Myocardial structure as a determinant of pre- and postoperative ventricular function and long-term prognosis after valve replacement for aortic stenosis


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Background Long-term results after aortic valve replacement for aortic stenosis can be correlated to a cardiaclrelated pre-operative risk profile. This predictability indicates that there is a common basis in subtle or overt structural abnormalities of left ventricular myocardium.

Methods and Results Forty-nine patients aged 24–82 (mean 61) years, with aortic stenosis had a full wall thickness transmural biopsy of the left ventricular antero-lateral free wall during aortic valve replacement. Echocardiography and radionuclide ventriculography were performed prior to, and 18 months (n=41) after, the operation. Postoperative follow-up to a maximum of 7.7 years was 100% complete. Pre-operatively, all patients had an increase in both the left ventricular mass index (202 ± 67 g. m⁻²) and the muscle cell diameter (41 ± 8 μm); other morphological data included a muscle cell nucleus volume of 752 ± 192 μm³, a muscle cell mass index of 163 ± 54 g. m⁻², and a fibrous tissue mass index of 39 ± 16 g. m⁻². Patients with a pre-operative episode of clinical left ventricular failure (n=19) had significantly greater morphological variables than those without. Preoperative ejection fraction and other measures of systolic function correlated inversely with the morphological data, except for the fibrous tissue mass index; diastolic function indices correlated inversely with all the morphological variables. At the 18-month re-study, the same general picture was noted, but with an underlying strengthening, especially of the muscle cell mass index. Overall, the mass index dropped to 152 ± 51 g. m⁻² (P <0.0001), but in 17% of the patients it became normal; the mass index at 18 months was directly correlated to morphological variables. A high muscle cell nucleus volume was identified as an independent predictor of early and late mortality.

Conclusions Abnormalities of the hypertrophied left ventricular muscle cell and the degree of muscle hypertrophy are, to some degree, underlying determinants of preoperative symptomatology, pre- and postoperative ventricular function, and early and late mortality after valve replacement for aortic stenosis. Incomplete hypertrophy regression after valve replacement, being indicative of impaired results, was related to preoperative myocardial structural abnormalities.

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Key Words: Aortic stenosis, myocardial histo-pathology, aortic valve replacement, systolic function, diastolic function, prognosis.

Introduction

Previous studies have indicated that long-term results after aortic valve replacement for aortic stenosis are correlated to patients’ pre-operative risk profile[1–4]. Prognostic indexes derived from the risk profile have primarily included factors related to advanced heart disease in general and impairment of left ventricular function in particular[1–4]. It is thus reasonable to hypothesize that the pre-operative status, risk profile and associated postoperative prognoses of such patients have a basis in subtle or overt pre-operative myocardial structural abnormalities. Further research should examine the relationship between myocardial structure and pre- and postoperative left ventricular function and long-term prognosis after surgery.

The aim of the present study was to relate myocardial structure, based on intra-operative full wall thickness left ventricular transmural biopsies, to pre-operative ventricular function as well as to postoperative
function and long-term prognosis after aortic valve replacement. The study includes 49 prospectively enrolled patients with aortic stenosis who had left ventricular function re-examined 18 months postoperatively and who were followed for a minimum of 6-4 years after surgery.

Methods
A total of 49 patients gave written informed consent to participate in the present prospective study, which was approved by the Ethical Committee of Aarhus County. Aiming to recruit 50 patients, 54 consecutive patients, who eventually underwent bivalve replacement for aortic stenosis during August 1988 to November 1989, were asked for their involvement: four declined and one was later excluded when re-evaluation of the aortic root angiogram showed significant aortic regurgitation within one year prior to the operation, and secondary kidney failure as increased S-creatinine (>110 µmol l⁻¹) in two successive pre-operative blood samples. ECG hypertrophy and strain score (0-12) was calculated according to Romhilt and Estes. Invasive data

Left ventricular peak systolic pressure, end-diastolic pressure, and peak-to-peak aortic valve gradient were recorded (mmHg), in all except six patients in whom the left ventricle could not be entered retrogradely. In a consecutive series of 100 patients (including those of the present report) with aortic stenosis awaiting operation, we noted a high correlation between peak-to-peak gradient and peak instantaneous Doppler gradient (PDG): PPG = 24 + 0.69 PDG (r = 0.68, P < 0.001) (unpublished material). In order to have an estimate of peak-to-peak gradient in all patients, it was calculated from the Doppler gradient using the above formula in the six without a measured value; left ventricular peak systolic pressure was calculated as estimated peak-to-peak gradient plus peak systolic aortic pressure measured during catheterization. Coronary artery disease was defined as luminal diameter reduction of at least 50% of a major epicardial vessel or a first branch.

