**SUPPLEMENTARY DATA**

**SUPPLEMENTARY METHODS**

**Patient population**

This cross-sectional, retrospective, observational and descriptive study included patients with severe isolated aortic stenosis (AS) referred to *Hospital Universitario de Navarra* for aortic valve (AV) replacement from June 2013 to February 2015. In agreement with the European Society of Cardiology recommendations,[1] AS was diagnosed as AV area ≤ 1 $cm^2$, transaortic mean pressure gradient ≥ 40 mm Hg, and peak velocity ≥ 4 m/s. Exclusion criteria included moderate or severe concomitant valvular disease; malignant tumor; contraindications to cardiac magnetic resonance (CMR), including claustrophobia, pacemaker and defibrillator implantation and an estimate glomerular filtration rate of < 30 mL/min. All patients were evaluated by transthoracic echocardiography and CMR imaging. Venous blood samples were drawn atraumatically and without stasis within the 24 hours before the surgery to quantify serum sST2 levels and other routine analytes. Serum samples were collected upon whole blood centrifugation at 1500 g, 10 minutes in a refrigerated centrifuge, aliquoted and transferred to −80º C until batch analyses. Conventional coronary angiography or computed tomography coronary angiography was performed for determination of significant concomitant coronary artery disease (CAD). Informed consent was obtained from each patient. The study protocol conformed to the ethics guidelines of the 1975 Declaration of Helsinki and was approved by the institution's human research committee (*Comité Ético de Experimentación Clínica. Gobierno de Navarra, Departamento de Salud*; Ethics number: 2015/26).

**Cardiac magnetic resonance**

CMR imaging studies were performed using a 1.5 T scanner (Avanto) under the standard protocols. Steady-state free precession sequences were used for the assessment of left ventricle (LV) dimensions,

mass and function, and AV area. Late-gadolinium enhancement (LGE) images were acquired 10 to 15 minutes after injection of intravenous gadolinium contrast agent (Dotarem, 0.1 mmol/kg body weight). Inversion recovery-prepared spoiled gradient echo images were acquired in standard short- and long-axis views to detect areas of LGE. Inversion delay times were optimized to null normal myocardium.

All examinations were analyzed by an experienced radiologist and a cardiologist with an interest in cardiac imaging, in consensus. Cine images (steady-state free precession) were used to assess LV volumes, mass, and function. The endocardial and epicardial LV borders were manually contoured in end-systole and end-diastole using the ARGUS software (Siemens Medical Solutions). AS severity was assessed using CMR-derived planimetry of the AV area as well as phase-contrast sequences to determine flow velocities. The presence and pattern of LGE was assessed by 2 independent observers who were blinded to the clinical data. Patients were grouped according to the presence or absence of LGE suggestive of replacement myocardial fibrosis (RMF). Patients with RMF were then divided into midwall or subendocardial fibrosis patterns. Patients with a mixed LGE pattern were classified according to the predominant fibrosis pattern. LGE mass was calculated semiautomatically by a single operator using QMass MR7 software. The endocardial and epicardial borders were traced for each short-axis slice. A region of interest was defined within normal myocardium in an area with uniform myocardial suppression free of artifacts. The extent of fibrosis was quantified in grams and as a percentage of total LV mass. Fibrosis within intramyocardial (diffuse or focal and including the interventricular junction) and subendocardial areas was considered pathological in this study. Focal fibrosis found around the mitral and aortic rings was not classified as pathological, as it is often seen in older patients, unless it was found as widely diffused from the mitral and aortic rings.

**Statistical analyses**

To determine that the sample size (n = 79) ensured a high power of our results, an intermediate effect size was calculated from previous studies of AS.[2] Briefly, Cohen's F effect size (f2 = 0.39) was used for an $\alpha = 0.05$. Variables are expressed as mean ± standard deviation or median [IQR], as appropriate. Comparisons of 2 continuous variables were performed using unpaired Student T test or Mann-Whitney U test, as appropriate. Comparison of ≥ 3 normal continuous variables was performed using 1-way analysis of variance (ANOVA), and *P*-values were adjusted by Bonferroni correction for multiple comparisons (post hoc analyses). Comparisons of ≥ 3 nonnormal variables were analyzed by Kruskal-Wallis and *P* values were adjusted applying Dunn's multiple post hoc test. Pearson's or Spearman's correlation coefficients were calculated. Categorical variables are expressed as percentages and compared using the chi-square test, or Fisher exact test, as appropriate. A 2-tailed *P*-value of < .05 was considered statistically significant. Receiver operating characteristic curves and Youden's statistic were used to determine the optimal sST2 cutoff value for determining the presence of RMF. The parametric bootstrap method was used to further validate the calculated sST2 cutoff.

