Left Ventricular Myocardial Structure in Aortic Valve Disease Before, Intermediate, and Late After Aortic Valve Replacement

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Left ventricular biplane cineangiography, micromanometry, and endomyocardial biopsies were performed in 27 patients with aortic stenosis (AS) and in 17 patients with aortic insufficiency (AI). Twenty-three patients with AS and 15 with AI were restudied at an intermediate time (18 months after successful valve replacement), and nine patients with AS and six with AI were restudied late (70 and 62 months after surgery). Biopsy samples were evaluated for muscle fiber diameter, percent interstitial fibrosis, and volume fraction of myofibrils. In control biopsy samples obtained from five donor hearts at transplantation, these morphometric variables averaged 21.2 μm, 7.0%, and 57.2%, respectively. After surgery, mass determined by cineangiography decreased from 186 to 115 and 94 g/m² in patients with AS and from 201 to 131 and 93 g/m² in patients with AI. At the three studies, muscle fiber diameter was 30.9, 28.0, and 28.7 μm in patients with AS and was 31.4, 27.6, and 26.4 μm in patients with AI. Percent interstitial fibrosis was 18.2, 25.8, and 13.7% in patients with AS and was 20.4, 23.7, and 19.2% in patients with AI. Left ventricular fibrous content decreased from 34.2 to 29.8 and to 12.7 g/m² in patients with AS and from 42.1 to 28.9 and to 18.9 g/m² in patients with AI. Volume fraction of myofibrils was 57.7, 56.8, and 49.0% in patients with AS and was 56.8, 56.6 and 48.8% in patients with AI. Thus, the decrease of muscle mass determined by cineangiography at the intermediate time after valve replacement is mediated by regression of myocardial cellular hypertrophy in patients with AS and AI and in addition by a decrease of fibrous content in patients with AI. Late after surgery, left ventricular fibrous content also decreases in patients with AS. This late decrease associated with minor changes of end-diastolic volume may be important for improvement of increased diastolic myocardial stiffness. Even 6–7 years after valve replacement, incomplete regression of structural abnormalities of left ventricular hypertrophy still exists compared with the normal myocardium. The residually increased relative interstitial fibrosis and the small late postoperative decrease of volume fraction of myofibrils, associated with a prosthesis-related slight left ventricular pressure increase, are at the origin of a persistent systolic overload at the myofibrillar level. (Circulation 1989;79:744–755)

Clinical Investigation

Chronic pressure or volume overload or both in aortic valve disease is associated with marked left ventricular hypertrophy, evidenced by angiography and microscopy. This process of secondary hypertrophy is accompanied invariably by an increase in interstitial connective tissue. Correction of the abnormal hemodynamic burden by aortic valve replacement has resulted in a regression of the increased left ventricular muscle mass early and intermediate after surgery. Eight years after valve replacement, we reported that the process of mass regression was essentially complete because no difference in angiographic mass was found between late postoperative patients with aortic stenosis or aortic insufficiency and control subjects. But what about the microscopic structure of the left ventricular myocardium after successful valve replacement? In a previous investigation of 21 patients of whom 10 had aortic stenosis, five had combined aortic valve lesion, and six had chronic aortic insufficiency, we reported a significant decrease

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of muscle fiber diameter 18 months after surgery. An increase of percent interstitial nonmuscular tissue occurred that was significant in the patients with aortic stenosis. No change of left ventricular fibrous content was observed at this intermediate postoperative time. The present study focuses on our entire experience in evaluating myocardial microscopic structure from left ventricular endomyocardial biopsies in patients with aortic valve disease intermediate and late after valve replacement.

Methods

Preoperative hemodynamic measurements and left ventricular endomyocardial biopsies were obtained in 27 patients (21 men and six women) with aortic stenosis (group 1) and in 17 patients (14 men and three women) with aortic insufficiency (group 2) between 1977 and 1982.

In group 1, mean age was 52 years (range, 25–67 years). Twenty patients had practically "pure" aortic stenosis; in 15 of them, a minimal aortic reflux was present. Aortic regurgitant fraction as determined by thermodilution was always less than 0.20 (mean, 0.13). The planimetered mean systolic pressure gradient varied between 45 and 105 mm Hg, and the aortic valve area, which was calculated by the Gorlin formula with the regurgitant volume taken into account, was between 0.3 and 0.8 cm². The remaining seven patients of group 1 had predominant aortic stenosis. Aortic valve area was always 1 cm² or less (mean, 0.86 cm²); the mean systolic pressure gradient was between 46 and 107 mm Hg. In group 1 patients, aortic valve area averaged 0.68 cm² (0.3–1.0 cm²), and the mean systolic pressure gradient averaged 71 mm Hg (45–107 mm Hg). Coronary arteriography was performed in all patients with aortic stenosis. In one patient, there was a 50% stenosis of the left anterior descending coronary artery with no regional contraction abnormalities on the left ventricular cineangiogram.