Echocardiographic data

The echocardiographies were performed by one of two experienced observers. Left ventricular variables were measured from the M-mode tracings (end-diastolic and end-systolic diameter (EDD; ESD), posterior free wall thickness (EDPWT; ESPWTh), and interventricular septal thickness (EDSWTh; ESSWTh). End-diastolic and end-systolic radius-wall thickness ratio (RTh) were calculated as EDD/(EDPWT + EDSWTh) and ESD/(ESPWTh + ESSWTh), respectively. Left ventricular mass (g) was calculated as 1.04 [(EDD + EDPWT + EDSWTh)³ – EDD³] – 14, while division by body surface area gave mass index (g m⁻²). Diastolic and systolic wall stress were calculated as end-diastolic RTh, end-diastolic pressure, 1333, and end-systolic RTh . LV peak systolic.
pressure, 1333 (dynes cm$^{-2}$), respectively\cite{16}, and fractional shortening as 100. (EDD - ESD)/EDD (%). A value of 108 g. m$^{-2}$ was used as the upper normal level for the left ventricular mass index\cite{5,15}, and a value of 2.50 as the lower normal level for end-diastolic $R \bar{c}T_h$\cite{17}.

Radionuclide cardiography

Our methods have been described and validated in detail previously\cite{19-22}. Right ventricular ejection fraction and mean pulmonary transit time were calculated from the first-pass study\cite{11,12}. The remaining left ventricular function indices were drawn from the ECG-gated equilibrium study\cite{12}, ejection fraction, peak ejection rate normalized for end-diastolic volume (EDV $\cdot$ s$^{-1}$), time to peak ejection (from ejection start)-systole duration ratio (an index of prolonged contraction), systole duration-heart cycle (duration of heart cycle) ratio (an index of prolonged contraction at the expense of diastolic filling time), peak filling rate in the first half of diastole (EDV $\cdot$ s$^{-1}$), fast filling fraction (filling during the first half of diastole as a percent of total filling volume), PQ filling fraction (filling during the ECG PQ interval, atrial contraction, in late diastole as a percent of total filling volume; the PQ interval was arbitrarily set at 190 ms in two patients with atrial flutter/fibrillation), time to peak filling (also if in the last half of diastole) – diastole duration ratio (an index of reduced and delayed early fast filling), and end-diastolic volume index (ml . m$^{-2}$). Lower or upper 95% confidence limits from a reference study of healthy individuals\cite{12} were used to identify patients with subnormal or supranormal (as appropriate) function indices as detailed in Appendix 1.

Left ventricular transmural biopsy

The endo- and epicardial half was designated according to the biopsy’s position on the millipore paper. The biopsy was placed in a groove and plastic embedded (glycol methacrylate, Technovit$^{\text{\textregistered}}$ 7100). Two micron-thick sections were cut and haematoxylin-eosin, toluidine blue, and van Gieson staining reactions undertaken. The relative volume of myocardial muscle and interstitial (fibrous) tissue was determined by point counting using a 10 $\times$ 10 grid at a total magnification of 375 and according to standard methods\cite{18}. Percent fibrous tissue was calculated as the average of a minimum of eight (mean 12) areas and muscle cell transverse diameter ($\mu$m) as the average of 50 measurements from each (epicardial and endocardial) half of the biopsy and subsequently by calculation for the full biopsy. The myocardial cell is known to represent a near cylinder and therefore measurements of the myocyte diameter were restricted to the nuclear areas which occupy the centre of the cells; the smallest diameter was chosen as the one representing the actual diameter\cite{19}. Fibrous tissue mass index (g . m$^{-2}$) was calculated as percent fibrous tissue. Left ventricular mass index/100 and muscle cell mass index (g . m$^{-2}$) as mass index – fibrous tissue mass index. Mean nucleus volume ($\mu$m$^3$) of myocardial muscle cells was estimated by point sampling of nuclear intercepts\cite{18} using all available sections of each half biopsy and of the full biopsy. A muscle cell diameter of 10–15 $\mu$m was considered normal\cite{19}.

