Inflammation and echocardiographic parameters of ventricular hypertrophy in a cohort with preserved cardiac function

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ABSTRACT
Objective: To investigate the association between inflammation and selective echocardiographic parameters (EP) characteristic for ventricular hypertrophy in cross-sectional and longitudinal population-based analyses.

Methods: Baseline (711 men, 659 women: 45–83 years) and 4-year follow-up data (622 men, 540 women) of the prospective, population-based CARdiovascular disease, Living and Ageing in Halle (CARLA) study after exclusion of participants with cardiovascular diseases were analysed. Inflammation parameters: soluble tumour necrosis factor receptor 1 (sTNF-R1), high-sensitivity C reactive protein (hsCRP) and interleukin 6 (IL-6). EPs: left ventricular mass (LVM), left atrial systolic dimension (LADS), interventricular septum diameter (IVSD), posterior wall dimension (PWD), left ventricular diastolic diameter (LVDD), ejection fraction according to Teichholz (EF). For the longitudinal analyses baseline to follow-up differences were considered. Effect sizes were determined by using multiple linear regression and mixed models. Missing values were replaced by means of multiple imputations.

Results: Men had higher sTNF-R1 levels; means of hsCRP and IL-6 were similar in men and women. In multiple regression models, sTNF-R1 was associated with LADS (1.4 mm/1000 pg/mL sTNF-R1, 95% CI 0.6 to 2.1) in men. Respecting confounder hsCRP was associated with LVM (5.2 g/10 mg/L hsCRP, 95% CI 1.6 to 8.8), IVSD (0.2 mm/10 mg/L hsCRP, 95% CI 0 to 0.3) and PWD (0.2 mm/10 mg/L hsCRP, 95% CI 0.1 to 0.3) in women, while there were no relevant effects in analysis of IL-6 in both sexes. The baseline to follow-up change in EPs was not relevantly associated with sTNF-R1, hsCRP or IL-6.

Conclusions: STNF-R1, hsCRP and IL-6 were inadequate predictors for structural changes of the heart at follow-up, while weak cross-sectional associations are restricted to certain EPs and depend on sex.

INTRODUCTION
The prognostic role of cytokines and their corresponding modulators, especially soluble tumour necrosis factor receptor 1 (sTNF-R1) and C reactive protein (CRP), in the development of lethal outcomes of congestive systolic and diastolic heart failure (CHF) has been revealed previously. The value of cytokines in predicting death in patients with myocardial infarction was the subject of further studies. They found that higher levels of sTNF-R1, but not interleukin 6 (IL-6) and CRP, were associated with an increased risk of death. This is in contrast to the findings reported by Tan et al who showed that IL-6 was an independent predictor of cardiovascular mortality. Further cross-sectional studies focused on the relationship between IL-6, sTNF-R1 and CRP, respectively, and left ventricular hypertrophy (LVH) in the general population and in asymptomatic hypertensive patients. The authors revealed a positive relationship between sTNF-R1 and LVH, but failed to find a similar effect for IL-6 and CRP, which was associated with LVH in another collective of asymptomatic participants with essential hypertension. Although there is evidence from experimental studies indicating the pathophysiological role of IL-6 in the development of cardiac hypertrophy the impact of IL-6 on cardiac hypertrophy in humans remains controversial.

Summarising previous findings, sTNF-R1 was suggested to be of prognostic value for the course of disease in cardiac patients. The stability of sTNF-R1 makes it an easily assessable marker of the larger TNF system. Interestingly, the type 1 receptor of TNF-α is the origin of several pathways in the human heart, affecting cell metabolism, apoptosis and remodelling. The soluble form of this receptor is released from its membrane-bound component by different stimuli (eg, TNF-α, lipopolysaccharides), and serves among other things as a ligand to
TNF-α. The shedding of sTNF-R1 is increased in patients with heart failure, myocardial infarction and coronary artery disease.

Although the association between sTNF-R1 and CHF or LVH, respectively, has been examined previously in clinical populations, there is a substantial lack of studies examining the longitudinal association of inflammation with LVH assessed via echocardiographic parameters of ventricular wall thickness, dimensions and ejection fraction (EF) in cardiovascularly healthy participants. Therefore, the objectives of the current study were

1. To analyse the cross-sectional association of inflammation—especially sTNF-R1—with echocardiographic parameters related to ventricular hypertrophy in a cardiovasculary healthy population-based cohort, avoiding possible distractions due to pre-existing cardiac defects;

2. To assess the prognostic value of inflammation for changes in the parameters aforementioned in a 4-year period as a longitudinal analyses in this group of participants.

**METHODS**

**Study design and study population**

We used data from the CARdio-vascular disease, Living and Ageing in Halle (CARLA) study, which is a prospective population-based cohort study of the elderly general population of the city of Halle in eastern Germany. The CARLA cohort comprises 1779 participants (baseline response 64.1%), aged 45–83 years at baseline (967 men and 812 women). The baseline examination took place between December 2002 and January 2006. A multistep recruitment strategy aimed to achieve a high response rate. The percentage final response after subtracting exclusions (individuals who were deceased prior to the invitation, had moved away, or were unable to participate due to illness) was 64%. From March 2007 until March 2010, the first 4-year follow-up examination was performed. The net sample (after exclusion of deceased or non-responding people) then comprised 1436 participants (86% response), consisting of 790 men and 646 women aged between 50 and 87 years. The study participants underwent a detailed medical examination and a standardised, computer-assisted interview, which collected information on sociodemographic and socioeconomic variables, behavioural, biomedical and psychosocial factors, medical history and the use of medication within the preceding 7 days. Medication was automatically coded according to the Anatomical Therapeutic Chemical Classification System (ATC code). Additionally, an analysis of non-respondents was performed in order to assess non-response bias by obtaining information about prevalent diseases, and selected behavioural and sociodemographic factors. A more comprehensive account of the CARLA study can be found elsewhere. In short, CHF was defined as follows: presence of symptoms of CHF (oedema, fatigue and dyspnoea) and an NT-probrain natriuretic peptide (NT-proBNP) above 220 pg/mL or a reduced EF (<50% according to the Teichholz et al’s formula), or a left ventricular (LV) dimension index above 3.8 cm/m² in men and 3.7 cm/m² in women and echocardiographic parameters of diastolic dysfunction, which is in accordance with international guidelines. Additionally, we excluded all participants with a history of myocardial infarction and presence of cardiovascular diseases (history of stroke, history of vascular intervention—self-reported and subsequently physician approved). Thus, the final study sample comprised 711 men (256 excluded) and 659 women (153 excluded) at baseline and 622 men and 540 women at follow-up, while 52 men and 33 women primarily included in the study died between baseline and follow-up.

**Echocardiographic assessment**

At baseline, Doppler echocardiographic examinations were conducted and evaluated by a specially trained and certified physician. At follow-up, echocardiography was performed by a trained and certified study nurse, and subsequently the stored echocardiographic recordings were evaluated by a trained physician. All echocardiographers underwent the same dedicated study certification procedures. Assessing intraobserver variability for the M-mode examinations the mean observer bias varied between 0.3% and 3.8% (2*SD between 15.3% and 27.7%), while the interobserver variability ranged between 0.1% and 2.7% (2*SD between 12.7% and 20.8%). All echocardiographic examinations at baseline and follow-up were performed using the GE Vivid ultrasound system (GE Vivid 4 and 5 at baseline, GE vivid 5 at follow-up). To quantify the LV dimensions and function, we chose echocardiographic parameters (table 1) that are recommended by the guidelines for chamber quantification in echocardiography. We took only dimensional parameters, rather than volume parameters, into account. To calculate the left ventricular mass (LVM), we used the ASE-cube formula, which is in accordance with international guidelines. In addition, the size of the left ventricle was quantified according to the LV diastolic dimension (LVDd). The LV wall thickness was assessed by measuring the diastolic interventricular septum thickness (IVSd) and the diastolic thickness of the posterior wall (PWD). In order to examine the cross-sectional association of inflammation with the myocardial geometry more closely, we differentiated between participants with signs of concentric remodelling and participants without such changes. According to Lang et al concentric remodelling was defined based on the relative posterior wall thickness (RPWD), which was calculated by the formula RPWD=...
# Table 1  Baseline characteristics