In group 2, mean age was 43 years (range, 21–63 years). Thirteen patients had "pure" or almost pure aortic insufficiency; in three of them, a small gradient (mean systolic pressure gradient between 6 and 14 mm Hg) was present. Aortic regurgitant fraction varied between 0.47 and 0.80. Four patients of group 2 had predominant aortic insufficiency. Aortic valve area ranged between 1.4 and 2.1 cm², aortic regurgitant fraction between 0.38 and 0.63. In group 2 patients, aortic regurgitant fraction averaged 0.58 (range, 0.38–0.80). Two patients had mild mitral regurgitation with a regurgitant fraction of 0.25 and 0.26. Coronary arteriography was performed in 14 of the 17 patients and showed no coronary artery disease. The remaining three patients who had no coronary arteriography were 21, 28, and 37 years old.

Twenty-three patients without or with minimal heart disease served as controls (group 3). Mean age was 37 years (range, 16–56 years). No biopsies were performed in these patients.

All 67 studied patients were in sinus rhythm, and the duration of the QRS complex did not exceed 0.11 seconds.

In 34 patients, the aortic valve was replaced with a bioprosthesis, and in 10 patients, a mechanical prosthesis (Björk-Shiley or Saint Jude Medical) was implanted. Prosthesis size averaged 27.2 mm. In three patients of group 1 and in one patient of group 2, a myectomy from an asymmetrically hypertrophied septum was performed. In one patient of group 2, anuloplasty of the mitral valve and aortic valve replacement were performed. After surgery, mean systolic pressure gradient was determined in 39 patients, and it varied between 0 and 24 mm Hg (mean, 9.3 mm Hg). Minimal aortic reflux was observed in nine of 39 patients in whom postoperative supravalvular cineangiography was performed.

Twenty-three patients of group 1 were recatheterized at an intermediate time, that is, 18 months (range, 9–28 months) and nine patients at a late time, that is, 70 months (range, 41–99 months) after successful aortic valve replacement. Five patients were catheterized three times (before surgery and intermediate and late after surgery). Fifteen patients of group 2 were recatheterized intermediate, that is, 18 months (range, 11–25 months) and six patients late, that is, 62 months (range, 49–70 months) after surgery. Four patients were catheterized three times.

At the late restudy, none of the 15 patients had signs of aortic insufficiency at auscultation. The systolic peak-to-peak gradient between the simultaneously measured pressures in the left ventricle (by micromanometer) and the brachial artery (by sphygmomanometer) averaged 4.5 mm Hg (range, 0–24 mm Hg). Prosthesis size varied between 25 and 31 mm (mean, 28.5 mm). In only one patient, the gradient exceeded 9 mm Hg. Left ventricular pressure loading, however, was not especially marked in this patient because left ventricular peak systolic pressure was practically the same (136 mm Hg) as in the whole group (138 mm Hg) of 15 late postoperative patients.

Catheterization, Cineangiography, and Endomyocardial Biopsies

Informed consent was obtained from all patients under a protocol approved by the human studies committee of the University Hospital. Regarding postoperative studies, the patients were asked by letter for their consent to undergo recatheterization. Among the 44 patients who were asked to undergo recatheterization at the intermediate time after surgery, 38 consented, and six consented later for the late postoperative study. Of the 38 patients who underwent the first restudy, nine consented for the late postoperative catheterization and 29 refused. Nine were not candidates for late restudy. Of these, three had to be reoperated on because of failure of the bioprosthesis, two were 70 years old or older, malignant lymphoma developed in one, one suffered a myocardial infarction, left bundle branch
block developed in one, and one had left the country. Premedication consisted of 10 mg chlordiazepoxide (Librium) given orally 1 hour before the procedure. Left ventricular pressure was measured with a transseptally introduced Statham SF-1 (Puerto Rico) or a 7F Millar micromanometer (Houston, Texas). The micromanometer was calibrated by superimposing the high-fidelity pressure tracing on the conventional pressure tracing. Aortic pressure was measured through a fluid-filled 8F pigtail catheter. No invasive arterial pressure measurements were performed late after surgery except in one of the 15 patients studied. A peripheral lead of the standard electrocardiogram and of the phonocardiogram was recorded together with the left ventricular high-fidelity pressure tracing and its first derivative at a paper speed of 200 or 250 mm/sec with an Electronics for Medicine (Pleasantville, New York) DR-16 or VR-12 oscillograph.7,13

Left ventricular cineangiography at a film speed of 50 frames/sec was performed in the right and left anterior oblique projections by our standard technique.18 For the quantification of end-diastolic and end-systolic volumes the area-length method was used. The range of normality for left ventricular biplane ejection fraction in our laboratory is between 57% and 83% (mean, 68%).18 Left ventricular end-diastolic wall thickness was measured from the anterior wall on the right anterior oblique cineangiogram, and muscle mass was calculated by the technique of Rackley et al.19 End-systolic wall thickness was calculated from the end-diastolic muscle volume and the end-systolic intracavitary dimensions assuming that the muscle volume remains constant throughout the cardiac cycle.