Values of RMF were modeled by univariate and multivariate linear regression models to identify the clinical and biological markers able to stratify/identify patients with RMF likely to benefit from early valve replacement interventions. First, each variable was modeled using a univariate generalized linear model in association with RMF as a response variable. Nonlinear data were modeled using generalized additive models. Next, all variables significantly associated with RMF in univariate analyses were further assessed by multivariate analysis. Variable selection was carried out by stepwise backward selection.

Normal distribution of residuals was assessed by the Kolmogorov-Smirnov test and homocedasticity by the White test. Model relevance and results in the absence of assumption fulfillment were tested against ideal lambda likelihood profile and using Yeo-Johnson transformation.[3, 4]

To circumvent the risk of an over-fitted model and to check the internal validity of the model, we used bootstrap sampling (R = 1999). When modeling across bootstrap samples, the prognostic variables that were truly important should be retained in most models fitted.

All variables with $P > 0.05$ were removed from the model, which included only the best explanatory variables. All *P*-values included in this manuscript are net *P*-values unless otherwise indicated (eg, Bonferroni adjustment). All statistical analyses were performed with SPSS version 28.0.1.0 and using the R statistical package, v. 3.6 (R Foundation for Statistical Computing. Vienna, Austria). The graphs were plotted using GraphPad Prism 9.0 software.

**References**

1. Joint Task Force on the Management of Valvular Heart Disease of the European Society of Cardiology, European Association for Cardio-Thoracic Surgery, Vahanian A, et al. Guidelines on the management of valvular heart disease (version 2012). *Eur Heart J*. 2012;33:2451-2496.

2. Lancellotti P, Dulgheru R, Magne J, et al. Elevated Plasma Soluble ST2 Is Associated with Heart Failure Symptoms and Outcome in Aortic Stenosis. *PLoS One.* 2015;10:e0138940.

3. Yeo IK, Johnson RA. A new family of power transformations to improve normality or symmetry. *Biometrika*. 2000;87:954-959.

4. Pena EA, Slate EH. Global Validation of Linear Model Assumptions. *J Am Stat Assoc*. 2006;101:341.

**Table 1 of the supplementary data.** Characterization of the echocardiographic and RMF parameters depending on sST2 level

| | sST2 > 28.2 | sST2 < 28.2 | *P* |
|---|---|---|---|
| *TTE parameters* | | | |
| Maximum gradient, mmHg | 76.2 ± 18.5 | 77.2 ± 21.3 | .841 |
| Mean gradient, mmHg | 49.5 ± 14 | 50.8 ± 14.1 | .715 |
| AVA (VTI), $cm^2$ | 0.79 ± 0.18 | 0.73 ± 0.16 | .263 |
| *CMR parameters* | | | |
| LVEDD, mm | 50.6 ± 6.8 | 47.7 ± 5.1 | .061 |
| IVST, mm | 13.9 ± 2.7 | 12.5 ± 2.1 | .022 |
| EDV index, $mL/m^2$ | 76.2 ± 27.7 | 66.6 ± 17.7 | .120 |
| ESV index, $mL/m^2$ | 31.1 ± 21.3 | 22.9 ± 17.1 | .092 |
| LV mass index, $g/m^2$ | 98 ± 28.7 | 81.5 ± 17.3 | .010 |
| LVEF, % | 62.7 ± 14.2 | 67.8 ± 13.5 | .151 |
| LGE mass, g | 5.1 [2.12-11.9] | 0 | |
| LGE LV mass, % | 3 [1-6] | 0 | |

AVA (VTI), aortic valve area (continuity equation by velocity time integral); CMR, cardiac magnetic resonance imaging; EDV, end-diastolic volume; ESV, end-systolic volume; IVST, interventricular septum thickness; LGE, late-gadolinium enhancement; LV, left ventricle; LVEDD, left ventricle end-diastolic diameter; LVEF, left ventricular ejection fraction; sST2, soluble ST2; TTE, transthoracic echocardiography.

Data are expressed as mean ± standard deviation or median [interquartile range].

