Atrial Structure and Function

Left Atrial Volumes and Function by Three-Dimensional Echocardiography
Reference Values, Accuracy, Reproducibility, and Comparison With Two-Dimensional Echocardiographic Measurements

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Background—Our study sought to (1) identify reference values for left atrial (LA) volumes and phasic function indices by 3-dimensional echocardiography (3DE) and compare them with those measured by 2-dimensional echocardiography (2DE) and (2) analyze their relationship with age, sex, body size, and left ventricular function. Accuracy and reproducibility of 3DE and 2DE have been also tested to evaluate the robustness of our data.

Methods and Results—We obtained maximal, minimal, and preA LA volumes by 3DE and 2DE in 276 healthy volunteers (18–79 years; 57% women). Limits of normality for LA volumes and total LA emptying fraction were larger with 3DE than with 2DE (maximal LA volume: 43 versus 35 mL/m²; preA LA volume: 31 versus 25 mL/m²; minimal LA volume: 18 versus 14 mL/m²; 53 versus 48%, respectively; $P<0.001$). 3DE LA volumes indexed by body surface area were similar in men and women and increased with age. On multivariable analysis, age, weight, and left ventricular systolic and diastolic function indices resulted as correlates of LA 3DE indices. LA volumes were tightly correlated with cardiac magnetic resonance measurements, yet more underestimated by 2DE versus 3DE (bias±SD: −17±16 versus −7±15 mL, respectively). Among all LA parameters, maximal LA volume and total emptying fraction were the most reproducible, including at test-retest and at expert versus trainee comparisons.

Conclusions—This study provides reference values for LA 3DE volumes and function from a relatively large cohort of healthy subjects with a wide age range. Our data may help clinicians to identify LA remodeling and dysfunction. (***Circ Cardiovasc Imaging***. 2016;9:e004229. DOI: 10.1161/CIRCIMAGING.115.004229.)

Key Words: left atrial function ■ left atrial volumes ■ left atrium ■ normal values ■ reference values ■ three-dimensional echocardiography

Left atrial (LA) size is a powerful prognostic marker in a variety of clinical conditions.[1] For estimating LA size by 2-dimensional echocardiography (2DE), the current guidelines recommend the calculation of maximum LA volume (Vmax) using either the disk summation (Simpson rule) or the area–length biplane algorithms.[2] However, both algorithms are heavily dependent on correct positioning and angulation of imaging planes and on geometric assumptions about LA geometry. Indeed, 2DE significantly underestimates LA volumes, in comparison with cardiac computed tomography[3] or cardiac magnetic resonance (CMR).[4]

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Because 3-dimensional echocardiography (3DE) does not imply any geometric assumption, it provides a more accurate[5,6] and reproducible[7] measurement of LA volumes than 2DE. Moreover, 3DE LA volume offers an incremental capability to predict cardiovascular events compared with 2DE.[8,9] Thus, 3DE has the potential to become the primary technique to assess LA size and function in clinical practice, pending the availability of robust reference values. At present, reference data of 3DE LA volumes and phasic function are limited.[2,10]

Accordingly, we designed this study to (1) identify the reference values for 3DE LA volumes and phasic function indices and compare them with those measured by 2DE and (2) analyze the effects of age, body size, sex, and left ventricular (LV) function on 3DE LA volumes. As secondary aims,

to validate our 3DE data, we aimed to assess (3) the accuracy and (4) the variability of LA measurement by 3DE and 2DE.

Methods

Study Design

To achieve aims 1 and 2, we have prospectively screened for eligibility 346 healthy volunteers among hospital employees, fellows in training, their relatives, and people who underwent medical visits for driving or working license. The inclusion and exclusion criteria are listed in Figure 1. During enrollment, we aimed to include at least 20 men and 20 women per decade to achieve a fairly uniform age and sex distribution, yet the fulfillment of this condition for 1 decade did not represent an exclusion criterion.

To achieve aim 3, we enrolled consecutive patients with wide ranges of LA volumes and function, undergoing clinically indicated CMR and echocardiography less than 24 hours apart. Exclusion criteria included known contraindications to CMR (pacemaker or defibrillator implantation, atrial arrhythmias, and claustrophobia).

To achieve aim 4, single-beat and multibeat (either 4 or 6) LA data sets have been acquired in sequence (ie, without changing probe position, gain, depth or sector size), from 15 consecutive subjects. For test–retest variability, a second multibeat LA data set has been acquired by the same operator at the end of the examination (21±5 minutes after the first 3DE LA acquisition and after repositioning the patient from supine to left lateral position).