Follow-up and statistical analysis

In addition to the 18-month investigation mentioned previously, the patients were all seen 3 months postoperatively and thereafter annually in the outpatient clinic. A 100% complete follow-up involving telephone contact to the patients who were alive and to the general practitioner if the patient had died was conducted during May 1996. A total of 13 patients had died; death certificates were available in all and autopsy reports in four. A total of 515 patient-years at risk had been accumulated with a minimum and maximum observation time of 6.4 and 7.7 years, respectively. All statistical tests were computerized using the BMDP Dynamic release 7-0 software package\cite{20}. Univariate comparisons between groups were done using a standard Pearson chi-square test, a non-paired t-test, a paired t-test, or a one-way analysis of variance as appropriate. Linear relations were checked with a standard least-squares linear regression analysis. Cumulative survival was estimated using Kaplan and Meier’s product-limit method and differences between survival curves tested with a log-rank test and a Gehan test. The Cox regression analysis in a step-wise and formalized test sequence\cite{2-4} was used to identify independent predictors of mortality. Quantitative data are given with ± one standard deviation and survival estimates with ± one standard error. The level of statistical significance was set at 0.05.

Results

The pre-operative clinical, paraclinical, and haemodynamic profile of the patients are given in Tables 1 and 2. The majority of the patients (61%) were men and 59% (n=29) were 60 years or older. All had left ventricular hypertrophy with a mass index >108 g . m$^{-2}$, while only 36% (n=18) had a dilated left ventricular chamber with an end-diastolic volume index >89 ml . m$^{-2}$. The patients had a concentric hypertrophic chamber geometry with a low end-diastolic radius–wall thickness ratio; only one patient had a value >2.50.

A total of 47% (n=23) of the patients had an impaired left ventricular systolic function with a subnormal ejection fraction (n=21), a subnormal peak ejection rate (n=7), or a supranormal time to peak ejection – systole duration ratio (n=5), while 67% (n=33) had impaired diastolic function with a subnormal peak filling rate (n=14), a subnormal fast filling fraction (n=23), a supranormal time to peak filling – diastole duration ratio (n=16), or a supranormal PQ filling fraction (n=25). Systole duration–heart cycle ratio was supranormal in 20 patients, all with impaired systolic or diastolic function.
Examination of the full left ventricular transmural needle biopsies showed a muscle cell diameter of $41 \pm 8$ (range 27–65) μm, a nucleus volume of $752 \pm 192$ (range 283–1224) μm$^3$, a percent fibrosis of $19 \pm 5$ (range 11–34), a muscle cell mass index of $163 \pm 54$ (range 81–309) g. m$^{-2}$, and a fibrous tissue mass index of $39 \pm 16$ (range 16–76) g. m$^{-2}$. The two biopsy halves differed only as regards percent fibrosis (epi- vs endocardial: $18 \pm 6$ vs $21 \pm 6\%$, P < 0.01).

Table 1  Pre-operative clinical, paraclinical, and invasive data (n=49)

