Effects of Triiodothyronine Supplementation After Myocardial Ischemia

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Cardiopulmonary bypass causes a “euthyroid-sick” state characterized by low levels of circulating triiodothyronine. Triiodothyronine supplementation in this setting has been postulated to improve postischemic left ventricular function by increasing the availability of myocardial high-energy phosphates. These postulates have not been substantiated, however, using load-independent parameters of left ventricular function and analysis of high-energy phosphate metabolism. To test these hypotheses, 14 healthy pigs (30 to 40 kg) were placed on cardiopulmonary bypass and instrumented with left ventricular minor-axis ultrasonic crystals and micromanometer-tipped pressure catheters. Hearts were subjected to 30 minutes of global, normothermic ischemia. Triiodothyronine (0.1 mg/kg; n = 7) or placebo (n = 7) was administered in a random, investigator-blinded fashion at the removal of the aortic cross-clamp and after 60 minutes of reperfusion. Hemodynamic, metabolic, and ultrastructural data were obtained before ischemia and after 30, 60, 90, and 120 minutes of reperfusion. By 90 minutes of reperfusion left ventricular contractility had returned to preischemic levels in hearts supplemented with triiodothyronine, despite postischemic myocardial adenosine triphosphate levels of 50% to 60% of baseline in both groups. Ultrastructurally, the sarcoplasmic reticulum and mitochondria were significantly better preserved in the group treated with triiodothyronine. This study suggests that triiodothyronine supplementation significantly enhances postischemic left ventricular functional recovery and that this recovery is due to mechanisms other than enhanced availability of myocardial high-energy phosphates.


Despite an operative mortality of less than 2% for elective coronary artery bypass grafting, postoperative left ventricular dysfunction remains problematic [1]. Transient decreases in left ventricular function in the immediate postoperative period have been reported in patients with normal preoperative ventricular ejection fractions [2-6], as well as in patients with impaired ventricular function. The cause of this transient, postoperative left ventricular mechanical derangement is usually attributed to either inadequate intraoperative myocardial protection or reperfusion injury. Alternatively, however, it has been suggested that alterations in thyroid hormone metabolism may contribute to cardiac dysfunction after open heart operations.

Cardiopulmonary bypass leads to a “euthyroid-sick” state, defined by depressed levels of free, unbound triiodothyronine (T₃) and elevated levels of reverse T₃, with normal or slightly elevated circulating free thyroxine levels [7-12]. Novitzky and associates [13-16] have suggested that this transient depression in free T₃ levels after cardiopulmonary bypass contributes to postoperative myocardial dysfunction and that T₃ supplementation in this setting improves ventricular systolic performance in the immediate postoperative period. However, studies describing the effects of acute T₃ supplementation on cardiac function have been limited by the use of highly load-dependent indices of contractility, making assessment of ventricular function difficult. Using an isolated rabbit heart model, we have previously demonstrated that T₃ has no intrinsic inotropic properties when given in either physiologic or pharmacologic doses to normal hearts [17]. However, T₃ supplementation significantly enhanced left ventricular functional recovery after ischemic injury [17].

The mechanisms by which acute T₃ supplementation enhances postischemic function are not clear. Novitsky and associates [15] suggested that T₃ supplementation increases the aerobic oxidative capacity of the myocardium, thereby leading to an increased availability of high-energy phosphates for contraction. The purpose of the present study was to evaluate left ventricular functional recovery after ischemia in an in vivo, large animal model using a load-independent index of left ventricular function. Additionally, we wished to test the hypothesis that T₃ supplementation enhances postischemic ventricular function by increasing the availability of myocardial high-energy phosphates available for contraction.


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Material and Methods

Instrumentation
Fourteen healthy pigs weighing between 30 and 40 kg were studied. All animals received humane care in compliance with the “Principles of Laboratory Animal Care” formulated by the National Society for Medical Research and the “Guide for the Care and Use of Laboratory Animals” prepared by the National Academy of Sciences and published by the National Institutes of Health (NIH publication 85-23, revised 1985).