| Miscellaneous                  | Baseline | Follow-up |
|--------------------------------|----------|-----------|
|                                | Men      | Women     | Men      | Women     |
|                                | Mean (95% CI) | Mean (95% CI) | Mean (95% CI) | Mean (95% CI) |
| **Age (years)**                | 711 62 (61 to 63) | 659 62 (61 to 63) | 621 66 (65 to 66) | 540 65 [64 to 66] |
| **BSA (m²)**                   | 711 1.98 (1.98 to 1.98) | 659 1.75 (1.74 to 1.77) | 618 1.98 (1.96 to 1.99) | 534 1.75 [1.73 to 1.76] |
| **AMP (mm Hg)**                | 711 106.57 (105.67 to 107.47) | 659 102.01 (101.02 to 103.01) | 619 100.06 (99.12 to 101.01) | 535 97.3 [96.32 to 98.28] |
| **BMI (kg/m²)**                | 711 27.67 (27.47 to 27.87) | 659 27.76 (27.5 to 28.01) | 618 27.79 (27.57 to 28.02) | 534 27.74 [27.44 to 28.06] |
| **LV mass (g)†**               | 691 228.64 (224.49 to 232.86) | 460 220.77 (215.99 to 225.65) | 460 220.77 (215.99 to 225.65) | 425 166.34 [162.71 to 170.05] |
| **PWD (mm)†**                  | 692 11.9 (11.79 to 12.02) | 463 11.1 (10.92 to 11.28) | 463 11.1 (10.92 to 11.28) | 425 10 [9.82 to 10.18] |
| **IVSD (mm)†**                 | 695 11.87 (11.75 to 12) | 462 11.63 (11.44 to 11.81) | 462 11.63 (11.44 to 11.81) | 428 10.57 [10.4 to 10.74] |
| **LAD (mm)†**                  | 683 40.08 (39.73 to 40.43) | 579 40.92 (40.53 to 41.31) | 579 40.92 (40.53 to 41.31) | 506 37.81 [37.41 to 38.21] |
| **LVDd (mm)†**                 | 693 49.39 (48.88 to 49.9) | 464 50.17 (49.66 to 50.68) | 464 50.17 (49.66 to 50.68) | 426 45.85 [45.36 to 46.35] |
| **EF (%)**                     | 690 62.78 (62.16 to 63.4) | 458 65.54 (64.67 to 66.43) | 458 65.54 (64.67 to 66.43) | 422 67.93 [67.03 to 68.84] |
| **Blood parameters**           |          |           |          |           |
| **TNF-R1 (pg/mL)**             | 672 1122.62 (1093.02 to 1153.03) | 614 1021.97 (994.12 to 1050.6) | 619 1.84 (1.71 to 1.99) | 531 1.85 [1.7 to 2.01] |
| **hsCRP (mg/L)**               | 681 1.67 (1.54 to 1.8) | 642 1.85 (1.7 to 2.01) | 619 1.84 (1.71 to 1.99) | 531 1.85 [1.7 to 2.01] |
| **IL-6 (pg/mL)**               | 672 1.99 (1.83 to 2.17) | 614 1.8 (1.65 to 1.97) | 619 1.84 (1.71 to 1.99) | 531 1.85 [1.7 to 2.01] |
| **Creatinine (µmol/L)**        | 708 78.41 (77.33 to 79.52) | 659 63.34 (62.48 to 64.22) | 619 82.37 (80.82 to 83.96) | 531 66.74 [65.66 to 67.83] |
| **HDL (mmol/L)**               | 690 3.11 (3.04 to 3.17) | 656 3.32 (3.25 to 3.39) | 606 3.05 (2.98 to 3.12) | 529 3.36 [3.28 to 3.44] |
| **HbA1c (%)**                  | 708 5.67 (5.61 to 5.73) | 659 5.64 (5.59 to 5.7) | 619 5.79 (5.73 to 5.84) | 531 5.79 [5.73 to 5.84] |
| **Numeric values**             |          |           |          |           |
| **Median n of medication**     | 711 4 | 659 4 | 619 4 | 535 4 |

Means are displayed as geometric means with the respecting 95% confidence limits.

†Adjusted for body surface area.

AMP, Arterial mean pressure; BMI, body mass index; BSA, Body surface area; CRP, high-sensitivity C reactive protein; EF, left ventricular ejection fraction; HDL, high-density lipoproteins; IL, interleukin 6; IVSD, interventricle septum diameter; LAD, left atrial diastolic diameter; LV, left ventricular mass; LVDd, left ventricular end-diastolic dimension; PWD, posterior wall dimension; TNF, soluble tumour necrosis factor-α receptor 1.
(2 × PWD)/LVDD. Finally, an RPWD above 0.42 was considered a concentrically altered LV. The cardiac output as a functional parameter was measured by the EF according to Teichholz et al. We added the systolic dimension of the left atrium (LADS) to our analysis, as this parameter was previously found to be related to cardiac risk.

**Laboratory measurements**

Blood samples were taken after a supine rest of 30 min at baseline and at follow-up. At baseline the parameters of sTNF-R1, high-sensitivity CRP (hsCRP) and IL-6 were assessed, while hsCRP was measured at follow-up as well. STNF-R1 and IL-6 were analysed by the Department of Medicine III, University Clinics Halle (Saale). After 10 min centrifugation (20°C, 1500 rpm, Acc=9, Dcc=3), the plasma was collected and stored at −80°C. Cytokines were assessed using commercially available sandwich ELISAs (IL-6, Opteia, BD Biosciences, Heidelberg, Germany; TNF-R1, Boehringer Mannheim, Mannheim, Germany).

The determination of hsCRP was undertaken by the Institute of Laboratory Medicine, Clinical Chemistry and Molecular Diagnostics at the Leipzig University Clinics. The laboratory has been accredited according to the accreditation norms ISO 15180 and ISO 17025. Serum hsCRP levels were measured using a high-sensitivity immunoturbidimetric method (CRP (Latex) HS, Roche, Mannheim, Germany) on a Hitachi autoanalyser (Roche Diagnostics Mannheim, Germany).

**Statistical methods**

All analyses were performed separately for men and women. Descriptive results are displayed as geometric means to illustrate the distribution of echocardiographic and inflammation parameters in the study population.

To assess the association of sTNF-R1, hsCRP and IL-6 (each as an independent variable) with all six echocardiographic parameters in a multivariate approach we used mixed models with an unstructured covariance matrix respecting echocardiographic parameters as a fixed effect. These analyses were separately conducted for sTNF-R1, hsCRP and IL-6, men and women, and the cross-sectional and longitudinal analyses, respectively (statistical results given in tables 2–4). Parameters were entered in the mixed model after standardisation of all included dependent and independent parameters (mean=0, SD=1) making the association across all echocardiographic parameters comparable between analyses. In addition, single regression coefficients and respective 95% CIs (association of each inflammation parameter with each echocardiographic parameter) were calculated by means of univariate linear robust regression models. Models were adjusted for arterial mean pressure, body surface area, age, medication, glycated haemoglobin (HbA1c), low-density lipoprotein and creatinine clearance. As medication, we considered antihypertensive drugs (ATC codes: C02/C03/C07/C08/C09), cardiac glycosides (ATC code: C01A) and lipid-lowering medication (ATC code: C10). In the longitudinal regression analysis, we used the difference in echocardiographic values between baseline and follow-up as outcome without consideration of baseline echocardiographic values as a further covariate. The idea beyond such a longitudinal analysis is to check for associations of a change in one parameter over time (from baseline to follow-up), depending on an explanatory variable, which influences the course of the dependent parameter. Possible differences in the blood level of inflammatory parameters between participants with signs of a concentrically altered LV wall thickness compared with participants without such signs were checked by performing a Mann-Whitney U test. Echocardiographic parameters adjusted for body surface area are specified in the descriptive statistics. Finally, the adequacy of the considered regression models was assumed when the residuals were normally distributed, which was tested via a Q-Q plot and Cook’s distance, which was required to be below one (achieved for all conducted tests).