<table>
<thead>
<tr>
<th></th>
<th>Before (n=49)</th>
<th>After (n=49)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>61 ± 14, 24–82</td>
<td>64 ± 16, 20–82</td>
</tr>
<tr>
<td>Male gender</td>
<td>61% (30)</td>
<td>62% (25)</td>
</tr>
<tr>
<td>NYHA class</td>
<td>I</td>
<td>II</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>27% (13)</td>
</tr>
<tr>
<td></td>
<td>II</td>
<td>61% (30)</td>
</tr>
<tr>
<td></td>
<td>IV</td>
<td>12% (6)</td>
</tr>
<tr>
<td>Left ventricular failure</td>
<td>39% (19)</td>
<td>39% (19)</td>
</tr>
<tr>
<td>Secondary kidney failure</td>
<td>18% (9)</td>
<td>19% (9)</td>
</tr>
<tr>
<td>Atrial flutter/fibrillation</td>
<td>4% (2)</td>
<td>5% (2)</td>
</tr>
<tr>
<td>Ventricular ectopic beats</td>
<td>10% (5)</td>
<td>10% (5)</td>
</tr>
<tr>
<td>AV conduction block grade</td>
<td>1†</td>
<td>10% (5)</td>
</tr>
<tr>
<td>Left bundle branch block†</td>
<td>4% (2)</td>
<td>4% (2)</td>
</tr>
<tr>
<td>ECG hypertrophy and strain score</td>
<td>$7.9 \pm 2.3$, 4-12</td>
<td>$8.0 \pm 2.3$, 4-12</td>
</tr>
<tr>
<td>Cardiac output index</td>
<td>0.52 ± 0.04, 0.43–0.59</td>
<td>0.53 ± 0.04, 0.43–0.59</td>
</tr>
<tr>
<td>Aortic valve gradient (mmHg)</td>
<td>84 ± 24, 40–140</td>
<td>85 ± 24, 40–140</td>
</tr>
<tr>
<td>LV systolic pressure (mmHg)</td>
<td>215 ± 26, 162–290</td>
<td>217 ± 26, 162–290</td>
</tr>
<tr>
<td>LV end-diastolic pressure (mmHg)</td>
<td>$24 \pm 9$, 8–45</td>
<td>$25 \pm 9$, 8–45</td>
</tr>
<tr>
<td>LV systolic wall stress (10$^3$ dynes . cm$^{-2}$)</td>
<td>$255 \pm 73$, 146–423</td>
<td>$260 \pm 73$, 146–423</td>
</tr>
<tr>
<td>LV diastolic wall stress (10$^3$ dynes . cm$^{-2}$)</td>
<td>$65 \pm 30$, 22–137</td>
<td>$68 \pm 30$, 22–137</td>
</tr>
<tr>
<td>Coronary artery disease</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>65% (32)</td>
<td>59% (30)</td>
</tr>
<tr>
<td>1-vessel</td>
<td>12% (6)</td>
<td>12% (6)</td>
</tr>
<tr>
<td>2-vessel</td>
<td>12% (6)</td>
<td>12% (6)</td>
</tr>
<tr>
<td>3-vessel or left main stem</td>
<td>11% (5)</td>
<td>11% (5)</td>
</tr>
</tbody>
</table>

Data are mean ± standard deviation, range, or % (n). See text for definitions and calculations. NYHA = New York Heart Association; AV = atrioventricular; LV = left ventricular; tno-one with grade 2 or 3; †no-one with right bundle branch block; §n=43.

Table 2  Radionuclide cardiography and echocardiography data before and 18 months after the operation

<table>
<thead>
<tr>
<th>Radionuclide cardiography</th>
<th>Before (n=49)</th>
<th>After (n=49)</th>
</tr>
</thead>
<tbody>
<tr>
<td>LV ejection fraction (%)*</td>
<td>$59 \pm 15$, 19–80</td>
<td>$64 \pm 16$, 20–82</td>
</tr>
<tr>
<td>LV peak ejection rate (EDV . s$^{-1}$)**</td>
<td>$3.22 \pm 0.92$, 1.38–5.55</td>
<td>$3.90 \pm 1.14$, 1.30–6.77</td>
</tr>
<tr>
<td>LV time to peak ejection-systole duration ratio</td>
<td>$0.54 \pm 0.12$, 0.23–2.90</td>
<td>$0.54 \pm 0.13$, 0.34–0.87</td>
</tr>
<tr>
<td>LV systole duration-heart cycle ratio</td>
<td>$0.49 \pm 0.07$, 0.24–0.61</td>
<td>$0.46 \pm 0.07$, 0.34–0.68</td>
</tr>
<tr>
<td>LV peak filling rate (EDV . s$^{-1}$)</td>
<td>$2.77 \pm 0.95$, 1.28–5.59</td>
<td>$2.96 \pm 1.09$, 1.35–7.00</td>
</tr>
<tr>
<td>LV time to peak filling-diastole duration ratio</td>
<td>$0.47 \pm 0.23$, 0.15–0.97</td>
<td>$0.53 \pm 0.23$, 0.13–0.97</td>
</tr>
<tr>
<td>LV fast filling fraction (%)</td>
<td>$59 \pm 13$, 31–87</td>
<td>$56 \pm 15$, 28–90</td>
</tr>
<tr>
<td>LV PQ filling fraction (%)</td>
<td>$40 \pm 21$, 12–95</td>
<td>$39 \pm 19$, 12–90</td>
</tr>
<tr>
<td>LV end-diastolic volume index (ml . m$^{-2}$)*</td>
<td>$86 \pm 30$, 48–203</td>
<td>$76 \pm 34$, 40–177</td>
</tr>
<tr>
<td>RV ejection fraction (%)</td>
<td>$58 \pm 10$, 28–75</td>
<td>$56 \pm 8$, 38–74</td>
</tr>
<tr>
<td>Pulmonary transit time (s)**</td>
<td>$11.2 \pm 3.5$, 6–7.84</td>
<td>$9.4 \pm 2.4$, 5.1–15.0</td>
</tr>
</tbody>
</table>