As a measure of left ventricular afterload, circumferential peak systolic wall stress (S peak) was calculated by the technique of Gaasch et al21 with end-diastolic and end-systolic left ventricular dimensions from cineangiography, wall thicknesses, and peak systolic pressure. End-diastolic circumferential wall stress was obtained from end-diastolic left ventricular dimensions, wall thickness, and pressure.7,20 Maximal rate of rise of left ventricular circumferential wall stress (dS/dt max) was determined from end-diastolic left ventricular dimensions, wall thickness, and pressure.22 The relation between dS/dt max and end-diastolic wall stress (preload) was used as an "isovolumic ventricular function" diagram.23 Peak measured velocity of shortening of the contractile elements (V peak) was obtained as the maximal instantaneous quotient of [(dP/dt)/28 × P].7

Left ventricular endomyocardial biopsies were performed with the King's College bioptome (Olympus, Tokyo, Japan), which was introduced into the left ventricle through the 11.5F Brockenbrough catheter (USCI, Billerica, Massachusetts) previously used for the insertion of the tipmanometer catheter. In each patient at each catheterization, 2–3 biopsy samples were obtained from the anterolateral wall of the left ventricle.13 The biopsy specimens were fixed in glutaraldehyde, embedded in epon, and evaluated by light microscopy in semithin sections.13 Morphometric techniques were used for the quantitative evaluation. From several sections of 2–3 biopsy samples of each catheterization, muscle fiber diameter and relative interstitial fibrosis were determined with a mechanical-optical pen (MOP, Kontron, Zurich, Switzerland). Occasionally, contraction bands and mechanical disruption of tissue structure were seen. Those portions of the biopsy samples were excluded from the analysis. Interstitial space or interstitial nonmuscular tissue was estimated with the point-counting system.11–13,16,24,25 Areas with arterioles and perivascular tissue were excluded. The intersection points of the counting grid overlying interstitial nonmuscular tissue were expressed as percentage of the entire number of evaluated intersection points and were referred to as relative interstitial fibrosis.12,16,26,27 The term "interstitial fibrosis" for this nonmuscular interstitial tissue is somewhat incorrect, but because fibrous tissue is the predominant component of the interstitial space, we13 like others12,16,26,27 have used the term "fibrosis" in this context. Usually, 1,000 intersection points were counted for the estimation of the average interstitial fibrosis in each patient at each catheterization. Previously determined interobserver variability for interstitial fibrosis was 6.2%.13

The morphometric method has yielded directionally similar results for the quantification of interstitial fibrosis as the determination of hydroxyproline concentration that was used as a marker of connective tissue in the experimental animal.28 The volume fraction of myofibrils was evaluated at a magnification of 1,000 × 1 with oil immersion and phase contrast microscopy. The technique was described previously by one of us (G.M.).29 For four randomly chosen sections of each patient and at least three random test areas from each section, the volume fraction of myofibrils was determined with a counting grid with 36 intersection points. This light microscopic technique has been validated by comparison with the electronmicoscopic method.29 A strong correlation was observed between light microscopic and electronmicroscopic values of volume fraction of myofibrils with a slight overestimation of the light microscopic values by 2.5 vol%. The interobserver variability with the light microscopic technique in 20 of the patients in the present study was 3.5%. Of importance, the determination of the volume fraction of myofibrils is not influenced by the myocardial cell length (which was not measured) because according to basic principles of morphometry, counting the number of points overlaying a certain structure (in this case myofibrils) permits quantitative determination of the volume of the structure under investigation in relation to the volume of the entire tissue (i.e. the intramyocyte space) within the boundaries of the grid.12

Fibrous content (FC, g/m²) of the left ventricle was calculated as FC = LMMI × IF/100 where LMMI
is left ventricular angiographic mass index (g/m²), and IF is percent interstitial fibrosis (%).

The endomyocardial biopsy samples obtained at catheterization are tiny, and the question arises how representative their structure is for the rest of the myocardium. In biopsy pairs obtained from patients with cardiomyopathies, Baandrup et al. have reported a coefficient of variance for determining fiber diameter of 18.6% and for determining the percentage of interstitium of 28.9%. In patients with aortic valve disease, morphologic changes in the myocardium seem, however, more uniformly distributed over the whole ventricle, although some focal processes may occasionally occur. Moreover, variability due to sampling appears of lesser importance in the preoperative and postoperative comparisons because the biopsy specimens were removed from the same endocardial area before and after surgery. Control morphologic data for muscle fiber diameter, nonmuscular tissue (percent interstitial fibrosis), and volume fraction of myofibrils were obtained from left ventricular endomyocardial biopsy samples of five donor hearts at the time of heart transplantation. Age of the donors (four men and one woman) averaged 26 years (19–34 years). Muscle fiber diameter averaged 21.2 μm (range, 18.4–22.8 μm), relative interstitial fibrosis averaged 7.0% (range, 5.7–9.4%), and volume fraction of myofibrils averaged 57.2% (range, 53.5–60.6%).

Statistics

One-way analysis of variance was used for comparing the preoperative, intermediate postoperative, and late postoperative patients in groups 1 and 2. If the analysis showed a significant difference, the Scheffe’s test was applied for the determination of the p values. When only two groups were compared, the unpaired Student t test was applied. Preoperative versus late postoperative comparisons in those specific patients in whom paired preoperative and late postoperative data were available were performed by the paired Student t test. In all tables, values are mean ± SD.