The study was approved by local Ethics Committee (protocol 2380P, approved on October 6, 2011), and all volunteers signed an informed consent. Body surface area (BSA) was calculated according to DuBois and DuBois formula.[11]

Echocardiographic Acquisition

All healthy subjects underwent a comprehensive 2DE and Doppler study using Vivid E9 (GE Vingmed, Horten, Norway) equipped with M5S probe. Apical 4- and 2-chamber views dedicated for LA quantification were acquired. Image optimization to avoid LA foreshortening was done by maximizing LA length and base in each view, so that the difference between LA lengths in the 2 apical views was minimal (<10%).[2] In addition, we acquired 3 apical views (4- and 2-chamber views and long-axis view) dedicated for LV and optimized for global longitudinal strain (GLS) analysis.[2,12]

Finally, 2 separate multibeat (4 or 6) 3DE data sets optimized for LA and for LV were acquired using a 4 V matrix-array transducer

Figure 1. Study enrollment flow chart.

from the apical approach. Care was taken to encompass the entire LV or LA cavity in the data sets, to optimize temporal resolution (by reducing depth and volume size), and to avoid artifacts.[12]

Echocardiographic Analysis

2DE LA volumes were measured using the biplane disk summation method at end systole (just before the mitral valve opening), at pre A (the frame at the peak of the P wave on the ECG), and at end diastole (just after mitral valve closure).[2] While tracing the LA endocardium, we excluded the LA appendage and the ostia of pulmonary veins, if visible. When dropout was encountered, a straight line joining the closest visualized structures completed the atrial outline.[2] Mitral annulus was considered as LA atrioventricular border.

Image quality of the 3DE LA data sets was judged visually, considering the signal-to-noise ratio and presence of dropouts or artifacts and was categorized on a 4-level scale from excellent to poor. 3DE LA analysis was performed by a single experienced observer (M.H.M.), unaware of the 2DE calculations, using a validated software dedicated for LA (4D LA Analysis; Tomtec Imaging Systems, Unterschleissheim, Germany).[5] The frames corresponding to the maximum (Vmax), minimum (Vmin), and pre A (VpreA) volumes were identified by operator using the same criteria described for 2DE. LA data set alignment was performed to obtain nonforeshortened views of the LA in all 3 apical views (Figure 2). In each apical view, the LA blood–tissue interface depicted on high-contrast/low-brightness images (to minimize intracavitary artifacts and endocardial blurring) was manually initialized by tracing the endocardium on the frames identifying LA Vmax and Vmin. For each consecutive frame, the voxel count inside the 3DE LA surface was used to measure LA volumes, resulting in a smooth interpolated time–volume curve (Figure 3) from which Vmax, Vmin, and VpreA were automatically calculated by a software.

From the LA volumes, we calculated (1) the total emptying volume (EV) (Vmax−Vmin), (2) the passive EV (Vmax−VpreA), and (3) the active EV (VpreA−Vmin).[13] Then, we computed the total emptying fraction (EF=total EV/Vmax), the passive EF (passive EV/Vmax), and the active EF (active EV/VpreA) as indexes of reservoir, conduit, and pump function, respectively. Finally, the LA expansion index, an index of LV reservoir function, was calculated as ([Vmax−Vmin]/Vmin)×100%.

Transmitral peak velocities during early (E) and late (A) diastole and the E-wave deceleration time were measured using pulsed-wave Doppler echocardiography. Peak myocardial velocities during systole (s) and early (e') and late (a') diastole were measured by tissue Doppler at the medial and lateral annulus level in the apical 4-chamber view, and septal, lateral, and average E/e' were calculated.

LV GLS was analyzed from the 3 LV apical views, and LV volumes were measured using a commercially available software (EchoPAC BT12, GE Vingmed, Horten, Norway).[12]

