Histologic Abnormalities of the Ascending Aorta: Effects on Aortic Remodeling after Intracardiac Repair of Tetralogy of Fallot

We evaluated aortic tissue specimens from patients undergoing tetralogy of Fallot repair, to determine whether histologic abnormalities affect postsurgical aortic remodeling and other patient-related variables.

Using light microscopy, we studied full-thickness aortic wall tissue operatively excised from 118 consecutive patients undergoing intracardiac repair of tetralogy of Fallot. We performed multiple linear regression analysis to identify independent predictors of change in aortic root dimensions, which we measured with echocardiography after repair and every 3 months thereafter.

Thirty histologically normal specimens were used as controls. Elastic fiber fragmentation was found in 74.6% of the abnormal specimens, mucoid extracellular matrix accumulation in 49.2%, smooth muscle cell nuclei loss in 39%, smooth muscle cell disorganization in 28.8%, and medial fibrosis in 52.5%. At a mean follow-up time of 83.55 ± 42.08 months, mean aortic sinotubular diameter decreased from 28.79 ± 9.15 to 27.16 ± 8.52 mm/m² (β = −0.43, P < 0.001). Aortic sinotubular diameter decreased by 0.6 mm/m² among females (β = 0.6, SE = 0.31; P = 0.05) and by 0.88 mm/m² in patients who had elastic fiber fragmentation or loss (β = 0.88, SE = 0.38, P = 0.02). In bivariate and multiple linear regression analysis, duration of follow-up emerged as an independent predictor of aortic remodeling.

The aortic histopathologic changes in our patients had an independent negative impact on the degree of aortic remodeling after surgery. We observed the most improved aortic sinotubular diameter in patients who had either histologically normal aortas or aortas with elastic fragmentation. (Tex Heart Inst J 2020;47(2):86-95)

Even after intracardiac repair of tetralogy of Fallot (TOF), many patients will have a large aortic root or progressive aortic root dilation, leading to aortic valve incompetence and necessitating reoperation.1,3 The Mayo Clinic group4 was the first to theorize that intrinsic aortic wall abnormalities are caused by long-standing volume overload, because most patients whom they studied were 10 years old at time of primary repair. Accordingly, early repair of TOF has been advocated.1,4-6 Progressive aortic root dilation has been reported to be related to factors such as right aortic arch, male sex, and palliative shunts having been placed before repair.1,4 Because so few patients are affected, the literature on remodeling and size changes of the ascending aorta and aortic root after intracardiac TOF repair is sparse.1 6

Earlier, we reported a statistically significant relationship between lamellar loss and the appearance of several histopathologic changes in the aortic walls of 98 patients who had TOF. The changes included elastic fiber fragmentation or loss, mucoid extracellular matrix accumulation, smooth muscle cell (SMC) nuclei loss, SMC disorganization, and medial fibrosis. These were present in infants and were more pronounced in older patients who had long-standing cyanosis and volume overload; in addition, most of the ascending aortic media was involved. The changes may reduce medial cohesive-ness and tensile strength and contribute to aortic root dilation. We postulated that the severity of pathologic changes in the ascending aorta has a role in remodeling of the ascending aorta and aortic root after intracardiac repair of TOF.

In our previous work, we investigated the sensitivity, specificity, and accuracy of a low lamellar count in predicting the presence of a histologically abnormal aorta. In the current study, we did not repeat that analysis. Instead, we analyzed echocardiographic measurements of aortic sinotubular diameter to determine whether relationships exist between histopathologic abnormalities of the aortic wall and changes in aortic root dimension from the first treatment for TOF or Fallot-type double-outlet right ventricle.
(DORV) to the latest evaluation, follow-up duration, and other patient-related variables.

**Patients and Methods**

The current study was approved by our Institutional Ethics Committee and conformed with the Declaration of Helsinki. Patients were enrolled after their parents or guardians provided written informed consent for removal of aortic tissue and anonymous analysis of data.

From January 2004 through December 2016 at our institution, we excised and analyzed aortic wall specimens from 138 consecutive patients who underwent repair of TOF or DORV with pulmonic stenosis. Specimens with more than half of an aortic valvular orifice supported by the right ventricle (RV) were considered to be double-outlet. We excluded 20 patients: 4 died immediately after repair (2 of massive pulmonary bleeding, 2 of low cardiac output syndrome and multiorgan failure), and 16 had insufficient tissue for evaluation or morphologic artifacts resulting from inadequate fixation or poor orientation. Consequently, samples from 118 patients (81 males) were suitable for analysis. The study group consisted of 98 patients from the previous cohort and 20 new patients.

All patients underwent 2-dimensional echocardiographic measurement of the internal diameter of the aortic root at the sinotubular junction, at midsystole, in the parasternal long-axis view. All patients remained eligible for the study if they had been monitored for at least 6 months, had available data on height and weight at each outpatient visit, and had at least 3 sequential good-quality echocardiograms of the aortic root available during follow-up.