**Table 2 of the supplementary data.** Post hoc comparisons among the profile groups of focal myocardial replacement fibrosis

| | +LGE MWF | +LGE SECF | −LGE | *P* (ANOVA/Kruskal-Wallis) |
|---|---|---|---|---|
| *Patients* | 23 (29.2) | 19 (24) | 37 (46.8) | n/a |
| *Biochemical analyses* | | | | |
| sST2, ng/mL | 38.5 [37.0-40.2] (*) († *P* = .006) | 31.5 [28.2-35.9] (^) (†) | 18.9 [16.0-22.7] (**P* = .001) (^*P* = .001) | < .001 |
| *CMR parameters* | | | | |
| LVEDD, mm | 51.0 [46.5-55.5] | 50.0 [47.0-55.5] (^) | 47.0 [44.0-50.0] (^*P* = .019) | .007 |
| IVST, mm | 13.0 [12.0-15.0] | 14.0 [12.2-15.0] | 12.0 [11.0-13.0] | .049 |
| EDV index, mL/m$^2$ | 77.0 [70.0-83.0] | 69.0 [56.5-102.0] (^) | 67.0 [54.8-71.2] (^*P* = .037) | .030 |
| ESV index, mL/m$^2$ | 30.0 [14.0-38.0] (*) | 32.0 [20.0-48.0] (^) | 19.0 [10.8-23.0] (**P* = .048) (^*P* = .001) | .002 |
| LV mass index, g/m$^2$ | 97.0 [87.0-128.0] (*) | 90.5 [78.8-107.0] | 84.0 [68.0-93.5] (**P* = .004) | .007 |
| LVEF, % | 61.8 ± 15.4 | 55.6 ± 16.9 (^) | 70.4 ± 9.7 (^ *P* = .001) | .001 |

*, statistical significance between −LGE and +LEG MWF; ^, statistical significance between −LGE and +LEG SECF; †, statistical significance between +LGE MWF and +LEG MWF.

Data are expressed as No. (%), mean ± standard deviation, or median [interquartile range].

**Table 3 of the supplementary data.** Multivariate linear regression model with and without use of the Yeo-Johnson transformation of 'fibrosis' as dependent variable and 'sST2 (ng/mL)' and 'ejection fraction (%)' as independent variables with a l = −0.75

| | Fibrosis | (Fibrosis + 1) ^-0.75)-1)/-0.75 |
|---|---|---|
| Intercept | 13.336 | 0.685 |
| *P*-value [95%CI] | < .001 [6.876-19.796] | .001 [0.279-1.092] |
| sST2, ng/mL | 0.169 | 0.030 |
| *P*-value [95%CI] | .003 [0.061-0.276] | < .001 [0.023-0.037] |
| LVEF, % | −0.220 | −0.016 |
| *P*-value [95%CI] | < .001 [−0.302 to −0.138] | < .001 [−0.021 to −0.010] |

95%CI, 95% confidence interval; LVEF, left ventricle ejection fraction.

**Table 4 of the supplementary data.** Multivariate linear regression model excluding recruited patients with CAD and prior AMI

| | Total | Patients without MI | Patients without CAD |
|---|---|---|---|
| **Intercept** | 13.336 | 10.469 | 4.894 |
| ***P*-value [95%CI]** | <.001 [6.876-19.796] | <.001 [5.404-15.535] | .041 [0.216-9.572] |
| **sST2 ng/mL** | 0.169 | 0.168 | 0.165 |
| ***P*-value [95%CI]** | .003 [0.061-0.276] | <.001 [0.083-0.253] | <.001 [0.083-0.246] |
| **LVEF, %** | −0.220 | −0.186 | −0.107 |
| ***P*-value [95%CI]** | <.001 [−0.302 to −0.138] | <.001 [−0.250 to −0.122] | 0.002 [−0.171 to −0.043] |
| **R2** | 0.437 | 0.517 | 0.458 |
| **R2 Adjusted** | 0.417 | 0.499 | 0.423 |
| **AIC** | 351.2 | 296.7 | 156.7 |
| **BIC** | 359.6 | 304.8 | 162.8 |
| **Log.Lik.** | −171.595 | −144.370 | −74.346 |
| **RMSE** | 4.22 | 3.19 | 2.15 |

95%CI, 95% confidence interval; AMI, acute myocardial infarct; AIC, Akaike information criterion; BIC, Bayesian information criterion; CAD, coronary artery disease; CI, confidence interval; Log.lik, log-likelihood function; LVEF, left ventricle ejection fraction; Num. obs., number of observations; RMSE, root mean squared error.

**Table 5 of the supplementary data.** Internal validity of the multivariate linear regression model using bootstrap sampling (R = 1999)

| | Bootstrap normal CI | | |
|---|---|---|---|
| | Estimate | 2.50% | 97.50% |
| Intercept | 13.336 | 3.970 | 22.400 |
| sST2, ng/mL | 0.168 | 0.073 | 0.264 |
| LVEF, % | −0.219 | −0.335 | −0.100 |

LVEF, left ventricle ejection fraction.