Missing values were replaced using the method of multiple imputation, which was previously found to cause little bias (see online supplementary appendix for further details). Deviations of the results between multiple imputations and the complete case analysis were compared in a sensitivity analysis, which is given in the appendix. A further sensitivity analysis was performed after exclusion of participant with the presence of self-reported and physician evaluated chronic diseases related to inflammation (cancer, rheumatoid arthritis, gout, liver disease, chronic kidney disease with a glomerular filtration rate below 30 mL/min).

The level of significance was taken as α=0.05, and consequently we report the 95% confidence limits. All statistical analyses and data management were performed using SAS, V.9.3 (SAS Inc, Cary, North Carolina, USA).

**RESULTS**

**Study population**

In the final sample, the mean age at baseline of male and female participants was 62 years (95% CI 61 to 63; table 1). We observed a higher EF but lower echocardiographic dimension and LV wall thickness parameters in women compared to men. While men showed a higher concentration of plasma sTNF-R1 (table 1), no considerable differences in plasma levels of hsCRP and IL-6 between sexes could be observed. At follow-up, our data indicated a decrease in LVM and an increase in EF, while there were no relevant changes in the mean of further echocardiographic parameters.

**Cross-sectional association of inflammation and echocardiographic parameters**

Soluble tumour necrosis factor receptor 1

Most of the participants had blood levels of sTNF-R1 between 500 and 2000 pg/mL, although 5.65% of men
| sTNF-R1 | Core | Adjusted | Core | Adjusted | Core | Adjusted |
|--------|------|----------|------|----------|------|----------|
|       | β    | (95% CI) | p Value | β    | (95% CI) | p Value | β    | (95% CI) | p Value |
|       |      |          |        |       |          |        |       |          |        |
| Cross-sectional* | 0.11 (0.07 to 0.14) | <0.0001 | 0.06 (0.02 to 0.1) | 0.0038 | 0.15 (0.1 to 0.2) | <0.0001 | 0.04 (0.01 to 0.09) | 0.0998 |
| LVM (g)  | 13.8 (5.6 to 21.9) | 0.0011 | 5.7 (−2.7 to 14) | 0.1820 | 22.8 (14 to 31.6) | <0.0001 | 3.5 (−4.5 to 11.5) | 0.3932 |
| PWD (mm) | 0.6 (0.4 to 0.8) | <0.0001 | 0.3 (0 to 0.5) | 0.0231 | 1 (0.8 to 1.3) | <0.0001 | 0.3 (0.1 to 0.6) | 0.0121 |
| IVSD (mm) | 0.6 (0.3 to 0.8) | <0.0001 | 0.2 (−0.1 to 0.4) | 0.1614 | 1 (0.7 to 1.3) | <0.0001 | 0.3 (0 to 0.6) | 0.0267 |
| LADS (mm) | 2.1 (1.3 to 2.8) | <0.0001 | 1.4 (0.6 to 2.1) | 0.0010 | 2.5 (1.7 to 3.4) | <0.0001 | 0 (−0.9 to 0.8) | 0.9755 |
| LVDD (mm) | 0 (−0.8 to 0.8) | 0.9796 | 0.2 (−0.7 to 1) | 0.6835 | 0 (−0.9 to 0.9) | 0.9925 | −0.7 (−1.6 to 0.2) | 0.1532 |
| EF (%) | −0.3 (−1.4 to 0.9) | 0.6442 | −0.1 (−1.4 to 1.2) | 0.8885 | 0.5 (−0.8 to 1.7) | 0.4666 | 0.6 (−0.9 to 2.1) | 0.4003 |
| Longitudinal* | −0.44 (−0.92 to 0.03) | 0.0675 | −0.02 (−0.08 to 0.04) | 0.4641 | 0.18 (−0.34 to 0.7) | 0.4873 | 0.44 (−0.14 to 1.02) | 0.1320 |
| LVM (g)  | −8.1 (−19.3 to 3.1) | 0.1568 | −3.3 (−16.4 to 9.8) | 0.6181 | −1 (−11 to 9.1) | 0.8455 | 5.8 (−6.1 to 17.7) | 0.3361 |
| PWD (mm) | −0.4 (−0.8 to 0.1) | 0.1031 | −0.2 (−0.7 to 0.3) | 0.4765 | −0.3 (−0.8 to 0.2) | 0.3043 | −0.1 (−0.7 to 0.5) | 0.6642 |
| IVSD (mm) | −0.1 (−0.6 to 0.3) | 0.6007 | 0.1 (−0.4 to 0.7) | 0.5619 | −0.3 (−0.8 to 0.2) | 0.2469 | 0 (−0.6 to 0.6) | 0.9177 |
| LADS (mm) | 0.1 (−0.8 to 1) | 0.8342 | 0.1 (−0.9 to 1.1) | 0.8406 | 0.6 (−0.3 to 1.5) | 0.2162 | 0.7 (−0.4 to 1.7) | 0.2073 |
| LVDD (mm) | −0.2 (−1.4 to 1.1) | 0.7971 | −0.3 (−1.7 to 1.1) | 0.6880 | 1.5 (0.1 to 2.8) | 0.0340 | 1.7 (0.1 to 3.3) | 0.0357 |
| EF (%) | −1.3 (−3.4 to 0.7) | 0.2084 | −1.7 (−4.1 to 0.6) | 0.1501 | −0.5 (−3.1 to 2) | 0.6742 | −1 (−3.9 to 1.9) | 0.4931 |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters. Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, LDL and creatinine clearance.

*grey-shaded line* = results from mixed models estimating the association of inflammation parameter across all six echocardiographic parameters in the cross-sectional, and longitudinal analyses, respectively. Estimates were standardised (mean=0, SD=1).

EF, ejection fraction; HbA1c, glycated haemoglobin; IL-6, interleukin 6; IVSD, interventricle septum diameter; LADS, left atrial diastolic diameter; LVDD, left ventricular end-diastolic dimension; LDL, low-density lipoprotein; LVM, left ventricular mass; PWD, posterior wall dimension; sTNF-R1, soluble tumour necrosis factor-α receptor 1.
|                  | Core                  | Adjusted               | Core                  | Adjusted               |
|------------------|-----------------------|------------------------|-----------------------|------------------------|
|                  | β (95% CI)            | p Value                | β (95% CI)            | p Value                |
| Cross sectional*| 0.02 (−0.03 to 0.06)  | 0.5024                 | 0.02 (−0.03 to 0.06)  | 0.4821                 |
| LVM (g)          | 1 (−8.7 to 10.6)      | 0.8442                 | −1.9 (−10.2 to 6.3)   | 0.6449                 |
| PWD (mm)         | 0.2 (0 to 0.5)        | 0.0800                 | 0.1 (−0.2 to 0.3)     | 0.5006                 |
| IVSD (mm)        | 0.2 (−0.1 to 0.5)     | 0.1788                 | 0.1 (−0.2 to 0.3)     | 0.5734                 |
| LADS (mm)        | 0.7 (−0.2 to 1.7)     | 0.1156                 | 0.4 (−0.4 to 1.3)     | 0.3073                 |
| LVDD (mm)        | −0.2 (−1.1 to 0.7)    | 0.6415                 | −0.4 (−1.3 to 0.5)    | 0.3904                 |
| EF (%)           | −0.3 (−1.6 to 1.1)    | 0.6807                 | −0.3 (−1.6 to 1.1)    | 0.6846                 |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters.

Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, LDL and creatinine clearance. *(grey shaded line)=results from mixed models estimating the association of inflammation parameter across all six echocardiographic parameters in the cross-sectional, and longitudinal analyses, respectively. Estimates were standardised (mean=0, SD=1).

hsCRP, high-sensitivity C reactive protein; EF, ejection fraction; IL-6, interleukin 6; IVSD, interventricle septum diameter; LADS, left atrial diastolic diameter; LVDD, left ventricular end-diastolic dimension; LDL, low-density lipoprotein; LVM, left ventricular mass; PWD, posterior wall dimension.
| IL-6 | Men | Women |
|------|-----|-------|
|      | Core | Adjusted | p Value | Core | Adjusted | p Value | Adjusted | p Value |
|      | β (95% CI) | p Value | β (95% CI) | p Value | β (95% CI) | p Value | β (95% CI) | p Value |
| Cross sectional* | −0 (−0.05 to 0.05) | 0.9471 | −0.01 (−0.05 to 0.04) | 0.7432 | 0.08 (−0.02 to 0.17) | 0.1031 | 0.07 (−0.01 to 0.14) | 0.0834 |
| LVM (g) | 2.1 (−2.3 to 6.5) | 0.3321 | −0.3 (−1.9 to 1.2) | 0.6782 | 2 (−1.5 to 5.5) | 0.2516 | 0.3 (−3 to 3.7) | 0.8377 |
| PWD (mm) | 0.1 (0 to 0.2) | 0.0761 | 0 (0 to 0.2) | 0.0794 | 0.1 (0 to 0.3) | 0.0956 | 0.1 (0 to 0.2) | 0.1962 |
| IVSD (mm) | 0.1 (0 to 0.2) | 0.1178 | 0 (−0.1 to 0.2) | 0.5380 | 0.1 (−0.1 to 0.2) | 0.2309 | 0.1 (0 to 0.1) | 0.1552 |
| LADS (mm) | 0.3 (−0.1 to 0.6) | 0.0984 | 0.1 (−0.1 to 0.3) | 0.5914 | 0.2 (−0.3 to 0.6) | 0.4621 | 0.2 (−0.2 to 0.5) | 0.2764 |
| LVDD (mm) | −0.1 (−0.5 to 0.3) | 0.4617 | −0.1 (−0.5 to 0.3) | 0.7001 | −0.1 (−0.3 to 0.2) | 0.6394 | −0.1 (−0.4 to 0.1) | 0.2281 |
| EF (%) | −0.1 (−0.6 to 0.3) | 0.5474 | −0.3 (−0.5 to 0) | 0.0468 | 0 (−0.7 to 0.8) | 0.9634 | 0 (−0.4 to 0.7) | 0.6011 |
| Longitudinal* | 0.05 (−0.46 to 0.55) | 0.8498 | 0.01 (−0.04 to 0.06) | 0.7329 | −0.67 (−1.44 to 0.11) | 0.0915 | −0.06 (−0.14 to 0.01) | 0.1070 |
| LVM (g) | −3.2 (−8.3 to 1.9) | 0.2067 | −1.7 (−7.4 to 4) | 0.5384 | −0.1 (−4.8 to 4.5) | 0.9562 | −0.9 (−3.7 to 1.9) | 0.5319 |
| PWD (mm) | −0.2 (−0.3 to 0) | 0.1101 | −0.1 (−0.4 to 0.1) | 0.2244 | −0.1 (−0.2 to 0.1) | 0.2689 | −0.1 (−0.2 to 0.1) | 0.2506 |
| IVSD (mm) | 0 (−0.2 to 0.2) | 0.8388 | 0.1 (−0.1 to 0.2) | 0.3519 | 0 (−0.3 to 0.2) | 0.6943 | −0.1 (−0.2 to 0.1) | 0.3989 |
| LADS (mm) | 0 (−0.4 to 0.3) | 0.7980 | 0 (−0.1 to 0.2) | 0.6791 | −0.1 (−0.7 to 0.4) | 0.6510 | −0.4 (−0.7 to 0) | 0.0656 |
| LVDD (mm) | −0.1 (−0.3 to 0.2) | 0.5102 | −0.1 (−0.3 to 0.1) | 0.4871 | 0.1 (−0.2 to 0.5) | 0.4453 | 0.1 (−0.2 to 0.5) | 0.5017 |
| EF (%) | 0 (−0.8 to 0.8) | 0.9729 | 0.2 (−0.6 to 1) | 0.6496 | 0 (−0.7 to 0.7) | 0.9795 | 0 (−0.6 to 0.7) | 0.9485 |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters. Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, LDL and creatinine clearance.

*(grey-shaded line)=results from mixed models estimating the association of inflammation parameter across all six echocardiographic parameters in the cross-sectional, and longitudinal analyses, respectively. Estimates were standardised (mean=0, SD=1).

EF, ejection fraction; HbA1c, glycated haemoglobin; IL-6, interleukin 6; IVSD, interventricle septum diameter; LADS, left atrial diastolic diameter; LVDD, left ventricular end-diastolic dimension; LDL, low-density lipoprotein; LVM, left ventricular mass; PWD, posterior wall dimension.
and 3.09% of women showed even higher values (see online supplementary figure A1). The estimates obtained from mixed models (covariate adjusted) respecting all six echocardiographic parameters indicated an overall association of sTNF-R1 with echocardiography in men, but not in women. The univariate analysis of echocardiographic values as a function of inflammatory parameters revealed a relevant unadjusted association of sTNF-R1 with LVM, IVSD, PWD and LADS (table 2). Nevertheless, after the models were adjusted for the considered covariates, the regression coefficients decreased considerably. A medium effect size remained only when LADS (1.4 mm/1000 pg/mL sTNF-R1, 95% CI 0.6 to 2.1) was the outcome in men, while the association of sTNF-R1 with PWD is of minor clinical relevance. The regression analysis of EF as a functional parameter revealed no relevant association with sTNF-R1 (see online supplementary figure A2). Consistent with the findings of the regression analysis of LVDD and PWD, the differentiation between concentric remodelling and ordinary/eccentric myocardial tissue (figures 1 and 2, see the online supplementary appendix for the results across both sexes) indicated that men and women with signs of concentric alterations had considerably higher blood levels of sTNF-R1.

High-sensitivity C reactive protein

The majority of the study population had plasma hsCRP levels below 10 mg/L (see online supplementary figure A1). A general association of hsCRP with echocardiographic parameters could only be found in women, which was mainly due to low statistical variance (0.03, 95% CI 0.01 to 0.05), while the estimate itself was lower than in the case of sTNF-R1. The multivariate analysis revealed lower effect estimates in men than in women. When covariates were taken into account, hsCRP was positively associated with LVM (5.2 g/10 mg/L hsCRP, 95% CI 1.6 to 8.8), PWD (0.2 mm/10 mg/L hsCRP, 95% CI 0.1 to 0.3) and IVSD (0.2 mm/10 mg/L hsCRP, 95% CI 0 to 0.3) in female participants, though the estimates

Figure 1  Box plot comparing inflammation parameters in men with (RPWD<0.42) and without (RPWD>0.42) signs of concentric hypertrophy. Lower and upper whiskers display the 2% and 98% quintiles, respectively. Notches display the 95% CI of the median (shortened horizontal line). The box represents participants between 25% and 75% quintiles. p Values refer to group comparison (RPWD<0.42 and RPWD>0.42) by means of a Mann-Whitney U test. hsCRP, high-sensitivity C reactive protein; IL-6, interleukin 6; RPWD, relative posterior wall dimension of the left ventricle; sTNF-R1, soluble tumour necrosis factor-α receptor 1.
appeared distinctly lower than in the unadjusted analyses (table 3). However, the effects were weak, which is especially obvious when the lower confidence limit is considered. Again, participants with signs of concentric remodelling had a higher CRP level in both sexes (figures 1 and 2).

Interleukin 6
Distributions of IL-6 and hsCRP showed a similar pattern with mainly low blood levels (see online supplementary figure A1). According to the large CIs of the regression analysis, the IL-6 levels were not related to the considered structural parameters (table 4) in men and women. After covariate adjustment the estimates declined, leading to negligible effect sizes. In the same line, IL-6 was not notably associated with the functional parameter of EF in men or women. Participants with echocardiographic signs of concentric hypertrophy and without such indications showed almost identical IL-6 blood levels (figures 1 and 2).

Longitudinal association of inflammation and echocardiographic parameters
In the longitudinal analysis, we found a slight association of sTNF-R1 and LVDD in women in the univariate (1.5 mm/1000 pg/mL, 95% CI 0.1 to 2.8) and multivariate models (1.7 mm/1000 pg/mL, 95% CI 0.1–3.3), which was much lower in men. However, this finding might be due to multiple testing, as there was no overall association of sTNF-R1 with the change of echocardiographic parameters when the results of the mixed models are taken into account. All other regression analyses of change in echocardiographic parameters depending on sTNF-R1, IL-6 and hsCRP revealed only minimal, clinically negligible associations (tables 2–4).