Data are mean ± standard deviation, range. LV = left ventricular; RV = right ventricular. See text for calculations and definitions. Paired t-test: ****P < 0.0001; ***P < 0.01; **P < 0.05.

Examination of the full left ventricular transmural needle biopsies showed a muscle cell diameter of $41 \pm 8$ (range 27–65) μm, a nucleus volume of $752 \pm 192$ (range 283–1224) μm$^3$, a percent fibrosis of $19 \pm 5$ (range 11–34), a muscle cell mass index of $163 \pm 54$ (range 81–309) g. m$^{-2}$, and a fibrous tissue mass index of $39 \pm 16$ (range 16–76) g. m$^{-2}$. The two biopsy halves differed only as regards percent fibrosis (epi- vs endocardial: $18 \pm 6$ vs $21 \pm 6\%$, P < 0.01).
Relationship between myocardial structure and pre-operative data

The relationships were examined for the values of the entire biopsy. There was no relationship between coronary artery disease, sex or age and any of the biopsy variables. Ten patients with significant stenoses of the vessels (left main stem, first two of three segments of the left anterior descending artery, first and second diagonal branch) supplying the biopsy area and 39 without did not differ from one another as regards any of the biopsy variables. The biopsy variables were unrelated to aortic anulus size (outer diameter of prosthetic valve).

Six patients in NYHA functional class IV had greater nucleus volume than those in classes II–III (n=43; 896 ± 209 vs 732 ± 184 μm³, P <0.05), while 13 in class II had a lower percent of fibrosis (17 ± 4 vs 20 ± 5%, P <0.05) and a lower fibrous tissue mass index (31 ± 16 vs 42 ± 15 g.m⁻², P <0.05) than those in classes III–IV (n=36). Nine patients with secondary kidney failure had greater nucleus volume (917 ± 194 vs 715 ± 174 μm³, P <0.01) and percent fibrosis (23 ± 6 vs 18 ± 5%, P <0.01) than those without (n=40). Eleven patients with conduction or rhythm disturbances (atrioventricular conduction block, bundle branch block, or atrial flutter or fibrillation; Table 1) had a greater muscle cell mass index (211 ± 71 vs 149 ± 40 g.m⁻², P <0.05) and left ventricular mass index (257 ± 83 vs 186 ± 50 g.m⁻², P <0.005) than those without (n=38). Clinical left ventricular failure was significantly related to all the biopsy variables (Fig. 1). Tables 3 and 4 show the significant correlations between myocardial structure and pre-operative quantitative variables. Generally, high nucleus volume, muscle cell mass index, and fibrous tissue mass index were related to indices indicating advanced disease and impaired left ventricular function. Aortic valve gradient, and systolic or diastolic wall stress did not correlate with any of the biopsy variables, neither did time to peak ejection–systole duration ratio, peak filling rate, or fast filling fraction.