Before instrumentation, pigs were sedated with an intramuscular injection of atropine sulfate (400 µg), acepromazine maleate (50 mg), and ketamine HCl (500 mg), and then anesthetized with pentobarbital (13 mg/kg intravenously). Subsequent doses of pentobarbital were administered every 1 to 2 hours as needed. Tracheostomy was performed to secure an airway, and animals were ventilated with a large animal respirator. The external jugular vein was isolated and cannulated for administration of intravenous fluids. Lidocaine HCl (1 mg/kg intravenously) and bretylum tosylate (8 mg/kg intravenously) were administered before median sternotomy for prophylaxis against ventricular arrhythmias. The left femoral artery was isolated, and a micromanometer-tipped catheter (Millar Instruments, Houston, TX) was introduced and positioned in the thoracic aorta for continuous monitoring of systemic blood pressure.

After median sternotomy, the right and left hemiazygos veins were ligated and the heart was suspended in a pericardial cradle. The right and left phrenic nerves were sectioned to eliminate diaphragmatic contraction. Left ventricular pressure was monitored with an intracavitary piezoelectric crystals was positioned across the left ventricular anteroposterior minor axis to record minor-axis velocity and was used to quantify ventricular performance [MI.

Assessment of Ventricular Performance
Hemodynamic and dimensional data were acquired online from the pressure catheters and ultrasonic crystals at 200 Hz on a Zenith 386 personal computer with interactive software developed in our laboratory. Analog data were digitized and passed through a recursive, low-pass Butterworth filter with a cut-off frequency of 30 Hz. The derivative of left ventricular pressure with respect to time (dP/dt) was calculated and the cardiac cycle defined such that maximally negative dP/dt defined end-systole. Left ventricular pressure–dimension work loops were produced over a physiologic range of end-diastolic volumes by emptying venous blood into the cardiopulmonary reservoir. The slope of the linear relationship between left ventricular stroke work and end-diastolic length has been demonstrated to be a load-insensitive index of contractility and was used to quantify ventricular performance [18].

Assessment of Myocardial Adenine Nucleotide Pool
Transmural myocardial biopsy specimens were obtained with a Tru-Cut needle (Travenol Laboratories, Deerfield, IL) before ischemia, at the end of ischemia, and after 30, 60, 90, and 120 minutes of reperfusion. Biopsy specimens were obtained from the anterior and posterior left ventricular walls randomly, with care to avoid injury to the coronary vasculature. Biopsy specimens were blotted and immediately frozen (within 3 seconds) in liquid nitrogen for storage until final analysis. Frozen tissue was homogenized in 0.4 mL 12% trichloroacetic acid for 30 minutes and centrifuged at 8,000 rpm for 10 minutes to separate denatured protein. The supernatant contained the soluble acid extract, from which adenine nucleotides and metabolites were measured using high-pressure liquid chromatography (model M-490; Waters Associates, Milford, MA) [19]. Protein content of the sediment was determined as previously described [20]. Final values of adenine nucleotides are expressed as nanomoles per milligram of protein.

Electron Microscopy
Myocardial ultrastructure was evaluated in five hearts from each group. Left ventricular transmural myocardial biopsy specimens were obtained before ischemia and after 120 minutes of reperfusion using a Tru-Cut needle. Biopsy specimens were immediately placed in fresh 3% glutaraldehyde, 1.5% paraformaldehyde in 0.1 mol/L sodium cacodylate buffer (pH 7.4) and stored until processing for transmission electron microscopy.

Before fixation, biopsy specimens were rinsed three
times in 0.1 mol/L sodium cacodylate buffer, pH 7.4 with 4% sucrose added, and postfixed for 2 hours with 2% osmium tetroxide, 0.8% potassium ferricyanide, in 0.1 mol/L sodium cacodylate on ice. Samples were rinsed with water and en bloc stained with saturated aqueous uranyl acetate for 2 hours at 60°C, followed by dehydration in a graded series of ethanols, then propylene oxide. After embedding in Spurr resin, silver sections were cut with a 0 (indicating no damage) to 3 (complete disruption) grading system for the nucleus, myofibrils, sarcolemma, mitochondria, and interstitium (Table 1). Samples were coded, prepared, and graded in a blinded fashion; the code was broken and the results were tabulated only after completion of ultrastructural grading.