Sensitivity analyses
The performance of a complete case analysis led to higher effect estimates and significant results in few instances (see online supplementary tables A1–3), which might be mainly due to bias driven by missing values.
and outliers. The exclusion of participants with chronic diseases associated with inflammation was not related to relevant alterations of effect estimates (see online supplementary tables A4–6).

**DISCUSSION AND LIMITATIONS**

We found minor associations of plasma sTNF-R1 with PWD and LADS in men, and IVSD in women, while plasma hsCRP was slightly associated with LVM, PWD and IVSD in female participants. In the longitudinal analyses no relevant associations could be deduced from our data. In summary this reflects only a weak association of systemic inflammation as measured by plasma levels with structural echocardiographic parameters of ventricular hypertrophy and atrial size. However, LVM and the size of the left atrium were both reported as being independently associated with hospitalisation and death, respectively. Owing to the exclusion of participants with major cardiac diseases, these associations might be driven by an endogenous inflammation that was independent of myocardial performance and blood pressure, but was probably due to causes such as age, infectious diseases or chronic inflammatory diseases.

The observed unadjusted cross-sectional association between cardiac mass and sTNF-R1 in men, which was weaker in women, is partly consistent with Takei et al. However, the inclusion of possible confounders led to a decrease in effect estimates in our study, which might be due to the different ethnic and social background of our collective and, thus, a different impact of confounders. Our unadjusted results also agree with the findings of Roselló-Lletí et al who described the sTNF-R1 as the most distinctive factor associated with LVH among various blood values related to inflammation (TNF, IL-6, interleukin-1ra, sTNF-R1) in a cohort of asymptomatic hypertensive patients, nevertheless estimates in our study were again lower after covariate adjustment and not associated with LVH, but LADS in men. The weak associations found in the longitudinal analysis are explainable by short-term effects of sTNF-R1, which were previously reported in survival analyses. However, as sTNF-R1 is related to a wide range of inflammation parameters, further inflammatory mechanisms and interactions are likely to be substantial in humans. We found no relevant correlation of sTNF-R1 with the functional parameter of EF. It is possible that the heart might sustain its function by adapting to alterations in the myocardial pattern. Additionally, the EF is characterised by high intrarater and inter-rater variability which limits the significance of EF as a parameter for statistical analyses. We failed to confirm additional associations of sTNF-R1 in the longitudinal analyses and, thus, we could not attribute further predictive value to sTNF-R1.

In women, we found a minimal association of hsCRP with structural echocardiographic parameters, which might reflect a general association of systemic inflammation with cardiac hypertrophy. Using ECG, Bo et al has found evidence that cardiac hypertrophy is a ‘status of inflammation’ causing increased hsCRP levels, which is consistent with our findings as we failed to find considerable longitudinal associations. Differences in estimates and, more importantly, statistical accuracy between men and women is in contrast to previous cross-sectional findings where similar effect sizes between male and female hypertension patients were found. It is likely that in our collective further factors such as kidney function and blood pressure (table 1) have a greater impact on ventricular hypertrophy in men than systemic inflammation represented by plasma hsCRP levels.

The associations of echocardiographic parameters representative for LVH with IL-6 were only minimal, and thus, a considerable association of IL-6 with cardiac hypertrophy was not present in our data, which is consistent with the findings in Takei et al.

Comparing echocardiographic parameters between baseline and follow-up we found an apparent improvement at follow-up. This finding might be affected by a coincident improvement in cardiovascular health and most importantly blood pressure (see table 1) reflecting a possible intervention effect due to study participation.

**Limitations**

Apart from our analyses, further unmeasured inflammation parameters might also be associated with cardiac structure and function. The long interval from baseline to follow-up might conceal short-term effects, which were found in mouse models. In addition, the inter-rater and intrarater variability of echocardiography might have contributed to inconsistent results. In this context, differences in measurement methods between baseline and follow-up can be due to different examinations at baseline and follow-up, could have biased the results. Nevertheless, to minimise observer bias, a quality assurance process was implemented during data acquisition and reading, including certification of the echocardiographic examination and reader certification. In addition, all echocardiographic examinations were supervised by a senior cardiologist. Concerning inflammation parameters only hsCRP was measured at follow-up, which limits the ability to assess the change in inflammation parameters in more detail. Despite the effort to adjust for potential confounders, there may be residual confounding factors that were not taken into account. From the clinical point of view, pathological findings in echocardiography without symptoms might not be as relevant as actual diseases, but as complex maladies are often difficult to objectify, we focused on these subclinical echocardiographic values. Finally, despite the attempt to treat missing values adequately in the statistical analysis, we cannot fully exclude a weakening or disruption of results because of missing values.
In conclusion we found a minor cross-sectional association of plasma sTNF-R1 (in both sexes) and plasma hsCRP (in women) with echocardiographic parameters in the general elderly population. Additionally, there are subtle indications of a longitudinal association of sTNF-R1, and hsCRP or IL-6 with LADS. Further studies that survey the changes in cardiac parameters in relation to inflammation, mainly sTNF-R1, over different time intervals and give further insights into the role of inflammation in biochemical processes of heart tissue are now required.35

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Contributors

DM wrote the article and performed all statistical analyses; he carries the responsibility for data analyses and is also the corresponding author. SD, AK, HGG, HT, SN, MR, DT, KW, JH and AF contributed essentially to the organisation of the study and data acquisition. Furthermore AK, HG, HT, DT, JH and KW revised the manuscript critically.

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Competing interests

None.

Ethics approval

The study was approved by the Ethics Committee of the Medical Faculty of the Martin-Luther-University Halle-Wittenberg, and conforms to the principles outlined in the Declaration of Helsinki.

Provenance and peer review

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Appendix

We imputed missing values for all considered echocardiographic and inflammatory parameters and the covariates taken into account (see Table 1 in the main text). For each regression analysis, we took 20 separate imputations of one imputation model into account; that is, missing values were simulated 20 times and all of these simulations were included in the regression analysis. In order to assess the reliability of the multiple imputation used, we analysed differences in the statistical analysis within several regression analyses obtained by the same multiple imputation model. The differences in estimates and confidence intervals within each regression analysis (each consisting of 20 imputations) were all smaller than 0.1% and, thus, indicated the sufficient reliability of this method. In order to obtain normally distributed variables, we transformed non-normally distributed variables using the Box-Cox transformation or a logarithmic transformation.

To apply the method of multiple imputation, we used the procedures PROC MI and PROC MIANALYZE, both implemented in SAS®, Version 9.3. The statistical analysis itself was conducted using PROC REG and PROC GPLOT, respectively.

Figures:

Figure A1: Distribution of plasma levels of sTNF-R1, IL-6, and CRP in the CARLA cohort.

Abbreviation: sTNF-R1: Soluble tumour necrosis factor-alpha receptor 1; hsCRP: High-sensitivity C-reactive protein; IL-6: Interleukin 6; CARLA: Cardiovascular Disease, Living and Ageing in Halle (CARLA) study.

Figure A2: Scatterplot of the cross-sectional analysis of EF in relation to plasma sTNF-R1 in men (blue) and women (red).

Abbreviation: EF: Ejection fraction; sTNF-R1: Soluble tumour necrosis factor-alpha receptor 1. Blue line represents the slope of the regression line in men (bright dashed blue lines illustrate 95% confidence intervals); red line represents the slope of the regression line in women (bright dashed red lines illustrate 95% confidence intervals)

Figure A3: Scatterplot of the cross-sectional analysis of LVM in relation to plasma sTNF-
R1 in men (blue) and women (red).