The 23 patients with impairment of left ventricular systolic function had a greater nucleus volume (819 ± 181 vs 693 ± 186 μm³, P <0.05) than the 26 with normal function. The patients with reduced diastolic function (n=33) had a greater muscle cell mass index (173 ± 61 vs 142 ± 28 g.m⁻², P <0.05) and left ventricular mass index (215 ± 70 vs 177 ± 43 g.m⁻², P <0.05) than those without (n=16).

Relationship between myocardial structure and ventricular function 18 months postoperatively

Ventricular function and mass 18 months postoperatively in the 41 patients who attended the investigation are given in Table 2. Left ventricular ejection fraction, peak ejection rate, and end-diastolic radius-wall thickness ratio had increased while end-diastolic volume, mass index, and pulmonary transit time had dropped significantly. Only seven patients (17%) had

Figure 1 Left ventricular myocardial structure in relation to clinical left ventricular failure (LVF) within a year before the operation. Cell diam = muscle cell diameter; Nucl vol = muscle cell nucleus volume. % FT = percent fibrous tissue; LVFTMi = left ventricular fibrous tissue mass index; LVMCMi = left ventricular muscle cell mass index. The bars represent mean values and the flags one standard error. See text for definitions and calculations and Table 3.
a normal mass index while 30 (73%) had a normal end-diastolic volume index; 26 patients were asymptomatic and the remaining 15 were in NYHA class II. The cardiothoracic index had dropped significantly to 0.49 ± 0.05 (P < 0.001) and ECG hypertrophy and strain score to 3.86 ± 2.50 (P < 0.0001; compare with Table 1). There were no relationships between these variables and the biopsy variables, except between cardiothoracic index and muscle cell mass index (r = 0.36, P < 0.05). Left ventricular mass and mass reduction (pre-operative minus 18-month values) were unrelated to peak Doppler gradient, the size (outer diameter) and orifice diameter of the prosthetic valve, and valve type. The correlations between myocardial structure and ventricular function at the 18-month investigation are shown in Table 5. The picture is generally the same as that for the relationship with pre-operative ventricular function (Table 4). Left ventricular mass index at 18 months was related to muscle cell diameter (r = 0.33, P < 0.05), nucleus volume (r = 0.36, P < 0.05), percent fibrosis (r = 0.31, P < 0.05), muscle cell mass index (r = 0.33, P < 0.05), and fibrous tissue mass index (r = 0.42, P < 0.05).

Figure 2 Cumulative survival after the operation in relation to muscle cell nucleus volume. Five- and 7-year survivals were 83 ± 7% and 83 ± 7%, respectively, for a nucleus volume of ≤ 820 μm³ (—–), and 74 ± 10% and 49 ± 14%, respectively, for a nucleus volume of > 820 μm³ (- - -).

Discussion

Previous investigations into the relationship between myocardial structure and left ventricular function in aortic stenosis were conducted more than 10 years ago by Schwarz et al., Raynebuehl et al., Oldershaw et al., and Baandrup et al., among others. Schwarz's group used transmural needle biopsies of the left ventricular free wall, while endomyocardial biopsies were performed in the remaining studies. The transmural biopsy studies showed that there was a relationship between the degree of abnormality of the myocyte and both the degree of left ventricular hypertrophy and impaired diastolic function, but only indirectly with impaired contractility. Using endomyocardial biopsies, on the other hand, the amount of interstitial fibrosis of the myocardium seemed to play a dominant role, both as regards reduced systolic and diastolic function. The latter results are, however, probably of limited value. The results were based on 2–3 biopsies, but Baandrup et al. have shown that at least five are needed due to extreme variability between the individual biopsies. Furthermore, the subendocardial portion of the left ventricular wall is subjected to more ischaemic injury than the other portions of the wall in concentric hypertrophy, indicating that the endomyocardium is not representative of functioning left ventricular myocardi- um. Schwarz et al. divided their transmural biopsies into three portions and showed that the percentage of fibrous tissue dropped successively and by a factor of two from subendocardial to a subepicardial third. They were unable to relate the average fibrous content of the entire transmural biopsy to ventricular function. The small number of patients with aortic stenosis (≤ 10) in these studies is probably another explanation for the conflicting results.