Statistical Analysis

All statistical analysis was performed using personal computer-based software from the SAS Institute (Cary, NC). Data obtained at multiple time points were compared using a one-way analysis of variance. The difference in ability to be weaned from cardiopulmonary bypass between groups was assessed using $\chi^2$ analysis with Fisher’s exact test. Electron microscopic ultrastructural scores between the two groups were compared using an unpaired $t$ test. Significance was accepted at the $p$ less than 0.05 level. Unless otherwise stated, all values are expressed as mean ± standard error of the mean.

Results

Hemodynamic Data

No significant differences in preischemic, baseline hemodynamic data were evident between groups (Table 2). Left ventricular systolic pressure, end-diastolic pressure, $dP/dt$, end-diastolic length, and heart rate were comparable between the two groups. After ischemia and 120 minutes of reperfusion, left ventricular peak pressure, end-diastolic pressure, $dP/dt$, and end-diastolic length were not significantly different from baseline in the group treated with T3. Hearts treated with T3 did have a significantly higher heart rate compared with baseline, however (see Table 2). After ischemia and 30 minutes of reperfusion, the slope of the left ventricular stroke work/end-diastolic relationship, a load-independent index of contractility, was significantly depressed compared with preischemic values in both groups (Fig 1). The group of animals treated with T3, however, demonstrated a gradual improvement in ventricular function over the ensuing 2-hour reperfusion interval, whereas ventricular function in the placebo group remained depressed (see Fig 1). This improvement in functional recovery approached significance after 60

### Table 1. Grading Scale for Myocardial Ultrastructurea

<table>
<thead>
<tr>
<th>Grade</th>
<th>Nuclei</th>
<th>Mitochondria</th>
<th>Myofibrils</th>
<th>Intersstitium</th>
<th>Sarcolemna</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Normal; no margination or membrane disruption</td>
<td>Normal; no swelling or dense bodies</td>
<td>Normal; no edema or disruption</td>
<td>Mild edema</td>
<td>Normal; continuous membrane, glycocalyx still intact</td>
</tr>
<tr>
<td>1</td>
<td>Slight margination and/or perinuclear edema</td>
<td>Slight swelling, no dense bodies</td>
<td>I-band-Z-line fuzziness</td>
<td>Mild to moderate edema</td>
<td>No membrane discontinuity, but glycocalyx disrupted</td>
</tr>
<tr>
<td>2</td>
<td>Margination, edema, and/or nuclear clearing</td>
<td>Swollen, dense bodies and/or clumping of cristae</td>
<td>Band fuzziness, hypercontraction and/or edema</td>
<td>Moderate to severe edema</td>
<td>Some membrane discontinuity, no glycocalyx</td>
</tr>
<tr>
<td>3</td>
<td>Dense margination and nuclear clearing</td>
<td>All or most of above; membrane disruption</td>
<td>Edema, fragmentation, contraction banding</td>
<td>Severe edema</td>
<td>Severely disrupted membrane</td>
</tr>
</tbody>
</table>

*a Organelles were scored on a 0 (no damage) to 3 (complete disruption) scale. All specimens were scored by an independent and blinded microanatomist (K.L.)