Results

Preoperative Left Ventricular Structure and Its Relation to Contractile Function

In patients with aortic stenosis (group 1) and with aortic insufficiency (group 2), muscle fiber diameter (30.9±4.7 and 31.4±4.4 μm) and relative interstitial fibrosis (18.2±6.2 and 20.4±4.9%) were increased (p<0.001) compared with the respective findings (21.2±2.0 μm; 7.0±1.8%) in the five heart donors (Figure 1). In contrast, the volume fraction of myofibrils did not differ between the three groups (57.7±5.9, 56.8±4.6, and 57.2±2.6%). No difference occurred in any of the morphometric variables between group 1 and group 2, including left ventricular fibrous content (34.2±17.2 and 42.1±16.5 g/m²). Thus, the morphometric findings of the two groups were pooled and compared with measures of left ventricular contractile function. Muscle fiber diameter showed a weak inverse correlation with left ventricular ejection fraction (r=−0.37, p<0.02) and Vpm (r=−0.32, p<0.05). No correlation occurred between percent interstitial fibrosis and ejection fraction and Vpm, respectively. Neither ejection fraction nor Vpm correlated with the volume fraction of myofibrils. This quantity also did not correlate with muscle fiber diameter or percent interstitial fibrosis. Muscle mass index determined by cineangiography correlated inversely with ejection fraction (r=−0.43, p<0.005) and Vpm (r=−0.49, p<0.001).

Preoperative and Postoperative Comparisons of Morphometric, Hemodynamic and Angiographic Findings

Intermediate postoperative evaluation. Muscle fiber diameter in groups 1 and 2 (28.0±3.6 and 27.6±3.2 μm, respectively) was smaller (p<0.05) at intermediate after surgery than before surgery (Figure 1). In group 1, percent interstitial fibrosis (25.8±8.7%) was greater (p<0.01) than before surgery; a slightly, but not significantly, increased percent interstitial fibrosis was observed in group 2 (23.7±8.6%). In both groups, muscle fiber diameter and percent interstitial fibrosis remained significantly increased compared with the control patients (Figure 1). Volume fraction of myofibrils did not change intermediate after valve replacement in either group 1 (56.8±4.8%) or in group 2 (56.6±3.5%). Left ventricular fibrous content remained unchanged in group 1 (29.8±11.0 g/m²), but in group 2, it was smaller (28.9±8.4 g/m²) than before surgery (p<0.05).

Ejection fraction and Vpm did not change intermediate after valve replacement, whereas muscle mass index determined by cineangiography was smaller (p<0.001) in both groups than before surgery (Table 1, Figure 1). Peak systolic wall stress decreased significantly in both groups (p<0.001 and p<0.05).

Late postoperative evaluation. In group 1, muscle fiber diameter tended to increase slightly (28.7±4.4 μm) late after surgery and, hence, was not different from the preoperative value (Figure 1). In group 2, a further slight decrease to 26.4±1.0 μm occurred (Figure 1), this value being different (p<0.05) from the preoperative one. Percent interstitial fibrosis decreased significantly (p<0.001) in group 1 (13.7±3.6%) and insignificantly in group 2 (19.2±6.2%) compared with the intermediate post-surgical data. Percent interstitial fibrosis late after surgery was similar to the value before surgery in both groups. Muscle fiber diameter and percent interstitial fibrosis remained significantly increased late after surgery compared with that in control patients (Figure 1). Volume fractions of myofibrils decreased (p<0.01) to 49.0±5.9% (group 1) and 48.8±3.6% (group 2) late after valve replacement and were reduced (p<0.05) compared with that in
the control patients (Figure 1). Fibrous content decreased in groups 1 and 2 and was significantly ($p<0.001$ and $p<0.01$, respectively) smaller ($12.7±3.2$ g/m² and $18.5±8.9$ g/m², respectively) than before surgery.

In both groups, ejection fraction and $V_{pre}$ did not change (Table 1). Muscle mass index determined by cineangiography elicited a further, although not significant, decrease late after surgery. A slight nonsignificant increase of peak systolic wall stress occurred in both groups late after surgery.

Because patient selection may have influenced the preoperative and the late postoperative comparisons, paired analyses for morphometric variables including fibrous content of the individual patients were performed (Figures 2 and 3). These paired comparisons yielded results similar to the group comparisons presented in Figure 1 except that the decrease of volume fraction of myofibrils in group 2 was not significant with the paired evaluation. To evaluate whether or not morphometric variables can predict an unsatisfactory functional outcome late after surgery, the preoperative data of seven patients with a depressed ejection fraction (<57%) late after surgery or an abnormal isovolumic function diagram (or both), that is, a $dS/dt_{max}$ less than 4,018 dynes×$10^3$/cm²/sec (i.e., smallest value in the 23 control patients) or an end-diastolic circumferential wall stress greater than 56.9 dynes×$10^3$/cm² (i.e., the largest value in the 23 control patients) were compared with those of eight patients whose ejection fraction and isovolumic function diagram were normal late after surgery. Preoperative muscle fiber diameter, percent interstitial fibrosis, volume fraction of myofibrils, and fibrous content were not different between these two groups. However, preoperative ejection fraction was higher (66%, $p<0.01$) in the patients with normal late postoperative contractile function than that (52%) in those with a depressed late postoperative left ventricular functional state. Thus, the best predictor of functional outcome late after surgery was preoperative left ventricular ejection fraction.
TABLE 1. Preoperative and Postoperative Hemodynamic and Angiographic Data