Absolute values (recorded in mm) were normalized for each patient’s body surface area (BSA) and age. Indexed aortic sinotubular diameter was grouped per year of follow-up after correction and was compared per patient between the time of repair and each year of follow-up. In addition, the difference in absolute aortic sinotubular diameter indexed to BSA (mm/m²) was calculated for each patient between the times of correction and final measurement, to detect regression or progression of aortic annular dilation.

We specifically analyzed the relationship between postsurgical aortic remodeling and abnormal aortic histopathologic conditions and other potential variables.

**Patient Characteristics**

Patient age at correction ranged from 6 months to 47 years (mean ± SD, 110.6 ± 94.8 mo; median, 74 mo). Forty patients (33.9%) were younger than 4 years, and 30 (25.4%) were 12 years or older (Table I). Angiocardiography was performed to confirm the diagnosis of TOF, define coronary artery anatomy, and identify major aortopulmonary collateral arteries (MAPCAs).

| Variable | Value |
|----------|-------|
| Age at operation (mo) | 110.6 ± 94.8 (6 mo–47 yr; 74 mo) |
| Age at operation (yr) | <4 40 (33.9) |
| | 4–12 48 (40.7) |
| | ≥12 30 (25.4) |
| Male | 81 (68.6) |
| Weight at operation (kg) | 22.42 ± 14.26 (2.5–80; 17) |
| Body surface area (m²) | 0.86 ± 0.42 (0.19–2; 0.8) |
| Previous Blalock-Taussig shunt | 30 (25.4) |

**Preoperative characteristics**

- Hemoglobin (g/dL) 14.8 ± 3.8 (14–25)
- Hematocrit (%) 67 (56.8)
- RV end-diastolic pressure (mmHg) ≥12 27 (22.9)
- <12 91 (77.1)
- SaO₂ (%) ≤80 57 (48.3)
- >80 61 (51.7)
- Coil embolization of MAPCAs 38 (32.2)
- Right aortic arch 26 (22)
- Dilated aortic annulus 76 (64.4)
- Aortic regurgitation
  - Present 14 (11.9)
  - Absent 104 (88.1)
- Aortic override (%)
  - ≥50 52 (44.1)
  - <50 66 (55.9)
- Indexed aortic diameter (mm/m²)
  - Preoperative 28.79 ± 9.15 (13.7–50; 28.5)
  - Postoperative 27.16 ± 8.52 (13–47; 27)

**Diagnosis**

- TOF with pulmonary stenosis 66 (55.9)
- Double-outlet RV (Fallot type) 52 (44.1)
- Lowest temperature at operation 28 °C
- Aortic cross-clamp time (min) 40 ± 13.8
- Peak systolic RV/LV
  - Preoperative 0.98 ± 0.08
  - Postoperative 0.49 ± 0.28
- Postoperative PAP (mmHg) 15.8 ± 4.2

MAPCAs = major aortopulmonary collateral arteries; PAP = pulmonary artery pressure; RV/LV = right-to-left ventricular pressure ratio; RV = right ventricle; SaO₂ = systemic arterial oxygen saturation; TOF = tetralogy of Fallot

Data are presented as mean ± SD, mean ± SD with range and median, or number and percentage.

Standard cardiopulmonary bypass and myocardial protection techniques were used in all patients. Intracardiac repair of TOF involved a trans-right atrial, transpulmonary approach in 95 patients and a trans-right atrial
A transannular patch was used in 78 patients. All operations were performed by a single surgeon (UKC) to maintain uniformity in the study protocol.

**Aortic Dilation.** Aortic root dimensions in normal hearts were determined in accordance with age, height, body weight, and sex.\(^6\) We used the standard nomogram for aortic root size at the sinotubular junction, as described by Roman and associates,\(^4\) indexed to BSA and age.\(^5,6\) We considered dilation to be present after TOF repair when aortic root diameter was 1.5 times greater than expected. Of the 118 patients, 76 (64.4%) had aortic dilation.

Table II shows the characteristics of the overall study cohort and the 76 patients who had dilated aortas. Of those 76, 50 (65.8%) had aortic override >50%, 32 (42.1%) had MAPCAs, 13 (17.1%) had aortic regurgitation, 21 (27.6%) had a modified Blalock-Taussig shunt, another 21 had right aortic arch, 36 (47.4%) had TOF with pulmonic stenosis, and 40 had DORV (Fallot type). Of the 76 patients, 73 (96.1%) also had histologic abnormalities in the aortic wall.

**Tissue Collection and Evaluation**

Our methods for collecting, processing, and staining specimens have been described.\(^7\) Briefly, at the aortic cannulation site, a button of full-thickness aortic wall tissue (width, 2–3 mm) was carefully excised from within the aortic pursestring suture on a side-biting aortic clamp. Each biopsy specimen was affixed in 10% buffered formalin solution at room temperature within the aortic pursestring suture on a side-biting aortic clamp. Each biopsy specimen was affixed in 10% buffered formalin solution at room temperature and embedded in paraffin, and thin sections of 4 µm to 5 µm were taken. Slides were stained with hematoxylin & eosin (H & E); or, when indicated, with Masson’s trichrome, Verhoeff-van Gieson elastic, or Alcian blue/periodic acid–Schiff stain. A Nikon Optiphot research light microscope (Nikon Instruments Inc.) was used to photograph the slides. The magnification data (orig. \(\times 40\), \(\times 100\), or \(\times 200\)) indicated both ocular and objective magnification.

