Myocardial Catecholamine Content After Heart Transplantation

Vera Regitz, MD, Claus Bossaller, MD, Rudolf Strasser, MD, Stephan Schüler, MD, Roland Hetzer, MD, and Eckart Fleck, MD

Myocardial catecholamine levels have not yet been determined in the transplanted human heart. We measured norepinephrine, epinephrine, and dopamine in endomyocardial biopsies from 19 short-term (organ age, $6.6 \pm 6$ months) and five long-term (organ age, $62 \pm 2$ months) heart transplant patients. Results were compared with those from 10 normal control subjects. In 17 of 19 short-term heart transplant patients, myocardial catecholamines were undetectable, indicating values below 0.1 pg/µg noncollagen protein, which was the detection threshold of our assay. In the remaining two patients, myocardial catecholamines (pg/µg noncollagen protein) were norepinephrine (1.4 and 3.2), epinephrine (0.8 and 1.9), and dopamine (0.9 and 2.3), respectively. In the five long-term heart transplant patients, myocardial catecholamines were not detected. Catecholamine concentrations in 10 healthy control subjects were norepinephrine (10.3±2.9), epinephrine (0.36±0.51), and dopamine (0.52±0.40). Low myocardial norepinephrine levels (<20% of control values) with unexplained high levels of epinephrine and dopamine were found in single transplant patients. In most heart transplant patients, however, myocardial catecholamines were undetectable up to five years after transplantation, indicating that the adrenergic response of these hearts probably depends on variations in plasma catecholamines or cardiac β-receptor density. (Circulation 1990;82:620–623)

Myocardial norepinephrine and epinephrine are depleted in all portions of short-term autotransplanted canine hearts.1,2 In long-term autotransplanted canine hearts about 50% of control norepinephrine and epinephrine are found.1,2 Dopamine content, however, is only modestly decreased in short- and long-term autotransplanted canine hearts.2,3 Corresponding data for the transplanted human heart do not yet exist. Therefore, we measured the myocardial norepinephrine, epinephrine, and dopamine content in endomyocardial biopsies of patients that had undergone heart transplantation less than 18 months or more than 5 years ago. The concentrations measured were compared with those in 10 healthy control hearts.

Methods

Patients

Endomyocardial biopsies were obtained from the right ventricular septum in 19 patients 1–18 months after heart transplantation and in five patients more than 5 years after transplantation; 10 control subjects with normal left ventricular function, without coronary, valvular, or hypertensive heart disease were studied. Immunosuppression in heart transplant patients consisted of cyclosporine A (dosage according to blood levels: 600 ng/ml during the first 6 months and 300–400 ng/ml later), prednisolone (2 mg/kg day with tapering doses), and azathioprine (0.5–1.5 mg/kg day). Cardiac catheterization in control subjects was performed because of chest pain; after exclusion of coronary or valvular heart disease, an endomyocardial biopsy was taken to preclude myocarditis. A complete left and right cardiac catheterization with left and right ventricular angiography, coronary angiography, and endomyocardial biopsy were performed in all patients. All patients gave informed consent.

Biochemical Studies

Endomyocardial biopsies were extracted in 4% perchloric acid/5 mM glutathion. After centrifugation, catecholamines in the supernatant were adsorbed on aluminum oxide, washed, eluted, and measured by high-performance liquid chromatography (HPLC) and electrochemical detection as
TABLE 1. Organ Age and Hemodynamic Parameters of Short-term Transplanted Patients Without Detectable Myocardial Catecholamines

<table>
<thead>
<tr>
<th>Patient</th>
<th>Organ age (mo)</th>
<th>Systemic BP (mm Hg)</th>
<th>LVEDP (mm Hg)</th>
<th>Cardiac index 1/m²/min</th>
<th>LVEF (%)</th>
<th>RVEF (%)</th>
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<td>±4.2</td>
<td>±8</td>
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BP, blood pressure; LVEDP, left ventricular end-diastolic pressure; LVEF, left ventricular ejection fraction; RVEF, right ventricular ejection fraction.

described previously. The sensitivity of the assay amounted to 5 pg (norepinephrine and epinephrine) and 10 pg (dopamine) per HPLC injection. This corresponds to a detection limit of approximately 20 pg/mg wet weight or 0.1 pg/μg noncollagen protein in a biopsy of 1–2 mg wet weight (corresponding to 160–320 μg noncollagen protein). The coefficient of variance for the determination of norepinephrine by HPLC was 7% in previous studies. For the determination of noncollagen protein, pellets were rehomogenized, collagen proteins were extracted overnight in 0.05N NaOH according to Lilienthal et al, and a protein determination with Coomassie brilliant blue R-250 was performed in the remaining supernatant.