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<th>LVSP (mm Hg)</th>
<th>LVEDP (mm Hg)</th>
<th>Vpms (ML/sec)</th>
<th>EDVI (ml/m²)</th>
<th>hwm (cm)</th>
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<td>Preoperative</td>
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<td>Intermediate postoperative</td>
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<td>138±14</td>
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<td>1.37±0.26</td>
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Patients with aortic insufficiency

|                                |             |               |              |              |         |             |       |                                 |                                 |                                 |
| Preoperative                   | 147±25      | 18.3±8.5      | 1.18±0.23     | 226±49       | 1.01±0.15| 201±49      | 60±9  | 538±129                         | 81.6±36.9                       | 6,803±1,484                     |
| (n=17)                         |              |               |              |              |         |             |       |                                 |                                 |                                 |
| Intermediate postoperative     | 135±16      | 8.8±2.5       | 1.37±0.26     | 128±40       | 0.92±0.18| 131±38      | 65±10 | 430±117                         | 33.3±7.7                       | 6,327±1,888                     |
| (n=25)                         |              |               |              |              |         |             |       |                                 |                                 |                                 |
| Late                           | 138±13      | 11.7±2.0      | 1.33±0.14     | 109±20       | 0.83±0.10| 93±25       | 64±9  | 465±81                          | 47.7±8.6                       | 8,261±2,305                     |
| postoperative                  |              |               |              |              |         |             |       |                                 |                                 |                                 |
| (n=6)                          |              |               |              |              |         |             |       |                                 |                                 |                                 |
| p values                       |              |               |              |              |         |             |       |                                 |                                 |                                 |
| Preoperative vs. intermediate  | NS           | <0.001        | NS            | <0.001       | NS       | <0.001      | NS    | <0.05                           | <0.001                          | NS                              |
| postoperative                  |              |               |              |              |         |             |       |                                 |                                 |                                 |
| Intermediate vs. late          |              | NS            | NS            | <0.001       | NS       | <0.001      | NS    | <0.05                           | NS                              | NS                              |

Controls

|                                | 118±12      | 7.7±2.8       | 1.69±0.35     | 82±16       | 0.79±0.10| 82±17       | 68±7  | 372±53                          | 30.2±12.0                       | 7,102±1,708                     |
|                                |             |               |              |              |         |             |       |                                 |                                 |                                 |

Data are mean±SD.

LVSP, left ventricular peak systolic pressure; LVEDP, left ventricular end-diastolic pressure; Vpms, peak measured velocity of shortening of the contractile elements in muscle lengths (ML)/sec; EDVI, left ventricular end-diastolic volume index; hwm, end-diastolic left ventricular wall thickness; LMMI, left ventricular muscle mass index; EF, biplane left ventricular ejection fraction; S peak, peak systolic circumferential wall stress; Sed, end-diastolic circumferential wall stress; dS/dt max, maximal rate of rise of left ventricular circumferential wall stress.

p values were obtained by one-way analysis of variance.

Discussion

Preoperative Left Ventricular Structure and its Relation to Contractile Function

In the preoperative patients with chronic pressure or volume overload (or both) from aortic stenosis and aortic insufficiency, we have found significant myocardial cellular hypertrophy and an increase of interstitial nonmuscular tissue, which is an index of relative interstitial fibrosis (Figure 1). No differences occurred in these morphometric variables between patients with aortic stenosis and aortic insufficiency. Cellular hypertrophy was assessed from the increased muscle fiber diameter, which is assumed to result from parallel addition of new myofibrils.\(^{34}\) In patients with chronic volume overload, series addition of new sarcomeres also occurs.\(^{34}\) To what extent such hypertrophy in length had occurred in our patients with aortic insufficiency cannot be determined because myocardial cell length was not measured. Muscle fiber diameter correlated weakly inversely with measures of left ventricular contractile function (ejection fraction and peak measured velocity of contractile element shortening), whereas percent interstitial fibrosis did not correlate with any of these variables. Similar observations occurred when mean values of morphometric findings and ejection fraction from previous studies were compared. In Tables 2 and 3, results of previous morphometric studies in patients with aortic stenosis\(^{10-13, 15, 16, 35}\) and aortic insufficiency,\(^{10-14}\) all using glutaraldehyde for fixation, are summarized. Regression analysis from these studies between muscle fiber diameter and ejection fraction yielded a significant inverse correlation (Figure 4), whereas no correlation existed between percent interstitial fibrosis and ejection fraction.
AS before and after AVR

FIGURE 2. Plots of preoperative and postoperative left ventricular morphometric findings in patients with aortic stenosis (AS) who were studied late after valve replacement (AVR). Statistical comparisons were made for paired preoperative and late postoperative data only because the data at the intermediate time were incomplete. Seventy months after aortic valve replacement, no significant change occurred in percent interstitial fibrosis (IF). Muscle fiber diameter (MFD) decreased slightly but not significantly. Volume fraction of myofibrils (VFM) and left ventricular fibrous content (FC) decreased significantly. Shaded areas represent the control values mean±SD obtained from five donor hearts (as in Figure 1).

