does not necessarily have to be expressed as increased RV pressure. The RV may also react with increased volume load at constant pressures. In our study, there was an increase in maximum developed RV pressure after addition of BDM to HTK solution, as well as an increase in RV balloon volume at maximum RV developed pressure of up to 30 mL in BDM-treated animals as compared with UW or HTK control animals. A 10-mL increase in RV volume would mean that the RV, paced at a rate of 130 beats/min, could “pump” 1.3 L of volume more per minute after BDM protection. UW control animals showed the lowest posts ischemic maximum developed RV pressures; HTK control animals showed good developed RV pressures but the lowest posts ischemic RV volume load. The good developed RV pressures in the HTK group, however, were recorded at much lower volumes, indicating well-maintained myocardial contractility but impaired compliance of the RV. This finding is strengthened by equally high diastolic pressures in HTK and UW animals on analysis of end-diastolic RV pressure-volume relationships and by the low RV compliance at maximum volume load in both the HTK and the UW group. Both BDM-treated groups show markedly improved RV diastolic properties by decreased RV diastolic pressures and thus increased RV compliance at maximum volume load, this well preserved RV compliance being a prerequisite for effective ventricular work.

The improved protection from ischemia-reperfusion injury in BDM animals was confirmed by a significantly lower release of troponin-T in BDM-treated groups after 2 hours of reperfusion. In our experimental model, CK-MB serum concentrations increased continuously with time in all groups, and after reperfusion HTK+BDM animals showed the lowest concentrations. CK activity did not differ significantly among groups, with a possible explanation being the ligation of the femoral arteries for cannulation, causing an increased lactate and CK release from the pigs’ lower extremities.

Although differences in the functional recovery of hearts and in the release of markers of tissue injury were not statistically significant between BDM in Krebs solution and BDM in HTK, BDM in HTK seems to provide superior preservation, as is shown by slightly better posts ischemic function and lower tissue injury marker release. However, the good functional recovery of hearts preserved with BDM in Krebs solution as compared with BDM in HTK was not expected and provides additional evidence for the excellent preservation properties of that agent.

The calcium concentration of a heart preservation solution has been reported to have an important influence on posts ischemic myocardial function. All preservation solutions used in this study have varying calcium concentrations and, above that, BDM is known to chelate free ionized calcium in a dose-dependent manner. At 30 mmol/L, BDM reduces ionized free calcium by 50%. Robinson and Harwood reported that reducing the calcium concentration in St Thomas’ Hospital solution from 1.2 to 0.6 mmol/L results in a significant improvement of recovery of posts ischemic function in rat hearts. Comparing the solutions used in our study, UW solution contains no calcium at all and the calcium concentration of HTK (0.015 mmol/L) is low. Although one could speculate that these low calcium concentrations might have caused part of the difference in RV function as compared with the BDM groups, this is unlikely considering the fact that the addition of BDM to HTK, which resulted in a significant improvement of RV function, should even lower its calcium concentration. On the other hand, it is not possible to conclude that relatively high calcium concentrations per se are deleterious, because BDM also provided significantly improved RV protection on addition to Krebs-Henseleit solution with a calcium concentration of 2.5 mmol/L (that should have been reduced by chelating to 1.25 mmol/L by BDM). However, the exact contribution of calcium concentration to RV protection in the solutions investigated in this study still needs to be determined.

In conclusion, our study demonstrates that RV function after transplantation is significantly improved after addition of BDM to HTK solution. Using an isovolumic right heart transplant preparation with total control of LV function, maximum volume load of the RV before the onset of RV failure was significantly higher and end-diastolic RV pressure was significantly lower after BDM application than in controls with HTK alone. Thus, administration of BDM-supplemented cardioplegic solution may protect the myocardium from ischemic injury after prolonged ischemia during transplantation.

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References