The present patient group was, unlike those of the above cited studies, of sufficient size as to be representative of the spectrum of adult patients in the western world today who come to operation for aortic stenosis. As in other recently published series, more than 70% of our patients had advanced disease, with NYHA functional class III or IV status, and an average age of more than 60 years. All our patients had significant concentric pre-operative hypertrophy and in all the myocardial muscle cell diameters had increased. The value of the present series is, furthermore, that the spectrum covers patients from minimal symptomatology (NYHA class II) to those with end-stage disease, and from normal to severely impaired left ventricular function.

The present study showed a clear connection between the clinical-paraclinical consequences of aortic stenosis and myocardial structure. Some of the present r-values were low but statistically significant. The main strength, however, was that all correlations pointed in the same general direction. A dvanced symptomatology, secondary kidney failure, and cardiac ectasia have been shown to be independent determinants of poor results after aortic valve replacement, and these variables were related to abnormalities of the myocyte (high nucleus volume) but also to the amount of left ventricular fibrous tissue. Most importantly, a pre-operative episode of clinical left ventricular failure was significantly related to the entire spectrum of left ventricular myocardial structural abnormalities. Clinical left ventricular failure has been shown to be a decisive predictor of both early and late mortality after surgery. The pre-operative risk profile of the patients and the postoperative prognosis it predicts is thus related to structural abnormalities of the left ventricular myocyte.

It is important to emphasize that associated coronary artery disease did not influence the present histopathological biopsy findings. Accordingly, both Wiger and Lund and Larsen have demonstrated that left ventricular microscopic and macroscopic scarring were more related to degree of hypertrophy than to associated coronary artery disease.
A direct relationship between left ventricular myocardial structural abnormalities and impaired contractility has not been shown previously. In the present study, both reduced left ventricular ejection fraction and the increased systole duration-heart cycle ratio were related to myocyte abnormalities (high nucleus volume), to pure muscle hypertrophy itself (high muscle cell mass index), and also to the increased amount of ventricular fibrous tissue. However, the relationship with muscle cell mass was generally stronger than that with fibrous tissue mass, and reduced peak ejection rate was exclusively related to the degree of muscle hypertrophy. Schwarz et al.\[22\] found an inverse relationship between left ventricular ejection fraction and mass, but unlike the present study, no relationship with the amount of fibrous tissue. However, their transmural biopsy study only included five patients with aortic stenosis\[22\].

An influence of myocardial structure on left ventricular diastolic function was indicated by the present direct relationship between muscle cell diameter and both end-diastolic pressure and volume index. However, our more direct measure of diastolic function, time to peak filling-diastole duration ratio, was related only to the amount of ventricular fibrous tissue. A normally increased filling fraction in late diastole during atrial contraction (PQ interval), compensating for reduced early filling, was, furthermore, directly related to increased myocyte nucleus volume, and also to both fibrous tissue mass index and muscle cell mass index. The present composite variables of overall left ventricular systolic and diastolic function were, finally, adversely influenced only by increased muscle cell nucleus volume and muscle cell mass index, respectively. The present results thus give a more differentiated insight into the significance of myocardial structural abnormalities in aortic stenosis than previous studies: abnormalities of the hypertrophied muscle cell and muscle cell mass are important determinants of left ventricular functional impairment while the amount of ventricular fibrous tissue plays a minor role.

The relationships between myocardial structure and ventricular function 18 months after the operation gave important supplementary information. Generally, the relationships were unaltered, with nucleus volume and muscle cell mass index being the foremost determinants. The adverse influence of muscle cell mass index was, if anything, stronger, especially as regards diastolic function at 18 months. Pulmonary transit time at the 18-month investigation was, furthermore, strongly related to left ventricular muscle cell mass index and not to fibrous tissue mass index. We have previously shown that pulmonary transit time in normal volunteers is related to left ventricular diastolic function (suction) and not to right ventricular ejection fraction\[122\]. Right ventricular ejection fraction of the present patients 18 months after the operation was, furthermore, directly related to both left ventricular muscle cell diameter and nucleus volume. Overall, these findings indicate that irreversible damage to the hypertrophied myocyte is an important factor. It seems that the adverse influence on left ventricular function was maintained 18 months after the operation with associated increased pulmonary transit time due to reduced left ventricular diastolic function and increased load, presumably with secondary hypertrophy and ‘boosting’ of right ventricular function.