(Figure 5). Thus, our observations, as well as those derived from previous investigations, indicate that in chronic mechanical overloading massive cellular hypertrophy is, in general, associated with impaired left ventricular contractile function. However, the correlations between muscle fiber diameter and indexes of contractile function are, unsurprisingly, not close because the latter are influenced by a variety of other factors, such as ventricular geometry and loading conditions. Moreover, at a given myocyte size, contractile capacities of the fibers and, hence, ventricular pumping performance may vary by the intracellular myofibrillar density. In the present study, volume fraction of myofibrils in both groups of patients with aortic stenosis and with aortic insufficiency, however, was not different from that in control subjects. Also, no correlation existed between volume fraction of myofibrils and ejection

Al before and after AVR

FIGURE 3. Plots of preoperative and postoperative left ventricular morphometric findings in patients with aortic insufficiency (AI) who were studied late after valve replacement (AVR). Statistical comparisons were made for paired preoperative and late postoperative data only because the data at the intermediate time were incomplete. Sixty-two months after aortic valve replacement, no change in percent interstitial fibrosis (IF) occurred. Muscle fiber diameter (MFD) decreased slightly but significantly. Volume fraction of myofibrils (VFM) also tended to decrease. Left ventricular fibrous content (FC) decreased significantly late after surgery. Shaded areas represent the control values mean±SD obtained from five donor hearts (as in Figure 1).
performance. Other morphologic variables that were not investigated but that may influence ventricular contractile function include isolation of myocardial cells, abnormalities of Z band material, proliferation of sarcoplasmatic reticulum, and increased thickness of basal laminae.36

**Left Ventricular Myocardial Structure and Contractile Function After Aortic Valve Replacement**

**Intermediate after aortic valve replacement.** A sizable decrease in macroscopic hypertrophy assessed by angiographically determined muscle mass was reported by our group8,13,17 and by others.2,3,5,6 When results from several studies were averaged, angiographically determined muscle mass in patients with aortic stenosis decreased by 31% and in patients with aortic insufficiency by 30% 14–15 months after surgery.8 In the present study at 18 months after surgery, the decreases in angiographically determined mass were somewhat more marked (−38% in patients with aortic stenosis and −35% in patients with aortic insufficiency). In both groups, muscle fiber diameter was significantly smaller 18 months after surgery compared with diameters before surgery. This decrease in cellular hypertrophy was associated with an increase in percent interstitial fibrosis, which was significant for the patients with aortic stenosis. Total left ventricular fibrous content remained unchanged in patients with aortic stenosis but decreased in those with aortic insufficiency. Thus, at least in patients with aortic insufficiency, there is evidence of some reversibility of interstitial fibrous tissue intermediate after valve replacement. The decrease of angiographically determined mass in patients with aortic insufficiency, therefore, was mediated by both a reduction of muscle fiber volume and interstitial fibrous content.

**Late after aortic valve replacement.** Recently, we reported that regression of angiographically determined mass after surgical correction of aortic valve disease continues, although more slowly, after the first two postoperative years.17 Morphometric structure was assessed at 70 months after surgery in patients with aortic stenosis and 62 months in patients with aortic insufficiency. Compared with the intermediate term, no further change in muscle fiber diameter occurred, but a reduction did occur in relative interstitial space (percent fibrosis), which was significant for the patients with aortic stenosis. Muscle fiber diameter and percent interstitial fibrosis, however, remained elevated late after aor-

**Table 2. Left Ventricular Morphometric Findings in Patients With Aortic Stenosis**

<table>
<thead>
<tr>
<th>Author</th>
<th>Yr</th>
<th>MFD (µm)</th>
<th>IF (%)</th>
<th>VFM (%)</th>
<th>EF (%)</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Warmuth et al</td>
<td>1978</td>
<td>25.8</td>
<td>62.0</td>
<td></td>
<td></td>
<td>7 (for MFD) resp,</td>
</tr>
<tr>
<td>Schwarz et al</td>
<td>1978</td>
<td>35.1±6.2</td>
<td>15.1±4.5</td>
<td>57</td>
<td>4 AVD</td>
<td></td>
</tr>
<tr>
<td>Nitenberg et al</td>
<td>1979</td>
<td>31±9</td>
<td>39±6</td>
<td></td>
<td>58</td>
<td>6 AS</td>
</tr>
<tr>
<td>Schwarz et al</td>
<td>1981</td>
<td>16.3±5.5</td>
<td>48.4±4.7</td>
<td>65</td>
<td>7 AS</td>
<td></td>
</tr>
<tr>
<td>Schaper et al</td>
<td>1981</td>
<td>23±8</td>
<td>19.5±5.3</td>
<td>44.9±14.3</td>
<td>65</td>
<td>11 AS</td>
</tr>
<tr>
<td>Kunkel et al</td>
<td>1982</td>
<td>46.2±3.4</td>
<td>≥50</td>
<td></td>
<td>59</td>
<td>27 AS</td>
</tr>
<tr>
<td>Hess et al</td>
<td>1984</td>
<td>31±3</td>
<td>15±3</td>
<td></td>
<td>58</td>
<td>10 AS</td>
</tr>
<tr>
<td>Present study</td>
<td></td>
<td>30.9±4.7</td>
<td>18.2±6.2</td>
<td>57.7±5.9</td>
<td>59</td>
<td>27 AS</td>
</tr>
</tbody>
</table>

MFD, muscle fiber diameter; IF, percent interstitial fibrosis; VFM, volume fraction of myofibrils; EF, left ventricular ejection fraction; AVD, aortic valve disease; AS, aortic stenosis.