Abbreviation: LVM: Left ventricular mass; sTNF-R1: Soluble tumour necrosis factor-alpha receptor 1. Blue line represents the slope of the regression line in men (bright dashed blue lines illustrate 95% confidence intervals); red line represents the slope of the regression line in women (bright dashed red lines illustrate 95% confidence intervals)

**Figure A4: Scatterplot of the longitudinal analysis of EF in relation to plasma sTNF-R1 in men (blue) and women (red).**

Vertical axis displays the absolute baseline to follow-up difference in EF. Abbreviation: EF: Ejection fraction; sTNF-R1: Soluble tumour necrosis factor-alpha receptor 1. Blue line represents the slope of the regression line in men (bright dashed blue lines illustrate 95% confidence intervals); red line represents the slope of the regression line in women (bright dashed red lines illustrate 95% confidence intervals)

**Figure A5: Scatterplot of the longitudinal analysis of LVM in relation to plasma sTNF-R1 in men (blue) and women (red).**

Vertical axis displays the absolute baseline to follow-up difference in LVM. Abbreviation: LVM: Left ventricular mass; sTNF-R1: Soluble tumour necrosis factor-alpha receptor 1. Blue line represents the slope of the regression line in men (bright dashed blue lines illustrate 95% confidence intervals); red line represents the slope of the regression line in women (bright dashed red lines illustrate 95% confidence intervals)

**Figure A6: Box plot comparing inflammation parameters in all subjects (men and women) with (RPWD <0.42) and without (RPWD >0.42) signs of concentric hypertrophy.**

Lower and upper whiskers display the 2% and 98% quintiles, respectively. Notches display the 95% confidence interval of the median (shortened horizontal line). The box represents subjects between the 25% and 75% quintile. P-values refer to group comparison (RPWD <0.42 and RPWD >0.42) by means of a Mann-Whitney U test. Abbreviation: RPWD: Relative posterior wall dimension of the left ventricle.
Figure A1:

Men

Women

![Graphs showing percentages of subjects for sTNF-R1 (pg/mL), IL-6 (pg/mL), and CRP levels in men and women.](image)
Figure A2:

![Figure A2](image1)

Figure A3:

![Figure A3](image2)
Figure A4:

Figure A5:
Figure A6:
Table A1: Estimates of the complete case regression analysis of sTNF-R1

| sTNF-R1               | Men                  |     | Women                  |     |
|-----------------------|----------------------|-----|------------------------|-----|
|                       | Core                 | Adjusted   | Core                  | Adjusted   |
|                       | Beta [95% CI]        | p  | Beta [95% CI]        | p  |
| Cross-sectional       |                      |     |                        |     |
| LVM [g]               | 12 [3.9, 20.1]       | 0.0035 | 2.7 [-5, 10.4]       | 0.4894 | 22.7 [14.3, 31.2] | <.0001 | 0.6 [-7.2, 8.4] | 0.8831 |
| PWD [mm]              | 0.6 [0.3, 0.8]       | <.0001 | 0.2 [-0.1, 0.4]      | 0.1402 | 1.1 [0.8, 1.4]   | <.0001 | 0.2 [0, 0.5]    | 0.0982 |
| IVSD [mm]             | 0.5 [0.3, 0.8]       | <.0001 | 0.1 [-0.1, 0.3]      | 0.3795 | 1 [0.8, 1.3]     | <.0001 | 0.3 [0, 0.5]    | 0.0557 |
| LADS [mm]             | 2.2 [1.5, 2.9]       | <.0001 | 1.1 [0.4, 1.9]       | 0.0030 | 2.6 [1.7, 3.4]   | <.0001 | -0.3 [-1.1, 0.6]| 0.5607 |
| LVDD [mm]             | -0.1 [-0.8, 0.7]     | 0.8507 | 0 [-0.8, 0.8]        | 0.9480 | -0.1 [-0.9, 0.8] | 0.8999 | -0.7 [-1.6, 0.2]| 0.1239 |
| EF [%]                | -0.2 [-1.3, 0.8]     | 0.6554 | 0 [-1.2, 1.3]        | 0.9575 | 0.6 [-0.7, 1.8]  | 0.3647 | 0.3 [-1.1, 1.8] | 0.6486 |
| Longitudinal          |                      |     |                        |     |
| LVM [g]               | -3.9 [-14.2, 6.5]    | 0.4678 | -1.8 [-13.3, 9.7]    | 0.7579 | 0.2 [-9.6, 10]   | 0.9695 | 5.7 [-4.8, 16.2]| 0.2886 |
| PWD [mm]              | -0.2 [-0.6, 0.3]     | 0.4490 | -0.1 [-0.6, 0.4]     | 0.6158 | -0.1 [-0.7, 0.4] | 0.6231 | 0.1 [-0.5, 0.7]| 0.6694 |
| IVSD [mm]             | 0.2 [-0.3, 0.6]      | 0.4479 | 0.4 [-0.1, 0.9]      | 0.1177 | -0.2 [-0.7, 0.3] | 0.3687 | 0 [-0.5, 0.5]   | 0.9911 |
| LADS [mm]             | 0.2 [-0.6, 1.1]      | 0.5845 | 0.2 [-0.7, 1.2]      | 0.6401 | 0.7 [-0.2, 1.6]  | 0.1140 | 0.8 [-0.2, 1.8] | 0.1040 |
| LVDD [mm]             | -0.5 [-1.6, 0.6]     | 0.3511 | -0.6 [-1.8, 0.7]     | 0.3705 | 1.6 [0.2, 2.9]   | 0.0202 | 1.5 [0, 3]      | 0.0479 |
| EF [%]                | -1.3 [-3.3, 0.7]     | 0.2065 | -2 [-4.2, 0.2]       | 0.0759 | -0.4 [-2.8, 2.1] | 0.7655 | -0.2 [-2.9, 2.6]| 0.9025 |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters. Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, low-density lipoprotein (LDL), and creatinine clearance. Abbreviation: sTNF-R1: Soluble tumour necrosis factor-alpha receptor 1; LVM: Left ventricular mass, PWD: Posterior wall dimension, IVSD: Inter-ventricle septum diameter, LADS: Left atrial diastolic diameter, LVDD: Left ventricular end-diastolic dimension, EF: Left ventricular ejection fraction.
### Table A2: Estimates of the complete case regression analysis of hsCRP

| hsCRP       | Men                                  | Women                                |
|-------------|--------------------------------------|--------------------------------------|
|             | Core Adjusted                        | Core Adjusted                        |
|             | Beta [95% CI] p                      | Beta [95% CI] p                      |
| LVM [g]     | 1.1 [-8.6, 10.8] 0.8205 [-6.7, 7.6] | 0.8917 13 [4.2, 21.8] 0.0037 5.6 [2.1, 9.2] 0.0019 |
| PWD [mm]    | 0.3 [0, 0.5] 0.0478 0.1 [0.1, 0.3] | 0.6 [0.3, 0.9] 0.0002 0.2 [0.1, 0.3] 0.0006 |
| IVSD [mm]   | 0.2 [-0.1, 0.5] 0.1396 0.1 [-0.2, 0.4] | 0.4456 0.5 [0.2, 0.8] 0.0003 0.2 [0.1, 0.3] 0.0038 |
| LADS [mm]   | 0.7 [-0.1, 1.6] 0.0974 0.4 [-0.3, 1.2] | 0.2611 0.7 [0.2, 1.1] 0.0035 0.3 [-0.1, 0.7] 0.1819 |
| LVDD [mm]   | -0.3 [-1.2, 0.6] 0.5628 -0.3 [-1.2, 0.5] | 0.4304 0.2 [-0.3, 0.6] 0.5182 -0.1 [-0.5, 0.3] 0.6197 |
| EF [%]      | -0.3 [-1.6, 1] 0.6933 -0.4 [-1.7, 0.9] | 0.5551 -0.2 [-0.9, 0.4] 0.5155 -0.3 [-1, 0.3] 0.3437 |
| LVM [g]     | 0.6 [-9.7, 11] 0.9033 0 [-10.6, 10.7] | 0.9951 -3 [-12.8, 6.8] 0.5510 -0.9 [-11.1, 9.3] 0.8608 |
| PWD [mm]    | -0.3 [-0.8, 0.1] 0.1713 -0.3 [-0.7, 0.2] | 0.2429 -0.2 [-0.7, 0.4] 0.5613 -0.1 [-0.7, 0.5] 0.6872 |
| IVSD [mm]   | 0 [-0.4, 0.5] 0.9485 0 [-0.5, 0.4] | 0.8978 0 [-0.5, 0.5] 0.8642 0.1 [-0.4, 0.6] 0.7725 |
| LADS [mm]   | 0.9 [0, 1.8] 0.0617 0.7 [-0.2, 1.6] | 0.1218 1.3 [0.3, 2.2] 0.0075 1.1 [0.2, 2.1] 0.0197 |
| LVDD [mm]   | 0.7 [-0.4, 1.8] 0.2045 0.6 [-0.5, 1.7] | 0.2728 0 [-1.4, 1.4] 0.9782 0 [-1.4, 1.4] 0.9498 |
| EF [%]      | -2 [-4, 0] 0.0552 -1.8 [-3.8, 0.3] | 0.0937 -0.2 [-2.9, 2.4] 0.8817 0 [-2.7, 2.8] 0.9722 |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters. Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, low-density lipoprotein (LDL), and creatinine clearance.