**Aortic Media Variables**

Evaluation of the aortic media included 6 variables: lamellar count, elastic fiber fragmentation or loss, mucoid extracellular matrix accumulation, SMC nuclei loss, SMC disorganization, and medial fibrosis. Lesions were graded as mild-to-moderate or severe in accordance with criteria adopted from Halushka and associates.\(^11\) Two independent pathologists, blinded to the study, reviewed each slide; they agreed that 30 tissue sections were normal and 88 were abnormal (kappa value, 1.00).

*Lamellar Count.* Corrugated, longitudinally arranged elastic lamellae extending across the length of the image parallel to the lumen were counted at the thickest and thinnest areas of the media, and the mean of these numbers was calculated. Grades were determined from the

| Variable | All Patients (n=118) | Aortic Dilation (n=76) |
|----------|---------------------|-----------------------|
| Age (yr) |                      |                       |
| ≥28      | 43 (36.4)            | 40 (52.6)             |
| <8       | 75 (63.6)            | 36 (47.4)             |
| Male     | 81 (68.6)            | 65 (85.5)             |
| Female   | 37 (31.4)            | 11 (14.5)             |
| Sa (%)   |                      |                       |
| <80      | 57 (48.3)            | 51 (67.1)             |
| ≥80      | 61 (51.7)            | 25 (32.9)             |
| Hematocrit (%) |              |                       |
| ≥45      | 67 (56.8)            | 50 (65.8)             |
| <45      | 51 (43.2)            | 26 (34.2)             |
| Aortic override (%) |          |                       |
| ≥50      | 52 (44.1)            | 50 (65.8)             |
| <50      | 66 (55.9)            | 26 (34.2)             |
| Aortic regurgitation |          |                       |
| Present  | 14 (11.9)            | 13 (17.1)             |
| Absent   | 104 (88.1)           | 63 (82.9)             |
| MAPCAs   | Present             | 32 (42.1)             |
| Absent   | 79 (66.9)            | 44 (57.9)             |
| Previous palliation |          |                       |
| Yes      | 30 (25.4)            | 21 (27.6)             |
| No       | 89 (74.6)            | 55 (72.4)             |
| RV end-diastolic pressure (mmHg) |          |                       |
| ≥12      | 27 (22.9)            | 23 (30.3)             |
| <12      | 91 (77.1)            | 53 (69.7)             |
| Right aortic arch |          |                       |
| Present  | 26 (22)              | 21 (27.6)             |
| Absent   | 92 (78)              | 55 (72.4)             |
| Diagnosis | TOF with pulmonary stenosis | 66 (55.9) |
| Double-outlet RV (Fallot type) | 52 (44.1) | 40 (52.6) |
| Lamellar count | Abnormal         | 68 (74.6) |
| Normalb  | 30 (25.4)            | 3 (3.9)               |
| Aortic wall histology | Abnormal   | 88 (74.6) |
| Normal   | 30 (25.4)            | 3 (3.9)               |
| Elastic fiber fragmentation or loss | Present | 88 (74.6) |
| Absent   | 30 (25.4)            | 3 (3.9)               |
| Mucoid extracellular matrix accumulation | Present | 58 (49.2) |
| Absent   | 60 (50.8)            | 22 (28.9)             |
| SMC nuclei loss | Present | 46 (39) |
| Absent   | 72 (61)              | 38 (50)               |
| SMC disorganization | Present | 34 (28.8) |
| Absent   | 84 (71.2)            | 46 (60.5)             |
| Medial fibrosis | Present | 62 (52.5) |
| Absent   | 56 (47.5)            | 20 (26.3)             |

MAPCAs = major aortopulmonary collateral arteries; RV = right ventricle; SMC = smooth muscle cell; Sa = systemic arterial oxygen saturation; TOF = tetralogy of Fallot

\(^a\) Mean ± SD, 40.85 ± 10.05; range, 15–59

\(^b\) Mean ± SD, 65.84 ± 4.27; range, 60–75

Data are presented as number and percentage.
degree of lamellar fragmentation. Extensive fragmentation was characterized by near loss of medial elastic fibers, which created increasingly extended translamellar spaces. The longer elastic lamellae parallel to the lumen were included in the count.