**Table 3. Left Ventricular Morphometric Findings in Patients With Aortic Insufficiency**

<table>
<thead>
<tr>
<th>Author</th>
<th>Yr</th>
<th>MFD (µm)</th>
<th>IF (%)</th>
<th>VFM (%)</th>
<th>EF (%)</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Schwarz et al</td>
<td>1978</td>
<td>28.8±6.2</td>
<td>16.7±4.8</td>
<td>54</td>
<td>5 resp, 7 AI</td>
<td></td>
</tr>
<tr>
<td>Nitenberg et al</td>
<td>1979</td>
<td>25±4</td>
<td>29±8</td>
<td>55</td>
<td>13 AI</td>
<td></td>
</tr>
<tr>
<td>Schaper12</td>
<td>1981</td>
<td>25±7</td>
<td>16.6±7.4</td>
<td>42.9±16.0</td>
<td>56</td>
<td>8 AI</td>
</tr>
<tr>
<td>Hess et al</td>
<td>1984</td>
<td>31±2</td>
<td>19±4</td>
<td>59</td>
<td>6 AI</td>
<td></td>
</tr>
<tr>
<td>Coulon et al</td>
<td>1986</td>
<td>31.2±2.8</td>
<td>21.0±2.4</td>
<td>normal</td>
<td>15 AI</td>
<td></td>
</tr>
<tr>
<td>Present study</td>
<td></td>
<td>31.4±4.4</td>
<td>20.4±4.9</td>
<td>56.8±4.6</td>
<td>60</td>
<td>17 AI</td>
</tr>
</tbody>
</table>

MFD, muscle fiber diameter; IF, percent interstitial fibrosis; VFM, volume fraction of myofibrils; EF, left ventricular ejection fraction; AI, aortic insufficiency; AS+AI, combined aortic valve lesion.
tic valve replacement compared with the respective data in the control patients (Figure 1). Of particular importance, left ventricular fibrous content late after surgery compared with the intermediate term had decreased significantly in the patients with aortic stenosis. Thus, late postoperative left ventricular fibrous content had decreased significantly in both groups compared with the preoperative value (Figure 1). The specific part of the interstitial tissue that regressed late after surgery could not be determined in this study. Because collagen is the most abundant component of the connective tissue network, the marked decrease of interstitial tissue (g/m²) late after surgery also probably reflected a decrease of collagen content. A complete "restitutio ad integrum", however, did not occur. This allows the postulation that myocardial hypertrophy was truly pathologic in these patients with operative aortic valve disease because in physiologic hypertrophy full reversibility of myocardial structural abnormalities occurs upon cessation of the stimulus to hypertrophy.23

Although one might expect beneficial effects of the late postoperative decrease of fibrous content on left ventricular systolic function, such influences could not be substantiated by our observations. Systolic left ventricular function as evaluated by ejection fraction and measured peak velocity of contractile element shortening was unchanged (Table 1) late after surgery compared with the value before surgery despite the reduction of the fibrous content. Because no correlation occurred at the preoperative evaluation between measures of left ventricular function and percent interstitial fibrosis from either the present investigation or based on the data from previous studies (Figure 5), the extent of interstitial fibrosis was probably insufficient to cause, by itself, a derangement of left ventricular systolic function. Alternatively, the decrease of left ventricular fibrous content might have influenced the pas-

**Figure 4.** Relation between left ventricular ejection fraction (EF) and muscle fiber diameter (MFD) based on mean values from previous studies. A significant inverse correlation occurred between ejection performance and degree of cellular hypertrophy. Individual data for patients with aortic stenosis and aortic insufficiency were taken from Tables 2 and 3. The only control data point was obtained from Schaper et al.12

**Figure 5.** Relation between left ventricular ejection fraction (EF) and percent interstitial fibrosis (IF) based on mean values from previous studies. No significant correlation occurred between the two variables. Individual data points for patients with aortic stenosis and aortic insufficiency were taken from Tables 2 and 3. The one control data point was obtained from Schaper et al.12
sive diastolic properties of the left ventricle. In a previous investigation, left ventricular diastolic myocardial stiffness constant ($\beta$) was curvilinearly related to the ratio of fibrous content (FC)/end-diastolic volume index (EDVI). Up to a ratio of FC/EDVI $\leq$ 0.20 g/ml, $\beta$ was normal. In six patients with aortic insufficiency, FC/EDVI did not change from the preoperative to the late postoperative study (0.17 and 0.17 g/ml, respectively), whereas in nine patients with aortic stenosis, FC/EDVI decreased from 0.22 to 0.14 g/ml ($p < 0.025$). Before surgery, FC/EDVI exceeded 0.20 g/ml in five of these nine patients with aortic stenosis, and late after aortic valve replacement, FC/EDVI exceeded 0.20 g/ml in only one of the nine patients. Hence, in patients with aortic stenosis, indirect evidence exists for improvement of preoperatively increased left ventricular diastolic myocardial stiffness late after aortic valve replacement.