### Table 2. Load-Dependent Hemodynamic Dataa

<table>
<thead>
<tr>
<th>Time</th>
<th>LV PKP</th>
<th>LVEDP</th>
<th>$dP/dt$</th>
<th>LV EDL</th>
<th>HR</th>
<th>Wean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>(-)T3</td>
<td>93.1 ± 4.6</td>
<td>6.6 ± 1.3</td>
<td>2310 ± 487</td>
<td>48.6 ± 2.6</td>
<td>140 ± 8</td>
</tr>
<tr>
<td></td>
<td>(+)T3</td>
<td>86.8 ± 4.2</td>
<td>6.5 ± 1.1</td>
<td>2504 ± 334</td>
<td>47.8 ± 0.8</td>
<td>143 ± 9</td>
</tr>
<tr>
<td>120 min R</td>
<td>(-)T3</td>
<td>70.8 ± 7.1</td>
<td>7.5 ± 1.8</td>
<td>1360 ± 335</td>
<td>51.0 ± 2.2</td>
<td>148 ± 8</td>
</tr>
<tr>
<td></td>
<td>(+)T3</td>
<td>81.2 ± 3.4</td>
<td>7.2 ± 1.4</td>
<td>2142 ± 276</td>
<td>50.5 ± 1.3</td>
<td>167 ± 6p</td>
</tr>
</tbody>
</table>

*a Load-dependent hemodynamic data in the control group [(-)T3] and the group treated with T3 [(+)T3] before ischemia and after 120 minutes of reperfusion are detailed. **p < 0.05 versus baseline by analysis of variance. ***p < 0.05 $\chi^2$ analysis.

dP/dt = first time derivative of pressure; HR = heart rate; LVEDL = left ventricular end-diastolic length; LVEDP = left ventricular end-diastolic pressure; LV PKP = left ventricular peak pressure; R = reperfusion; Wean = ability to be weaned successfully off cardiopulmonary bypass after 120 minutes of reperfusion.
Fig 1. The slope of the stroke work/end-diastolic length (SWEDL) relationship was used to quantify left ventricular contractility. Contractile function decreased significantly after ischemia and 30 minutes of reperfusion in both groups. Hearts treated with triiodothyronine (T3), however, demonstrated a significant improvement in systolic function compared with controls, such that after 120 minutes T3-treated hearts had returned to baseline function, whereas systolic function in hearts treated with placebo remained approximately 50% of baseline throughout the reperfusion period. (ANOVA = analysis of variance.)

minutes of reperfusion ($p = 0.10$) and was statistically significant after 90 and 120 minutes of reperfusion ($p < 0.05$). Left ventricular systolic function in the vehicle-treated group did not improve and remained significantly depressed compared with baseline and the T3-treated group.

After 2 hours of reperfusion, all animals treated with T3 were weaned and maintained off cardiopulmonary bypass without the use of inotropic support. In comparison, only 2 of 7 animals in the placebo group were able to be sustained off cardiopulmonary bypass at the end of 2 hours of reperfusion ($p = 0.02$ by $\chi^2$ analysis) (see Table 2). Hemodynamic data from the animals treated with placebo and unable to be fully weaned from bypass were obtained after optimization of preload and with partial support from the cardiopulmonary bypass pump.

Metabolic Data
At baseline, there was no difference in left ventricular adenosine triphosphate (ATP) levels or levels of ATP metabolites between groups (Fig 2; Table 3). After 30 minutes of normothermic ischemia, myocardial ATP levels were significantly depressed in both groups. In both groups, ATP levels remained depressed compared with baseline, preischemic levels throughout reperfusion. There was no difference in ATP levels in the group treated with T3 compared with control.

Similarly, T3 supplementation did not affect ATP metabolism after ischemia (see Table 3). There was no significant difference in myocardial adenosine diphosphate, adenosine monophosphate, adenosine, hypoxanthine, xanthine, or total adenine nucleotide content between the group treated with T3 and the group treated with placebo, either after ischemia or during reperfusion.

Myocardial Ultrastructure
There were no ultrastructural differences between groups before ischemia. Figures 3 and 4 are representative micrographs after ischemia and 120 minutes of reperfusion from the group treated with T3 and the group treated with placebo, respectively. In the postischemic hearts treated with placebo, the sarcolemma is disrupted and incongruent, whereas in the T3-treated group the integrity of the sarcolemmal membrane is preserved. Similarly, the mitochondria in the hearts treated with T3 are less swollen, with less disruption of the cristae. These qualitative differences were significant when myocardial ultrastructure was assessed quantitatively using the schema outline above (see Table 1). The sarcoplasmic reticulum and mitochondria were significantly better preserved when treated with T3 (Table 4). No differences in other cellular organelles were observed.