Elastic Fiber Fragmentation or Loss. Fragmentation or loss of elastic fibers of the media increases translamellar spaces, revealed by staining. Because this degeneration can be patchy, focal, or multifocal, grading as mild, moderate, or severe is essential. Multifocal fiber disruption with widened, extended translamellar spaces is considered severe.11

Mucoid Extracellular Matrix Accumulation. Increased aortic medial mucoid extracellular matrix accumulation (MEMA) creates intralamellar expansion, translamellar expansion, or both, including extracellular pools. In intralamellar MEMA, the increase in the mucoid extracellular matrix does not substantially alter the arrangement of lamellar units, whereas in translamellar MEMA, variable alteration occurs.11

Loss of Smooth Muscle Cell Nuclei. In one region of the aortic media, SMC nuclei, involving multiple lamellae, are not clearly identifiable on H & E stain. The term “SMC nuclei loss” replaces terms such as medionecrosis or SMC necrosis, which merely imply a loss of SMCs. The SMC nuclei can be lost in patches or in a band-like pattern. This lesion can be noted in the absence of medial laminar collapse.11

Smooth Muscle Cell Disorganization. Nonparallel arrangement or disarray of medial SMCs results in disorganization or, sometimes, nodular aggregates of SMCs. Elastic fiber disorganization may concomitantly be seen.11

Medial Fibrosis. An increase in collagen fibers creates areas of substitutive fibrosis or wider intralamellar spaces in the media, along with a progressively nonparallel arrangement of the elastic lamellae (or lamellar units). Increased intralamellar collagen does not substantially alter arrangements of lamellar units, whereas increased translamellar collagen is more scarlike and does cause changes. Increases are graded mild, moderate, or severe.11

Apoptosis. Apoptosis, a form of programmed cell death, is a central feature of fundamental biological processes, including embryonic morphogenesis, remodeling of mature tissues, and cell replacement in certain adult tissues. In contrast with necrosis, apoptosis occurs in isolated cells without an accompanying cellular reaction.12 Although apoptosis was not among the 6 variables that we formally evaluated, we remained aware that it might affect our observations.

Statistical Analysis
We used Intercooled STATA 9.0 Software (StataCorp LLC) for statistical analysis. Independent variables were expressed as mean ± SD with range and median, and categorical variables as number and percentage. We used the Wilcoxon signed-rank test to compare aortic sinotubular diameter at yearly intervals after TOF repair. We used a sign test to compare diameters at correction and at the last measurement in the same patient, and we compared changes in diameter between various categories of predictors by using a t test for independent samples.

Regression analysis enabled us to identify predictors of change in diameter. Multiple linear regression analysis was performed by considering variables with P values <0.2 in the simple regression analysis to identify independent predictors of change in aortic root dimensions, and also relationships between abnormalities, dilation, and remodeling. Results are presented as β (regression coefficient) with SE. P <0.05 was considered statistically significant.

Results
Follow-up was 100% complete; we collected 821.6 patient-years of data with a mean follow-up time of 83.55 ± 42.08 months (range, 1–156 mo). There were no late deaths. The actuarial survival rate at 156 months was 97.8% ± 0.01% (95% CI, 0.91–0.99). The 118 patients underwent postoperative clinical, electrocardiographic, and echocardiographic examinations every 3 months. At their final examinations, 116 patients (98.3%) were in New York Heart Association functional class I or II. Two patients (1.7%) who were taking diuretics and vasodilators late postoperatively were in functional class III. Grade I aortic regurgitation occurred in 10 patients (8.5%). No patient needed reoperation, reintervention, or new surgery for aortic problems.

Histopathologic Analysis
The 30 histologically normal tissue specimens had layers of longitudinally arranged elastic lamellae interspersed with SMCs and collagen fibrils in a mucopolysaccharide ground substance (Fig. 1).

Elastic fiber fragmentation or loss was found in 88 (74.6%) of the study specimens (Fig. 2). MEMA in 58 (49.2%) (Fig. 3), SMC nuclei loss in 46 (39%) (Fig. 4A) and SMC disorganization in 34 (28.8%) (Fig. 4B), and medial fibrosis in 62 (52.5%) (Fig. 5 and Table II).

Postoperative Analysis and Follow-Up
Preoperative echocardiograms showed dilated aortic sinotubular diameter in 76 patients (64.4%). At 83.55 ± 42.08 months postoperatively (range, 1–156 mo), mean diameter had decreased significantly from 28.79 ± 9.15 mm/m2 (range, 13.7–50 mm/m2; median, 28.5; 95% CI, 27.12–30.46) to 27.16 ± 8.52 mm/m2 (range, 13–47 mm/m2; median, 27; 95% CI, 25.61–28.71) (r = −0.43; P <0.001) (Table III). The relationship between mean reduction of aortic sinotubular diameter and follow-up duration was significant.
After comparing the mean reduction in aortic sinotubular diameter with the 6 individual aortic histopathologic variables, we found greater reduction in patients who had histologically normal aortas than in patients who had abnormalities (Table III).