Abbreviation: hsCRP: High-sensitivity C-reactive protein, LVM: Left ventricular mass, PWD: Posterior wall dimension, IVSD: Inter-ventricle septum diameter, LADS: Left atrial diastolic diameter, LVDD: Left ventricular end-diastolic dimension, EF: Left ventricular ejection fraction.
Table A3: Estimates of the complete case regression analysis of IL-6

| IL-6 | Men | | | Women | | |
|------|-----|-----|-----|-------|-----|-----|
|      | Core | Adjusted | Core | Adjusted |
|      | Beta [95% CI] | p | Beta [95% CI] | p | Beta [95% CI] | p |
| Cross-sectional | | | | | | |
| LVM [g] | 3.7 [0.4, 7] | 0.0300 | -0.6 [-2.1, 1] | 0.4777 | 3.1 [0.3, 5.8] | 0.0270 | 1.6 [-0.5, 3.7] | 0.1329 |
| PWD [mm] | 0.1 [0.2] | 0.215 | 0.2 [0.1, 0.2] | 0.0005 | 0.2 [0.1, 0.3] | 0.0001 | 0.1 [0.1, 0.2] | 0.0004 |
| IVSD [mm] | 0.2 [0.1, 0.3] | 0.0002 | 0.2 [0.1, 0.3] | 0.0006 | 0.1 [0.2] | 0.0114 | 0.1 [0.1] | 0.0560 |
| LADS [mm] | 0.4 [0.1, 0.8] | 0.0162 | 0.4 [0.1, 0.7] | 0.0592 | 0.2 [-0.3, 0.6] | 0.4391 | 0.4 [0.1, 0.6] | 0.0024 |
| LVDD [mm] | -0.3 [-0.6, 0] | 0.0959 | -0.5 [-0.8, -0.2] | 0.0033 | -0.1 [-0.3, 0.2] | 0.5911 | -0.1 [-0.4, 0.1] | 0.2391 |
| EF [%] | -0.3 [-0.5, 0] | 0.0345 | -0.2 [-0.5, 0] | 0.0736 | 0 [-0.6, 0] | 0.9448 | 0.3 [-0.1, 0.7] | 0.1682 |
| Longitudinal | | | | | | |
| LVM [g] | -4.7 [-9.6, 0.1] | 0.0574 | -7.2 [-13.4, -1] | 0.0231 | -1.8 [-4.3, 0.6] | 0.1489 | -1.9 [-4.4, 0.6] | 0.1367 |
| PWD [mm] | -0.1 [-0.3, 0] | 0.1292 | -0.2 [-0.4, 0] | 0.0351 | -0.1 [-0.3, 0] | 0.1226 | -0.1 [-0.2, 0] | 0.1651 |
| IVSD [mm] | 0 [-0.2, 0.2] | 0.9041 | -0.1 [-0.4, 0.2] | 0.4882 | -0.1 [-0.2, 0] | 0.0843 | -0.1 [-0.2, 0] | 0.1579 |
| LADS [mm] | -0.1 [-0.5, 0.2] | 0.4454 | 0.1 [-0.1, 0.2] | 0.5370 | -0.2 [-0.7, 0.3] | 0.3523 | -0.6 [-0.8, -0.3] | <0.0001 |
| LVDD [mm] | -0.4 [-0.8, 0.1] | 0.1197 | -0.1 [-0.3, 0.1] | 0.3216 | 0.1 [-0.3, 0.4] | 0.6065 | 0 [-0.3, 0.4] | 0.8429 |
| EF [%] | 0 [-0.8, 0.8] | 0.9694 | -0.3 [-1.3, 0.7] | 0.5247 | -0.1 [-0.8, 0.5] | 0.6916 | -0.1 [-0.7, 0.6] | 0.8607 |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters. Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, low-density lipoprotein (LDL), and creatinine clearance. Abbreviation: IL-6: Interleukin 6, LVM: Left ventricular mass, PWD: Posterior wall dimension, IVSD: Inter-ventricle septum diameter, LADS: Left atrial diastolic diameter, LVDD: Left ventricular end-diastolic dimension, EF: Left ventricular ejection fraction.
Table A4: Estimates of regression analysis of sTNF-R1 (after exclusion of subjects with cancer, rheumatoid arthritis, gout, liver disease, and chronic kidney disease with a glomerular filtration rate below 30 mL/min)

| sTNF-R1 | Men | | | Women | | |
|---------|-----|-----|-----|-------|-----|-----|
|         | Core | Adjusted | | Core | Adjusted | |
|         | Beta [95% CI] | p | Beta [95% CI] | p | Beta [95% CI] | p | Beta [95% CI] | p |
| Cross-sectional | | | | | | | | |
| LVM [g] | 16.4 [6.6, 26.3] | 0.0012 | 6 [-3.8, 15.8] | 0.2309 | 22.4 [11.8, 33] | <.0001 | 1 [-8.4, 10.4] | 0.8323 |
| PWD [mm] | 0.7 [0.4, 0.9] | <.0001 | 0.3 [0, 0.6] | 0.0464 | 1 [0.7, 1.4] | <.0001 | 0.3 [0, 0.6] | 0.0834 |
| IVSD [mm] | 0.6 [0.3, 0.9] | 0.0002 | 0.1 [-0.2, 0.4] | 0.3774 | 1.1 [0.7, 1.4] | <.0001 | 0.4 [0, 0.7] | 0.0434 |
| LADS [mm] | 2 [1.1, 2.9] | <.0001 | 1.1 [0.1, 2] | 0.0253 | 2.9 [1.8, 4] | <.0001 | 0.2 [-0.8, 1.2] | 0.6889 |
| LVDD [mm] | 0.4 [-0.5, 1.4] | 0.3374 | 0.2 [-0.8, 1.2] | 0.6747 | -0.1 [-1.2, 0.9] | 0.8393 | -1.1 [-2.2, 0] | 0.0553 |
| EF [%] | 0.2 [-1.1, 1.6] | 0.7570 | 0.2 [-1.3, 1.8] | 0.7561 | 0.1 [-1.4, 1.7] | 0.8575 | 0.7 [-1.2, 2.5] | 0.4698 |
| Longitudinal | | | | | | | | |
| LVM [g] | -14.9 [-28.5, -1.2] | 0.0327 | -10.5 [-25.8, 4.8] | 0.1775 | -4.3 [-17.1, 8.4] | 0.5017 | 2.7 [-12.1, 17.5] | 0.7196 |
| PWD [mm] | -0.6 [-1.1, 0] | 0.0545 | -0.4 [-1, 0.2] | 0.2103 | -0.2 [-0.8, 0.4] | 0.5579 | 0 [-0.7, 0.7] | 0.9324 |
| IVSD [mm] | -0.3 [-0.8, 0.3] | 0.3242 | 0 [-0.6, 0.6] | 0.9839 | -0.4 [-1.1, 0.3] | 0.2641 | -0.2 [-0.9, 0.5] | 0.5640 |
| LADS [mm] | 0.2 [-0.8, 1.2] | 0.7216 | 0.3 [-0.8, 1.4] | 0.5912 | 0.1 [-0.9, 1.2] | 0.8282 | 0.2 [-0.9, 1.4] | 0.6854 |
| LVDD [mm] | -0.5 [-2.1, 1.1] | 0.5333 | -0.6 [-2.3, 1.2] | 0.5343 | 1.4 [-0.4, 3.1] | 0.1184 | 2 [0, 4] | 0.0557 |
| EF [%] | -0.4 [-2.8, 2.1] | 0.7713 | -0.8 [-3.6, 1.9] | 0.5543 | 0.3 [-2.6, 3.2] | 0.8299 | -0.5 [-3.7, 2.7] | 0.7493 |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters.

Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, low-density lipoprotein (LDL), and creatinine clearance.

Abbreviation: sTNF-R1: Soluble tumour necrosis factor-alpha receptor 1; LVM: Left ventricular mass, PWD: Posterior wall dimension, IVSD: Inter-ventricle septum diameter, LADS: Left atrial diastolic diameter, LVDD: Left ventricular end-diastolic dimension, EF: Left ventricular ejection fraction
Table A5: Estimates of the regression analysis of hsCRP (after exclusion of subjects with cancer, rheumatoid arthritis, gout, liver disease, and chronic kidney disease with a glomerular filtration rate below 30 mL/min)

| hsCRP  | Men                  |                          | Women                     |                          |
|--------|----------------------|--------------------------|---------------------------|--------------------------|
|        | Core                 | Adjusted                 | Core                      | Adjusted                 |
|        | Beta [95% CI]        | p                        | Beta [95% CI]             | p                        |
| Cross-sectional |                      |                          |                           |                          |
| LVM [g]       | 1.3 [-10.6, 13.3]    | 0.8257                   | -0.7 [-10.5, 9.1]         | 0.8938                   |
| PWD [mm]     | 0.2 [-0.2, 0.6]      | 0.3198                   | 0 [-0.3, 0.3]             | 0.8342                   |
| IVSD [mm]    | 0.1 [-0.2, 0.5]      | 0.4472                   | 0 [-0.3, 0.4]             | 0.8195                   |
| LADS [mm]    | 0.6 [-0.6, 1.8]      | 0.3032                   | 0.4 [-0.7, 1.4]           | 0.4859                   |
| LVDD [mm]    | 0.1 [-1.1, 1.2]      | 0.9290                   | -0.1 [-1.2, 1]            | 0.8825                   |
| EF [%]       | -0.8 [-2.4, 0.7]     | 0.2943                   | -0.9 [-2.6, 0.7]          | 0.2541                   |
| Longitudinal |                      |                          |                           |                          |
| LVM [g]       | -1.2 [-16, 13.6]     | 0.8717                   | -1.2 [-16.3, 13.9]        | 0.8770                   |
| PWD [mm]     | -0.3 [-0.9, 0.2]     | 0.2458                   | -0.3 [-0.9, 0.2]          | 0.2641                   |
| IVSD [mm]    | -0.2 [-0.7, 0.4]     | 0.6079                   | -0.2 [-0.7, 0.4]          | 0.5866                   |
| LADS [mm]    | 0.3 [-0.8, 1.4]      | 0.5576                   | 0.2 [-0.9, 1.3]           | 0.6792                   |
| LVDD [mm]    | 0.6 [-1, 2.2]        | 0.4693                   | 0.6 [-1, 2.2]             | 0.4750                   |
| EF [%]       | -1.6 [-4.1, 0.8]     | 0.1958                   | -1.6 [-4, 0.9]            | 0.2012                   |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters.
Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, low-density lipoprotein (LDL), and creatinine clearance.
Abbreviation: hsCRP: High-sensitivity C-reactive protein, LVM: Left ventricular mass, PWD: Posterior wall dimension, IVSD: Inter-ventricle septum diameter, LADS: Left atrial diastolic diameter, LVDD: Left ventricular end-diastolic dimension, EF: Left ventricular ejection fraction
Table A6: Estimates of the regression analysis of IL-6 (after exclusion of subjects with cancer, rheumatoid arthritis, gout, liver disease, and chronic kidney disease with a glomerular filtration rate below 30 mL/min)

| IL-6       | Men                                     | Women                                   |
|------------|-----------------------------------------|-----------------------------------------|
|            | Core                                    | Adjusted                                | Core                                    | Adjusted                                |
|            | Beta [95% CI]                           | p                                       | Beta [95% CI]                           | p                                       |
| Cross-sectional |                                             |                                          |                                             |                                          |
| LVM [g]    | 1.2 [-3.4, 5.9]                         | 0.5936                                  | -0.4 [-2.7, 2]                          | 0.7667                                  | 0.8 [-3.1, 4.6]                         | 0.6923                                  | -1.2 [-4.3, 1.8]                         | 0.4238                                  |
| PWD [mm]   | 0.1 [-0.1, 0.2]                         | 0.3169                                  | 0 [-0.1, 0.2]                          | 0.7915                                  | 0.1 [-0.1, 0.2]                         | 0.2835                                  | 0 [0.1, 0.1]                            | 0.7607                                  |
| IVSD [mm]  | 0 [-0.1, 0.2]                           | 0.4432                                  | 0 [-0.1, 0.1]                          | 0.8824                                  | 0.1 [-0.1, 0.2]                         | 0.4044                                  | 0 [-0.1, 0.2]                            | 0.8196                                  |
| LADS [mm]  | 0.3 [-0.1, 0.6]                         | 0.1862                                  | 0.1 [-0.2, 0.3]                        | 0.5756                                  | 0.2 [-0.2, 0.6]                         | 0.3342                                  | 0.1 [-0.3, 0.4]                          | 0.7062                                  |
| LVDD [mm]  | 0 [-0.3, 0.3]                           | 0.9732                                  | 0.1 [-0.1, 0.2]                        | 0.5427                                  | -0.1 [-0.5, 0.3]                        | 0.6252                                  | -0.2 [-0.5, 0.1]                         | 0.2374                                  |
| EF [%]     | -0.3 [-0.7, 0]                          | 0.0685                                  | -0.3 [-0.6, -0.1]                      | 0.0174                                  | -0.1 [-0.8, 0.5]                        | 0.6999                                  | -0.1 [-0.8, 0.5]                         | 0.6825                                  |
| Longitudinal |                                             |                                          |                                             |                                          |
| LVM [g]    | -3.1 [-8.8, 2.6]                        | 0.2807                                  | -2 [-8.2, 4.2]                         | 0.5111                                  | 0.7 [-4.5, 4.5]                         | 0.7762                                  | 0.9 [-4.5, 8.4]                          | 0.7083                                  |
| PWD [mm]   | -0.1 [-0.3, 0.1]                        | 0.4502                                  | 0 [-0.2, 0.2]                          | 0.8504                                  | 0 [-0.3, 0.2]                          | 0.7137                                  | 0 [-0.3, 0.2]                           | 0.7334                                  |
| IVSD [mm]  | 0 [-0.1, 0.2]                           | 0.6522                                  | 0.1 [0, 0.2]                           | 0.0700                                  | 0 [-0.3, 0.2]                          | 0.7448                                  | 0 [-0.3, 0.2]                           | 0.9052                                  |
| LADS [mm]  | -0.1 [-0.5, 0.3]                        | 0.6413                                  | 0 [-0.2, 0.3]                          | 0.8681                                  | -0.1 [-0.6, 0.3]                        | 0.5374                                  | -0.1 [-0.6, 0.3]                         | 0.5266                                  |
| LVDD [mm]  | -0.2 [-0.7, 0.3]                        | 0.3753                                  | -0.1 [-0.4, 0.1]                       | 0.3313                                  | 0.2 [-0.4, 0.8]                         | 0.4646                                  | 0.2 [-0.4, 0.8]                          | 0.5282                                  |
| EF [%]     | 0.1 [-0.8, 1]                           | 0.8152                                  | 0.3 [-0.5, 1]                          | 0.4844                                  | 0.3 [-0.7, 1.3]                         | 0.5514                                  | 0.3 [-0.7, 1.3]                          | 0.4969                                  |

Estimates of the longitudinal analyses mean the absolute baseline to follow-up difference of echocardiographic parameters. Adjusted for arterial mean pressure, body surface area, age, medication, HbA1c, low-density lipoprotein (LDL), and creatinine clearance. Abbreviation: IL-6: Interleukin 6, LVM: Left ventricular mass, PWD: Posterior wall dimension, IVSD: Inter-ventricle septum diameter, LADS: Left atrial diastolic diameter, LVDD: Left ventricular end-diastolic dimension, EF: Left ventricular ejection fraction.