Comment
It is increasingly evident that cardiopulmonary bypass affects thyroid hormone metabolism, leading to a euthyroid-sick state characterized by low levels of circulating free T3. Although the pathophysiologic significance of this acute stress-induced hypothyroidism is not clear, Novitsky and associates [11, 13-16] have suggested that acute correction of the low T3 state has significant hemodynamic benefit. In extensive laboratory and clinical work, they have suggested that T3 is a positive inotrope,
Table 3. Effect of Triiodothyronine Supplementation on Adenine Nucleotides, Purine Bases, and NAD* After Ischemia

<table>
<thead>
<tr>
<th></th>
<th>Before Ischemia</th>
<th>After Ischemia</th>
<th>30 min Reperfusion</th>
<th>60 min Reperfusion</th>
<th>90 min Reperfusion</th>
<th>120 min Reperfusion</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ADP</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>(-)T₃</td>
<td>5.8 ± 0.4</td>
<td>7.5 ± 0.6</td>
<td>4.0 ± 0.4</td>
<td>4.0 ± 0.6</td>
<td>3.3 ± 0.7</td>
<td>3.2 ± 0.7</td>
</tr>
<tr>
<td>(+)T₃</td>
<td>6.7 ± 1.0</td>
<td>6.5 ± 0.6</td>
<td>5.4 ± 2.0</td>
<td>3.9 ± 0.5</td>
<td>4.3 ± 0.3</td>
<td>3.6 ± 0.4</td>
</tr>
<tr>
<td><strong>AMP</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(-)T₃</td>
<td>0.7 ± 0.3</td>
<td>1.5 ± 0.6ᵇ</td>
<td>0.1 ± 0.08</td>
<td>0.2 ± 0.08</td>
<td>0 ± 0</td>
<td>0.2 ± 0.1</td>
</tr>
<tr>
<td>(+)T₃</td>
<td>0.2 ± 0.07</td>
<td>2.1 ± 0.7ᵇ</td>
<td>0.2 ± 0.07</td>
<td>0.4 ± 0.2</td>
<td>0.2 ± 0.06</td>
<td>0.3 ± 0.09</td>
</tr>
<tr>
<td><strong>Adenosine</strong></td>
<td></td>
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<tr>
<td>(-)T₃</td>
<td>0.2 ± 0.07</td>
<td>1.9 ± 0.5ᵇ</td>
<td>0.14 ± 0.07</td>
<td>0.14 ± 0.06</td>
<td>0.10 ± 0.06</td>
<td>0.1 ± 0.8</td>
</tr>
<tr>
<td>(+)T₃</td>
<td>0.04 ± 0.02</td>
<td>0.8 ± 0.3ᵇ</td>
<td>0.1 ± 0.05</td>
<td>0.1 ± 0.13</td>
<td>0.13 ± 0.08</td>
<td>0.08 ± 0.06</td>
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<tr>
<td><strong>Hypoxanthine</strong></td>
<td></td>
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<td></td>
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</tr>
<tr>
<td>(-)T₃</td>
<td>0</td>
<td>1.2 ± 0.4ᵇ</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>(+)T₃</td>
<td>0</td>
<td>1.6 ± 0.4ᵇ</td>
<td>0.4 ± 0.3</td>
<td>0.06 ± 0.05</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><strong>Xanthine</strong></td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>(-)T₃</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>(+)T₃</td>
<td>0</td>
<td>0.6 ± 0.7</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><strong>NAD</strong></td>
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<td></td>
<td></td>
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<tr>
<td>(-)T₃</td>
<td>4.8 ± 0.4</td>
<td>4.2 ± 0.2</td>
<td>4.8 ± 0.4</td>
<td>4.7 ± 0.6</td>
<td>4.7 ± 1.0</td>
<td>3.9 ± 0.8</td>
</tr>
<tr>
<td>(+)T₃</td>
<td>5.1 ± 0.6</td>
<td>3.1 ± 0.6</td>
<td>4.2 ± 0.3</td>
<td>5.2 ± 0.8</td>
<td>5.3 ± 0.6</td>
<td>4.4 ± 1.5</td>
</tr>
<tr>
<td><strong>EadN</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(-)T₃</td>
<td>34.3 ± 2.2</td>
<td>25.1 ± 3.1ᵇ</td>
<td>20.6 ± 2.4ᵇ</td>
<td>22.7 ± 3.4ᵇ</td>
<td>19.9 ± 4.8ᵇ</td>
<td>20.6 ± 4.3ᵇ</td>
</tr>
<tr>
<td>(+)T₃</td>
<td>36.9 ± 3.9</td>
<td>23.8 ± 4.4ᵇ</td>
<td>22.1 ± 4.7ᵇ</td>
<td>19.1 ± 4.1ᵇ</td>
<td>20.9 ± 3.8ᵇ</td>
<td>16.4 ± 3.2ᵇ</td>
</tr>
</tbody>
</table>