Regression analysis revealed the following unadjusted independent predictors of reduced aortic sinotubular diameter (aortic remodeling): systemic arterial oxygen saturation ($\text{Sao}_2$) $<80\%$ ($\hat{\beta}=0.82, \text{SE}=0.25; \ P=0.001$), hematocrit $\geq 45\%$ ($\hat{\beta}=0.74, \text{SE}=0.25; \ P=0.004$), right aortic arch ($\hat{\beta}=0.71, \text{SE}=0.31; \ P=0.02$), lamellar count $<60$ ($\hat{\beta}=0.76, \text{SE}=0.27; \ P=0.007$), follow-up duration ($\hat{\beta}=-0.14, \text{SE}=0.003; \ P<0.001$), elastic fiber fragmentation ($\hat{\beta}=0.62, \text{SE}=0.25; \ P=0.01$), SMC nuclei loss ($\hat{\beta}=0.53, \text{SE}=0.26; \ P=0.04$), and SMC disorganization ($\hat{\beta}=0.56, \text{SE}=0.28; \ P=0.05$) (Table IV).

In the comparison of indexed aortic sinotubular diameter at correction with that at last measurement, significant decrease occurred over time, regardless of histopathologic abnormality. When diameter change was compared between various categories of independent predictors, the reduction was significantly greater in patients with histologically normal aortas (Table IV).

We adjusted for variables, performed multiple linear regression analysis, and identified independent predic-

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**Fig. 1** Light microscopic photomicrograph of an ascending aortic biopsy specimen from a patient with tetralogy of Fallot shows normal aortic media with corrugated, longitudinally arranged lamellar units of fenestrated elastic laminae enclosing smooth muscle cells, collagen fibers, and a large amount of proteoglycans (Verhoeff-van Gieson stain, orig. ×40).

**Fig. 2** Light microscopic photomicrographs (Verhoeff-van Gieson stain, orig. ×200) of biopsy specimens from the ascending aortas of patients with tetralogy of Fallot show A) fragmented elastic fibers no longer extending across the length of the image, B) even worse fragmentation, C) extensive fragmentation and near loss of elastic fibers of the media, creating increasingly extended translamellar spaces, and D) severe fragmentation (multifocal, with widening of translamellar spaces).
tors of change in aortic sinotubular diameter. Diameter reduction was 0.6 mm/m² ($\beta=0.6; SE=0.31; P=0.05$) in females, and it was 0.88 mm/m² ($\beta=0.88, SE=0.38; P=0.02$) in patients who had elastic fiber fragmentation or loss (Table IV).

Follow-up duration emerged as an independent predictor of the degree of aortic remodeling and dilation reduction in the entire group (Table IV). Maximal reduction in mean aortic sinotubular diameter was observed immediately after surgery. Late during follow-up, the monthly reduction was 0.01 mm/m² ($\beta=-0.01, SE=0.004; P=0.04$) (Table IV). Finally, we found no effects of apoptosis.