Data are presented as mean±SD.

Results

The concentration of norepinephrine in the right ventricle of 10 healthy control subjects was 10.9±2.9 pg/μg noncollagen protein. Concentrations of epinephrine (0.36±0.51) and dopamine (0.52±0.40) were low in this group.

In 17 of 19 patients studied 1–18 months after heart transplantation, norepinephrine could not be detected in the endomyocardial biopsy. Because the detection limit is 0.1 pg/μg noncollagen protein, it is safe to say that these biopsies contained less than one hundredth of the normal myocardial norepinephrine content. In these 17 patients without measurable amounts of norepinephrine, neither epinephrine nor dopamine could be detected. On average, the 17 patients were examined 6.6 months (range, 1–18 months) after transplantation (Table 1). Mean blood pressure, left ventricular end-diastolic pressure, cardiac index, and left and right ventricular ejection fractions were all within the normal range (Table 1).

In two of the short-term heart transplant patients, catecholamines (pg/μg noncollagen protein) were detected in the myocardium: norepinephrine (1.4

TABLE 2. Organ Age and Hemodynamic Parameters of Short-term Transplanted Patients With Detectable Myocardial Catecholamines

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<th>Patient</th>
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<td>52</td>
</tr>
</tbody>
</table>

BP, blood pressure; LVEDP, left ventricular end-diastolic pressure; LVEF, left ventricular ejection fraction; RVEF, right ventricular ejection fraction.
and 3.2) was about 20% of control values; epinephrine (0.8 and 1.9) and dopamine (0.9 and 2.3) were also found. These patients did not differ from the whole group with respect to organ age, mean blood pressure, left ventricular end-diastolic pressure, cardiac index, and left and right ventricular ejection fractions (Table 2).

In five long-term heart transplant patients, norepinephrine, epinephrine, and dopamine were not present in measurable amounts in the biopsies. Hemodynamic parameters in this group were comparable to short-term heart transplant patients (Table 3).

### Discussion

For the first time, myocardial catecholamines have been measured in endomyocardial biopsies from transplanted human hearts. In most short-term and long-term heart transplant patients, we found a complete depletion of myocardial norepinephrine, epinephrine, and dopamine. This is partially in agreement with findings in experimental animals, in which depletion of norepinephrine and epinephrine has been found shortly after heart transplantation.1,2 However, in long-term autotransplanted canine hearts, norepinephrine content was about 50% of control values.2 This could not be confirmed in our patients. Because the follow-up period encompassed only 5 years in our study, further investigations in humans are needed.

In contrast to depleted norepinephrine, dopamine levels remained relatively high in short-term autotransplanted canine hearts.2 This suggests an analogy with other pathological states, such as heart failure, in which norepinephrine is also depleted and relative concentrations of dopamine are also increased.4,8 To explain such findings, nonneuronal pathways for dopamine synthesis have been postulated in transplanted canine hearts.2 We obtained conflicting data in human hearts: Although we can confirm relatively high levels of dopamine in heart failure,4 we did not find high concentrations of dopamine in most transplanted hearts. Only two of the 24 patients had measurable myocardial dopamine levels, and in these patients, norepinephrine and epinephrine were also found. Thus, catecholamine metabolism in transplanted human hearts may differ in some quantitative or qualitative aspects from the described animal models.1-3

Reinnervation occurs frequently in canine hearts but has not been documented in human hearts.9 The presence of all three myocardial catecholamines in unusual concentrations in some of the transplanted hearts may be explained by the storage of catecholamines in the remaining nervous structures, in postganglionic parasympathetic nerves, or by binding or storage of plasma catecholamines in nonneuronal structures.2,9 Finally, ingrowth of nerve endings together with blood vessels from the surgically manipulated pericardium cannot be excluded. Simple blood contamination of the biopsies is not a sufficient explanation because plasma catecholamines occur at a concentration of about 1/1,000 of tissue catecholamines.

Because myocardial catecholamines cannot be detected in most heart transplant patients, the function of the transplanted human heart must increase due to plasma catecholamines, if mediated through the adrenergic system. In a recent study,10 we compared circadian plasma catecholamine levels in 10 heart transplant patients with levels in 11 age-matched control subjects. No differences were found between patients and control subjects.10 The effect of normal amounts of circulating catecholamines on the transplanted heart may be enhanced if β-receptor density is increased. Indeed, increased β2-receptor density of the denervated transplanted heart has been reported, although total β-receptor density was unchanged.11,12 Thus, it cannot be excluded that the adrenergic support of the transplanted human heart involves activation of up-regulated β2-adrenergic receptors by circulating catecholamines.

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### References


**KEY WORDS**  norepinephrine  epinephrine  dopamine  heart transplantation
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