* Myocardial adenine nucleotide and metabolite levels were measured at baseline, immediately before reperfusion (after ischemia), and every 30 minutes during reperfusion. Values are expressed as nanomoles per milligram of protein. ᵃᵇ p < 0.05 versus baseline by analysis of variance.

ADP = adenosine diphosphate; AMP = adenosine monophosphate; EadN = total adenine nucleotides; NAD = nicotinamide adenine dinucleotide; T₃ = triiodothyronine.

perhaps acting by improving the "aerobic capacity" of the myocardium after ischemia and thereby leading to an enhanced availability of myocardial high-energy phosphates available for contractile work [15]. However, their studies have lacked a load-independent demonstration of enhanced contractility.

Thyroid hormone has multiple systemic effects and is a potent mediator of cellular thermogenesis and local arterial vasodilatation [21-24]. Chronic hyperthyroidism is well recognized to lower systemic vascular resistance. Acute T₃ administration may also profoundly alter ventricular loading conditions. Load-dependent estimates of ventricular function such as cardiac output or stroke work, therefore, may be significantly altered on this basis alone, without any change in myocardial contractility [25]. Additionally, cardiopulmonary bypass induces large fluid shifts, which alter ventricular loading conditions.

In an earlier study using an isolated rabbit heart model to rigorously control ventricular loading conditions, we were unable to demonstrate an inotropic effect of acute T₃ administration in normal hearts, although acute T₃ administration significantly and rapidly improved left ventricular systolic function after ischemia [17]. This study was not designed to determine the mechanism of action of T₃ activity after ischemia.

The present study confirms these earlier findings of a rapid enhancement of myocardial mechanical perfor-

Fig 3. A representative electron micrograph from the left ventricle of a heart treated with placebo after 30 minutes of ischemia and 120 minutes of reperfusion. The arrow points to a break in the sarcolemma. Mitochondria (m) are swollen and edematous, whereas the contractile filaments are well preserved.
Mitochondria and the sarcoplasmic reticulum from the group of animals treated with triiodothyronine (T3) showed a significant preservation of normal architecture after ischemia and reperfusion compared with controls. Data are shown as mean ± standard error of the mean.

Our data, however, do not support the hypothesis that T3 improves myocardial performance by enhancing the availability of myocardial high-energy phosphate stores. In contrast to Novitsky and associates’ work, we did not demonstrate an effect of T3 on ATP metabolism after ischemia. In both placebo- and T3-treated groups, myocardial ATP levels decreased 50% to 60% from baseline after ischemia and remained depressed throughout 2 hours of reperfusion. Similarly, there was no difference in ATP degradation products between groups. Stores of diffusible metabolites (adenosine monophosphate, hypoxanthine, and xanthine) were increased in both groups after ischemia but before reperfusion, and were washed out with reperfusion. Whether this is consistent with Novitsky and associates’ study is unknown because ATP metabolites were not reported in their report [15].