**Discussion**

Excluding our earlier investigation,$^7$ we are aware of only 5 English-language studies of intrinsic aortopathy in TOF (3 surgical and 2 autopsy series).$^{13-17}$ None
| Variable                        | Value (n=118) | Reduction in Aortic Sinotubular Diameter [mm/m²] | P Value | 95% CI       |
|--------------------------------|---------------|-----------------------------------------------|---------|--------------|
| Age (yr)                       |               |                                               |         |              |
| ≥8                             | 43 (36.4)     | 1.75 ± 1.35 (0.19–6.59; 1.8)                  | 0.5     | 1.34–2.16    |
| <8                             | 75 (63.6)     | 1.55 ± 1.45 (0.19–6; 0.9)                     | 0.22    | 1.22–1.89    |
| Sex*                           |               |                                               |         |              |
| Male                           | 81 (68.6)     | 1.47 ± 1.33 (0.19–6.59; 2)                    | 0.05    | 1.17–1.76    |
| Female                         | 37 (31.4)     | 1.97 ± 1.52 (0.19–6; 1.29)                    |         | 1.46–2.48    |
| SaO₂ (%)                       |               |                                               |         |              |
| <80                            | 61 (51.7)     | 2.02 ± 1.47 (0.19–6.59; 2)                    | 0.001   | 1.64–2.4     |
| ≥80                            | 57 (48.3)     | 1.20 ± 1.20 (0.19–6; 0.8)                     |         | 0.88–1.52    |
| Hematocrit* (%)                |               |                                               |         |              |
| ≥45                            | 67 (56.8)     | 1.94 ± 1.48 (0.19–6; 1.8)                    | 0.001   | 1.58–2.31    |
| <45                            | 51 (43.2)     | 1.20 ± 1.18 (0.1–6; 0.8)                      |         | 0.87–1.54    |
| Aortic override (%)            |               |                                               |         |              |
| ≥50                            | 52 (44.1)     | 1.64 ± 1.27 (0.19–6.59; 1.1)                  | 0.9     | 1.29–1.99    |
| <50                            | 66 (55.9)     | 1.61 ± 1.50 (0.19–6; 0.9)                     |         | 1.24–1.98    |
| Aortic regurgitation           |               |                                               |         |              |
| Present                        | 76 (64.4)     | 1.56 ± 1.36 (0.19–6.59; 1)                    | 0.5     | 1.26–1.87    |
| Absent                         | 42 (35.6)     | 1.74 ± 1.49 (0.19–6; 1)                      |         | 1.27–2.22    |
| MAPCAs                         |               |                                               |         |              |
| Present                        | 14 (11.9)     | 2.15 ± 1.30 (0.19–4.59; 2.1)                  | 0.14    | 1.39–2.9     |
| Absent                         | 104 (90.1)    | 1.56 ± 1.41 (0.19–6; 1)                      |         | 1.28–1.83    |
| Previous palliation            |               |                                               |         |              |
| Yes                            | 30 (25.4)     | 1.84 ± 1.62 (0.19–6; 1)                      | 0.34    | 1.23–2.44    |
| No                             | 88 (74.6)     | 1.55 ± 1.32 (0.19–5.8; 1)                    |         | 1.27–1.83    |
| RV end-diastolic pressure (mmHg) |           |                                               |         |              |
| ≥12                            | 27 (22.9)     | 1.81 ± 1.44 (0.19–6.59; 1.8)                  | 0.43    | 1.24–2.38    |
| <12                            | 91 (77.1)     | 1.57 ± 1.40 (0.19–6; 1)                      |         | 1.28–1.86    |
| Right aortic arch*             |               |                                               |         |              |
| Present                        | 26 (22)       | 2.18 ± 1.55 (0.5–6.6; 2)                      | 0.02    | 1.55–2.81    |
| Absent                         | 92 (78)       | 1.47 ± 1.33 (0.5–5.9; 0.9)                    |         | 1.19–1.74    |
| Diagnosis                      |               |                                               |         |              |
| TOF with PS                    | 66 (55.9)     | 1.42 ± 1.25 (0.5–5; 0.9)                      | 0.06    | 1.11–1.72    |
| Double-outlet RV (Fallot type) | 52 (44.1)     | 1.89 ± 1.55 (0.2–6.59; 1.2)                   |         | 1.46–2.32    |
| Lamellar count*                |               |                                               |         |              |
| <60                            | 83 (70.3)     | 1.85 ± 1.44 (0.19–6.59; 1.4)                  | 0.03    | 1.54–2.17    |
| ≥60                            | 35 (29.7)     | 1.09 ± 1.15 (0.5–8.9; 0.8)                    |         | 0.69–1.48    |
| Elastic fiber fragmentation or loss* | 88 (74.6) | 1.30 ± 1.13 (0.6–0.79) | 0.008 | 1.52–2.32 |
| Absent                         | 30 (25.4)     | 1.92 ± 1.56 (0.19–6.59; 1.29)                 |         | 1–1.6        |
| Mucoid extracellular matrix accumulation | 58 (49.2) | 1.44 ± 1.18 (0–6; 1) | 0.07 | 1.39–2.23 |
| Absent                         | 60 (50.8)     | 1.81 ± 1.59 (0.19–6.59; 1)                    |         | 1.13–1.75    |
| SMC nuclei loss*               |               |                                               |         |              |
| Present                        | 46 (39)       | 1.42 ± 1.29 (0–6; 0.89)                      | 0.02    | 1.5–2.4      |
| Absent                         | 72 (61)       | 1.95 ± 1.52 (0.19–6.59; 2)                    |         | 1.11–1.72    |
| SMC disorganization*           |               |                                               |         |              |
| Present                        | 34 (28.8)     | 2.02 ± 1.58 (0.19–6.59; 2)                    | 0.02    | 1.47–2.58    |
| Absent                         | 84 (71.2)     | 1.46 ± 1.30 (0–6; 0.94)                      |         | 1.18–1.75    |
| Medial fibrosis*               |               |                                               |         |              |
| Present                        | 62 (52.5)     | 1.25 ± 0.95 (0–3.7; 0.94)                     | 0.04    | 1.43–2.07    |
| Absent                         | 56 (47.5)     | 1.75 ± 1.51 (0.19–6.59; 1)                    |         | 0.89–1.16    |

MAPCAs = major aortopulmonary collateral arteries; PS = pulmonary stenosis; RV = right ventricle; SaO₂ = systemic arterial oxygen saturation; SMC = smooth muscle cell; TOF = tetralogy of Fallot

*Variables with higher risk

Data are presented as number and percentage or as mean ± SD with range and median. P <0.05 was considered statistically significant.
focused on the relationship between abnormal aortic histopathologic factors and their possible effects on aortic remodeling after intracardiac TOF repair.

We found that abnormal aortic histopathology can affect aortic remodeling after TOF repair. We made 5 principal observations:

• Preoperative aortic dilation was present in 64.4% of patients.
• Substantial lamellar loss and abnormal aortic histopathologic conditions were seen in 96.1% of patients with aortic dilation.
• Intrinsic abnormal aortic histopathologic factors were present in patients as young as 4 years of age.
• Postoperative aortic sinotubular diameter (chiefly dependent on the presence or absence of intrinsic aortopathy) was significantly reduced in all patients over time.
• Postsurgical aortic remodeling in patients undergoing TOF was independently negatively affected by individual intrinsic aortic pathologic conditions.