The above results point to the importance of regression in left ventricular hypertrophy after aortic valve replacement. It has been proposed that complete hypertrophy regression is a primary aim of valve replacement in aortic stenosis, since this hypothetically is the underlying factor securing the patients a sex- and age-specific normal survival and a normal ventricular function after surgery\[41\]. In the present study, significant hypertrophy regression did take place: the left ventricular mass index dropped from an average of 202 g. m\(^{-2}\) pre-operatively to 152 g. m\(^{-2}\) at the 18-month investigation when only 17% had a normal ventricular mass. The mass index 18 months after the operation was significantly related to both muscle cell diameter, nucleus volume, muscle cell mass index, and fibrous tissue mass index from the intra-operative transmural biopsies. The degree of hypertrophy regression after removal of the hypertrophy trigger thus seems to be predetermined by presumably irreversible changes of the hypertrophied myocyte in many patients. This hypothesis was further strengthened by the observation that hypertrophy regression was not related to prosthetic valve size, type, or Doppler gradient. Residual hypertrophy was, furthermore, related to significant ventricular fibrosis. The latter is related to end-stage disease in aortic stenosis\[35,36\].

So far, the present results have hinted that the hypertrophied myocyte may irreversibly damaged prior to valve replacement, which may cause postoperative mortality. The damage may be subtle but may prevent the myocyte from returning to its normal size. The present patient group and its follow-up were of sufficient quantity to allow a direct multivariate prognostic analysis: myocyte nucleus volume was identified as an independent risk factor which seemed to maintain its influence on mortality throughout follow-up (Fig. 2). Nucleus volume thus seemed to be among the most useful of the left ventricular transmural biopsy variables: it was related both to clinical–paraclinical status and ventricular function pre-operatively, to ventricular function and mass 18 months after the operation, and to early and late prognosis after valve replacement.

Increase in nucleus size is a general feature of hypertrophy, but we have not measured the DNA content of myocytic nuclei. The changes we have seen do not resemble (nucleus) oedema, and by combining morphological and functional parameters of the present study we find there is ample evidence that the observed morphological changes represent genuine functional alterations.

We conclude that abnormalities of the left ventricular myocyte in aortic stenosis involve a nucleus as well as large myocyte hypertrophy and are underlying determinants of advanced symptomatology, paraclinical consequences of aortic stenosis, and pre- and
postoperative ventricular function, which in turn could lead to early and late mortality after valve replacement for aortic stenosis. The amount of left ventricular fibrous tissue had a somewhat lesser influence. Left ventricular myocardial structure thus played a part, as an underlying basis, in the risk profile of adult patients with severe aortic stenosis. Impaired left ventricular systolic and diastolic function and excess mortality after surgery were related to damage to the hypertrophied myocardium, which was probably irreversible because it had lost the ability to dwindle to normal size after removal of the outflow tract obstruction.

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References


[34] Lund O, Pilegaard HK, Nielsen TT, Knudsen MA, M agnussen K. Thirty-day mortality after valve replacement.


Appendix 1

Lower or upper 95% confidence limits from a reference study of healthy sedentary individuals aged 20–70 years were used to identify patients with subnormal or supranormal (as appropriate) ventricular function indices. Subnormal or supranormal levels were:

- Right ventricular ejection fraction <49%; pulmonary transit time >5·5 s (age ≤ 49) or >6·3 s (age ≥ 50);
- Left ventricular ejection fraction <61%; peak ejection rate <2·29 EDV . s⁻¹;
- Time to peak ejection–systole duration ratio >0·67;
- Systole duration–heart cycle ratio >0·47 (age ≤ 59) or >0·50 (age ≥ 60);
- Peak filling rate <2·86 EDV . s⁻¹ (age ≤ 49) or <2·00 (age ≥ 50);
- Fast filling fraction <55% (age ≤ 49) or <32% (age ≥ 50);
- PQ filling fraction >23% (age ≤ 49) or >32% (age ≥ 50);
- Time to peak filling–diastole duration ratio >0·41 (age ≤ 49) or >0·55 (age ≥ 50);
- End-diastolic volume index >89 ml . m⁻².