Although total myocardial adenine nucleotide stores can be measured quite accurately, the intracellular distribution of high-energy phosphates available for contraction. In an earlier study, they reported a significant increase in myocardial ATP levels after ischemia in a group of baboons treated with T3 compared with controls. Data detailing ATP degradation products were not presented in their report. It was hypothesized that the reported increase in myocardial ATP stores was mediated through the activity of T3 on the adenine nucleotide translocase moiety located on the inner mitochondrial membrane. This enzyme, a regulator of adenosine diphosphate/ATP exchange across the inner mitochondrial membrane, binds T3 and has been demonstrated to be rapidly up-regulated by exogenous administration of T3 [26].

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distribution of ATP stores is difficult to quantify. Possibly, changes in intracellular compartmentalization of ATP may be affected by T3, perhaps through the adenine nucleotide translocase moiety, and thereby affect contractile performance. These data do not address this hypothesis.

Although thyroid hormone exerts powerful norepinephrine-mediated effects on protein synthesis, the enhanced recovery of ventricular function is rapid enough to preclude a nuclear-mediated, transcriptional-based mechanism for enhanced contractility within this early reperfusion period [27]. It is doubtful that any significant change in myosin isoform expression could account for the functional improvement seen in this acute setting, although a posttranscriptional mechanism affecting protein synthesis could have a rapid effect on contractility.

Triiodothyronine has several sites of action at the plasma membrane that may affect ventricular function. Of particular interest are the nonnuclear sites of action that affect calcium cycling within the cell. Triiodothyronine has been demonstrated to increase intracellular calcium content in isolated myocytes by upregulating the activity of the sarcoplasmic reticulum Ca2+ adenosine triphosphatase [28, 29]. Also, acute T3 administration has been demonstrated to affect sodium channels on the plasma membrane, resulting in an increased inward sodium flux, which may affect contractility [30]. Both of these effects of T3 administration on ion transport are rapid in onset and may enhance myofibrillar shortening. Whether the anatomical observation of superior sarcolemmal preservation with T3 infusion is the ultrastructural correlate to the functional changes observed is not clear. However, both the sarcolemmal membrane and the mitochondria have been postulated to be mechanistically related to improved cardiac function with T3 infusion after ischemic injury. Further work will be required to test this hypothesis.

In summary, T3 supplementation significantly improved left ventricular functional recovery after ischemia but did not affect ATP metabolism. Levels of ATP and its metabolites were depressed after ischemia in both the group treated with T3 and placebo. This suggests that the enhancement of left ventricular systolic performance after ischemia with T3 supplementation is not mediated by an increased availability of myocardial high-energy phosphates. Although not immediate, the improvement in function was rapid enough to likely preclude a nuclear-mediated mechanism involving myosin isoform switches. The improvement in recovery of the sarcoplasmic reticulum and mitochondria on transmission electron micrographs suggests that these organelles may be related to the improvement in postischemic performance. Changes in ion flux across the plasma membrane and sarcoplasmic reticulum remain an attractive hypothesis for the enhancement of left ventricular functional recovery after ischemia that occurs with acute T3 supplementation.

References
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DISCUSSION

DR JULIE A. SWAIN (Las Vegas, NV): Your conclusion is that you do not know the mechanism, but I am sure you have speculated on the mechanism. What are your speculations? My second question is that your study shows that giving T3 during an operation appears to be beneficial. Have you considered preoperative treatment? Clinically we would think that this might help the heart failure and valve replacement patients to try to effect a V1-V3 switch of the myosin heavy chain isoforms as suggested by the work from Mahdavi and Nadal-Ginard [1].