Aortic media consists of layers of longitudinally arranged lamellar units of fenestrated elastic lamellae that enclose SMCs, collagen fibers, and a large amount of proteoglycans. Each lamella with its SMCs synthesizes the connective-tissue matrix and forms a lamellar unit approximately 11 µm thick. The media is not a static structure. As people age, elastic lamellae reduplicate with corresponding fragmentation, wider interlamellar spaces, MEMA, and eventual fibrosis. Although elastin has a long half-life (40–70 yr), in TOF it is unclear whether elastin loss is caused by increased elastolysis stimulated by shear force within the dilated aorta, or by intrinsic problems of elastin synthesis. Regardless, these abnormalities reduce medial cohesiveness and tensile strength, causing aortic dilation.

Table IV. Predictors of Mean Reduction of Aortic Sinotubular Diameter by Multiple Linear Regression Analysis in All Patients

| Variable                          | Unadjusted | Adjusted |
|-----------------------------------|------------|----------|
|                                   | β          | SE       | P Value | β          | SE       | P Value |
| Age >8 years                      | 0.19       | 0.27     | 0.4     | —          | —        | —       |
| Female sex*                       | 0.5        | 0.27     | 0.07    | 0.6        | 0.31     | 0.05    |
| SaO₂ <80%*                        | 0.82       | 0.25     | 0.001   | 0.49       | 0.58     | 0.4     |
| Hematocrit ≥45%*                  | 0.74       | 0.25     | 0.004   | −0.02      | 0.52     | 0.9     |
| Aortic override ≥50%              | 0.03       | 0.26     | 0.9     | —          | —        | —       |
| Aortic dilation (n=76)            | −0.17      | 0.27     | 0.5     | —          | —        | —       |
| Aortic regurgitation              | 0.59       | 0.39     | 0.14    | −0.22      | 0.42     | 0.6     |
| MAPCAs                            | 0.06       | 0.27     | 0.8     | —          | —        | —       |
| Previous palliation               | 0.28       | 0.29     | 0.9     | —          | —        | —       |
| RVEDP ≥12 mmHg                    | 0.24       | 0.31     | 0.4     | —          | —        | —       |
| Right aortic arch*                | 0.71       | 0.31     | 0.02    | 0.3        | 0.34     | 0.9     |
| Diagnosis                         | 0.47       | 0.26     | 0.07    | 0.31       | 0.26     | 0.2     |
| Follow-up duration*               | −0.14      | 0.003    | <0.001  | −0.01      | 0.004    | 0.04    |
| Abnormal aortic wall histology (n=88) | 0.62     | 0.25     | 0.01    | 0.88       | 0.38     | 0.02    |
| Lamellar count <60 (n=83)*        | 0.76       | 0.27     | 0.007   | 0.5        | 0.31     | 0.1     |
| Elastic fiber fragmentation or loss (n=88)* | 0.62     | 0.25     | 0.01    | 0.88       | 0.38     | 0.02    |
| MEMA (n=58)                       | 0.37       | 0.26     | 0.15    | −0.19      | 0.36     | 0.59    |
| SMC nuclei loss (n=46)*           | 0.53       | 0.26     | 0.04    | −0.24      | 0.37     | 0.5     |
| SMC disorganization (n=34)*       | 0.56       | 0.28     | 0.05    | −0.04      | 0.38     | 0.9     |
| Medial fibrosis (n=62)            | 0.51       | 0.29     | 0.09    | −0.84      | 0.46     | 0.06    |

β = regression coefficient; MAPCAs = major aortopulmonary collateral arteries; MEMA = mucoid extracellular matrix accumulation; RVEDP = right ventricular end-diastolic pressure; SaO₂ = systemic arterial oxygen saturation; SMC = smooth muscle cell

*Variables with higher risk

P <0.05 was considered statistically significant.
We found reports of aortic dissection in 2 patients whose root diameters were 6.45 and 6.7 cm, respectively.22,23

In the current study, 64.4% of patients undergoing TOF repair had aortic root dilation before operation. None have since needed surgery for mild aortic regurgitation. Of note, 96.1% of patients with dilation had histologic abnormalities. The patients with dilated aortas and histologically normal tissue had DORV with pulmonic stenosis. Of the patients whose tissue was normal, 73.3% had TOF, and 26.7% had DORV.

Logistic regression analysis in our previous investigation,7 which accounted for the effects of other variables, revealed a relationship between aortic dilation and patients 8 years or older, aortic override ≥50%, and the presence ( singly or in combination) of DORV, elastic fiber fragmentation, MEMA, SMC nuclei loss, SMC disorganization, and medial fibrosis.7 The relationship between lamellar loss and the appearance of histopathologic changes was significant. We then hypothesized that the severity of pathologic changes in the ascending aorta plays a role in remodeling the ascending aorta in patients with repaired TOF, and our current results support this.