The late postoperative reduction of volume fraction of myofibrils in both groups (Figure 1) was an unexpected finding. This decrease may be due to either intracellular myofibrillar lysis or decreased synthesis of myofibrils or a combination of the two mechanisms. In any case, the decrease of volume fraction of myofibrils appeared not to be the morphologic correlate of an impairment of cardiac function because ejection fraction, peak measured velocity of contractile element shortening, and maximal rate of rise of circumferential wall stress remained essentially unchanged late after aortic valve replacement (Table 1). One may speculate whether or not the decrease of volume fraction of myofibrils was a late effect of the surgical correction of the abnormal left ventricular loading conditions. Peak systolic circumferential wall stress was, indeed, lower, though not significantly, in both groups late after surgery compared with values before surgery (Table 1). However, true systolic unloading of the myofibrils would only have taken place if the percentage of the myofibrils per unit cross-sectional area of the myocardium would have been constant. This latter condition was certainly not met because the percentage of nonmuscular tissue and the percentage of the myofibrils within the intramyocyte space varied from the preoperative to the intermediate and late postoperative study. As a matter of fact, calculation of myofibrillar peak systolic circumferential wall stress revealed only small and insignificant differences for patients with aortic stenosis (1,057 x 953 x 1,028 x 10$^3$ dynes/cm$^2$ myofibrillar area) and for those with aortic insufficiency (1,197 x 1,057 x 1,185 x 10$^3$ dynes/cm$^2$ myofibrillar area) at the preoperative, intermediate, and late postoperative investigations. All these values seem larger than in normal subjects. By combining the morphometric data of the five donor control patients (interstitial nonmuscular tissue 7% and volume fraction of myofibrils 57.2%) with the average myocardial peak systolic circumferential wall stress of 372 x 10$^3$ dynes/cm$^2$ of the 23 hemodynamic control patients, an approximate normal myofibrillar peak systolic circumferential wall stress of 700 x 10$^3$ dynes/cm$^2$ myofibrillar area is obtained. Thus, in patients with aortic stenosis and aortic insufficiency, continuing systolic overloading occurs at the myofibrillar level even late after surgery with myofibrillar peak systolic stress being 47–69% above that in control subjects. This excess systolic loading may interfere with myofibrillar shortening and ultimately be the cause why systolic ejection fraction did not increase in these patients after valve replacement.

The preoperative morphometric findings were not useful for predicting the late postoperative functional outcome. Similarly, in patients with dilated cardiomyopathy, muscle fiber diameter and percent nonmuscular interstitial tissue have also not been predictive of the long-term course of hemodynamic function. In contrast to the morphometric findings, preoperative left ventricular function was important for late postoperative functional outcome because patients with depressed late postoperative contractile function had a significantly smaller preoperative ejection fraction than those with a normal postoperative left ventricular functional state.

**Limitations**

The number of biopsy specimens obtained in a given patient per cardiac catheterization was 2–3 in the present study. In studies of patients with cardiomyopathies, Baandrup et al. concluded that at least five biopsy samples are necessary to establish whether or not correlations between structural changes and the functional state of the heart exist. In comparison, the number of biopsy samples we obtained appears suboptimal. However, in patients with aortic valve disease, morphologic changes in the myocardium appear more uniformly distributed over the whole ventricle and hence, 2–3 biopsy samples should reflect adequately left ventricular endomyocardial structure in this setting.

Fibrous content was calculated as IF x LMMI/100. Interstitial fibrosis (IF) determined from endomyocardial biopsy samples was assumed to be representative for the entire left ventricular wall. Morphometric studies from transmural biopsies obtained at surgery, however, have shown that in patients with predominant left ventricular pressure overload interstitial fibrosis is significantly higher in the subendocardial compared with the subepicardial region (19% vs. 13%). Thus, the magnitude of fibrous content probably was overestimated in the patients with aortic stenosis. This systematic overestimation, however, should not have invalidated comparisons between the preoperative and postoperative investigations.

For most patients with preoperative and postoperative comparisons, no coronary artery disease was assumed to develop between the preoperative and the postoperative catheterizations. Among the 38 patients restudied intermediate after surgery, 16 had a coronary arteriogram. The results from 15 were normal, and in one patient, the same 50% stenosis of the left anterior descending coronary artery was visualized as...
on the preoperative coronary arteriogram. Coronary arteriography was performed late after surgery in only one of the 15 patients: the coronary arteries in this patient were normal. Although none of the patients who were restudied complained of anginal pain after aortic valve replacement, newly developed coronary artery disease was not excluded in 27 of the 44 patients. Because no regional wall motion abnormalities were detected on postoperative cineangiograms and because percent interstitial nonmuscular tissue was the same late after surgery as before surgery, significant asymptomatic ischemia probably did not develop in these patients.

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References


KEY WORDS • myocardial structure • left ventricular hypertrophy • interstitial fibrosis • regression of muscle mass
Left ventricular myocardial structure in aortic valve disease before, intermediate, and late after aortic valve replacement.

H P Krayenbuehl, O M Hess, E S Monrad, J Schneider, G Mall and M Turina

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