DR DYKE: Thank you for your comments. The mechanism, as we alluded to, has not been well described or proved; however, we think that one particular area, namely, the cycling of intracellular calcium or sodium ions, either through the calcium adenosine triphosphatase or the sodium-potassium adenosine triphosphatase, respectively, may contribute to the improvement in function that we see after ischemia. We believe that the rapid response, within an hour, rules out a nuclear-based, transcription-mediated mechanism.

Regarding your second question, we have not looked at preoperative T3 supplementation in this model, although similar studies in an isolated heart model are ongoing. However, one of the rationales for looking at T3 supplementation perioperatively is that most patients are euthyroid preoperatively, and the initiation of the euthyroid-sick syndrome seems to occur with cardiopulmonary bypass and continues through the early postoperative period.

DR DIMITRI NOVITZKY (Tampa, FL): I would like to congratulate you for this excellent presentation, which further supports the work that was initiated in Cape Town using T3 for nonthyroid conditions. Our first observation of a low free T3 level was inducing experimental brain death in baboons. This free T3 reduction followed an initial catecholamine storm; the thyroid-stimulating hormone level remained unchanged. A similar response was observed during and after cardiopulmonary bypass.

Triiodothyronine was administered to pigs subjected to cardiopulmonary bypass for 3 hours of aortic cross-clamping and cardioplegic arrest; these animals were easily weaned from the heart-lung machine, sustaining good hemodynamics for 2 hours. However, animals not receiving T3 remained bypass-dependent and died by the end of the experiment. Further studies performed in baboons showed that the administration of T3 definitely prevented the cardiac tissue lactic acidosis and normalization of ATP levels.

Approximately 8 years ago, I administered T3 to a patient who underwent emergent mitral valve replacement. She was septic, had bacterial endocarditis and pneumonia, and was intubated for pulmonary edema. After the valve replacement, in spite of multiple defibrillation attempts, maximum inotropic support, and boluses of calcium, she remained dependent on cardiopulmonary bypass. The anesthesiologist in charge of the case suggested that we give T3 to this patient. We already had gained significant experience administering T3 and rescuing hemodynamically unstable brain-dead organ donors dependent on high-dose inotropic support, observing excellent hemodynamics in the recipients after cardiac transplantation. The patient received 6 μg of T3 as a bolus; within few minutes, the fibrillation became more pronounced and the heart was defibrillated successfully, starting in sinus rhythm and allowing us to discontinue cardiopulmonary bypass within 20 minutes with only minimal inotropic support.

The low free T3 state is part of the “euthyroid sick syndrome” observed in acute shock states and chronic conditions such as cancer or starvation. I have found T3 replacement beneficial after acute insults, and I believe this condition should be totally differentiated from the chronic states. In the acute phase such as sepsis, hemorrhagic shock, in brain-dead organ donors, and in patients on cardiopulmonary bypass, the administration of T3 was beneficial in the recovery of the myocardium, allowing rapid reduction of inotropic support. Triiodothyronine also has systemic effects and may also have a beneficial effect on other organs.

At the present stage, two mechanisms of action are apparent: the first seems to be extranuclear, affecting various calcium-dependent adenosine triphosphatases acting on the contractile apparatus and also on the mitochondria-stimulating aerobic metabolism, and the second is mediated via DNA-RNA-protein synthesis.

Have you measured free T3 levels and the impact of heparin on these? Have you measured levels of other hormones besides T3 such as reverse T3, total thyroxine, free thyroxine, thyroid-stimulating hormone, and thyrotropin-releasing hormone?

I congratulate you for this excellent piece of work.

DR DYKE: Thank you, Dr Novitzky. We are well aware of and appreciate your work beginning in the early 1980s. A stimulus for this study was the increase in ATP with T3 supplementation that you described in one of your early studies. Although we demonstrated an improvement in cardiac performance, we were unable to show a change in ATP levels with T3 supplementation as your earlier work did, and that has led us away from an increase in oxidative phosphorylation capacity of the myocardium as a primary mechanism of action. We have not looked at the effect of heparin on T3 levels.

Reference