Focal fragmentation of the elastic lamellae is the start of the disease process, and medial fibrosis is the end stage of noninflammatory degeneration. In the current study, 74.6% of patients had elastic fiber fragmentation, and 52.5% had medial fibrosis. The mean reduction in aortic sinotubular diameter was greater in patients with fragmentation (P=0.008) than in those with fibrosis (P=0.04). Diameter reduction was 0.88 mm/m² (P=0.02) in patients with fragmentation. Some individual intrinsic pathologic factors also had an independent negative impact on postsurgical remodeling.

Our cohort of 118 patients is perhaps the largest thus far to be evaluated for aortic remodeling after TOF repair. Age distribution was heterogeneous: 33.8% were aged 6 months to 4 years, and 25.4% were 12 years or older. Despite a median age of 6 years, only 25% of patients had undergone shunt correction. The patients’ socioeconomic profiles and lack of healthcare insurance led to their referrals for surgery at older ages.

Although degenerative changes occurred more frequently in patients 8 years or older, we had previously observed similar changes in a subset of patients younger than 2 years. In our earlier population,7 patients 8 years or older had higher risks than did younger patients, with odds ratios (95% CIs) as follows: fiber fragmentation, 20.48 (1.82–225.28); SMC nuclei loss, 4.30 (1.17–15.75); SMC disorganization, 3.11 (1.62–13.93); and medial fibrosis, 12.14 (1.19–117.46). We think that these results support early TOF repair. These histopathologic changes resemble those observed in patients with Marfan syndrome and aortic dilation.24

Earlier,7 we found that exonic DNA variants involve the fibrillin-1 gene in one or more of exons 24 through 28. The DNA sequence variants were more pronounced in patients with TOF and dilated aorta in the presence of aortic histopathologic abnormalities.25 Some investigators have postulated that a combination of intrinsic premature cellular injury or cell death and programmed stress-induced activation of tissue enzymes might cause such apoptotic changes2,29; however, we found no association.

Few investigators have focused on progressive aortic root dilation after TOF repair.6,26 In the first series,4 surgical results were excellent, yet many patients developed aortic dilation and needed reoperation on the root. In a subset of 43 patients who underwent repair before the age of one year, Bacha and associates6 reported normal root diameters after 7 years; however, this study used only 2 measurements, and meaningful data were lacking. Francois and colleagues26 showed that, in patients with TOF who had dilated aortic roots and who underwent early repair, the aortic annulus and sinotubular junction became normal within 7 years. This process seems to be delayed at the level of the aortic sinuses.

Regardless of histopathologic changes, no patient in our study had surgery on the ascending aorta or aortic root at the time of TOF repair or late postoperatively. Furthermore, the larger data set enabled us to elucidate some of the mechanical effects of histopathologic abnormalities on aortic remodeling after TOF repair. Our findings indicate that the indexed aortic sinotubular diameter decreased slowly, but consistently, in all patients, regardless of pathologic condition. Follow-up duration was an independent predictor of the degree of aortic remodeling and dilation reduction in the entire group, in bivariate and multiple linear regression analyses. Maximally reduced mean diameter was observed immediately after surgery. Late postoperatively, monthly reduction was 0.01 mm/m² (P=0.04). Thus, the longer the follow-up time, the greater the diameter reduction.

Among the independent predictors of reduced aortic sinotubular diameter, the greater reduction in patients with elastic fiber fragmentation than in patients with medial fibrosis supports early TOF repair. Early repair may slow RV dilation and fibrosis by decreasing volume and pressure overload, thus limiting hemodynamic stress that can eventually lead to aortic dilation and regurgitation. Nevertheless, patients need lifelong monitoring with use of echocardiography or magnetic resonance imaging to track aortic size and RV outflow.

Study Limitations. Our study has limitations. It was observational and conducted in a single center. Whereas the greatest stress generated on the volume-loaded aorta in TOF is at the aortic root and ascending aorta, the tissue samples were obtained from the aortic cannulation site, to avoid destroying the aortic root structures. The small number of patients with MAPCAs and aortic regurgitation may explain why no relationships were observed between these variables and aortic remodeling.
To corroborate our current findings of changes in aortic root dimension, we plan a complete analysis of all our patients with TOF, to include echocardiographic measurements of the aortic annulus, sinus of Valsalva, and sinotubular junction, and examination of their relationship to body growth, aortic and pulmonary histopathologic conditions, and other variables. A multi-institutional prospective study is needed to test whether the severity of aortic histopathologic abnormalities plays a role in remodeling of the ascending aorta.

Conclusions

The ascending aortic media in patients with TOF exhibits substantial lamellar loss and intrinsic aortopathy. Within 7 years of repair, the diameter of the aortic root regresses at the level of the aortic sinotubular junction. The degree of postsurgical aortic remodeling is greater in patients who have histologically normal aortas or aortas with elastic fragmentation. Histologic abnormalities have individual and significant negative effects on postsurgical aortic remodeling. Early repair of TOF should slow degeneration and aortic root enlargement, and, with time, promote regression in root diameter. Despite improvement over time, patients who have undergone TOF repair should have aortic size and RV outflow tract condition evaluated regularly.

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