CMR Image Acquisition and Analysis

All CMR imaging studies were acquired using a 1.5 Tesla (Siemens AVANTO, Erlangen, Germany). Cine images were obtained by using end-expiratory, breath-hold, ECG-gated, balanced, steady-state, free precession sequences from short-axis orientation from the atrioventricular ring to the base of the atria (slice thickness 6 mm with no gap).[14] Volumetric analysis was performed with commercially available software (CMR 42, Circle, Canada) independently by 2 investigators experienced in CMR (M.P.M. and X.J.) who were blinded to echocardiographic results. On short axes, the LA boundary was manually traced in every slice at end systole and at end diastole. The most distal slice was defined as the slice that showed the LA cavity with no >50% of the circumference surrounded by LV myocardial tissue.[5] LA volumes were calculated using the disk summation method and were used as reference for comparison with the 2DE and 3DE measurements.
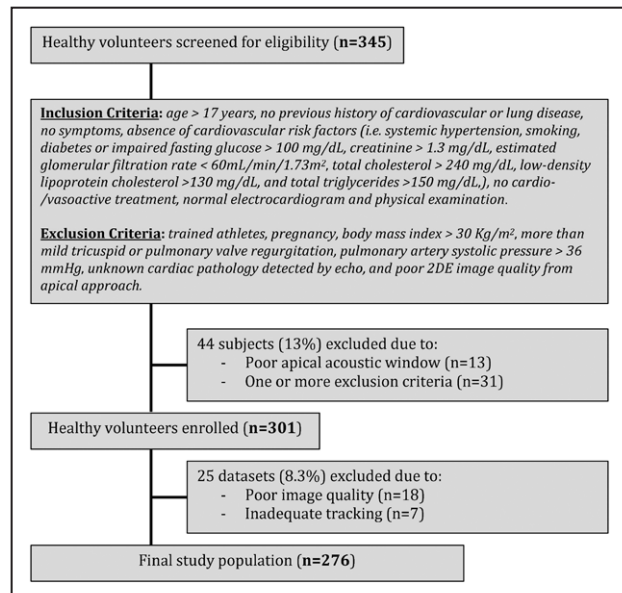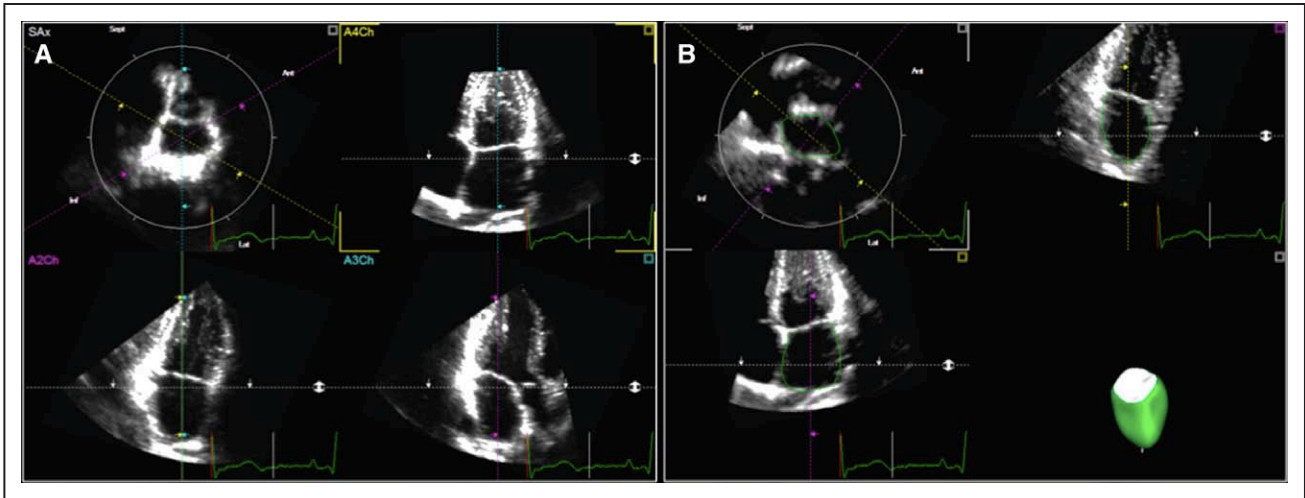
Figure 2. Three-dimensional echocardiography (3DE) quantification of left atrial (LA) volumes and function. **A**, The nonforeshortened LA at end diastole, realigned so that the 3 apical views (A4Ch, A2Ch, and A3Ch) share the same longitudinal axis, which passes through the LA cavity center (SAx). **B**, The endocardial contouring and the 3DE LA cast reconstruction.

Reproducibility Analysis

Intraobserver variability was tested on 15 good-quality data sets by an experienced researcher who reanalyzed the same beat twice (2 weeks apart and blinded from the first measurements). For the interobserver variability, the same images were analyzed independently by a different experienced researcher. Apical 4- and 2-chamber images from the same 15 subjects were also analyzed for assessing 2DE observer variability. Test–retest variability and single-beat versus multibeat variability and expert versus trainee comparisons (having several years versus 1 week training in 3DE LA measurements achieved in the same research laboratory) comparison were tested in LA data sets of different image quality obtained from 15 consecutive subjects.

Statistical Analysis

Normal distribution of variables was assessed by Kolmogorov–Smirnov test.

For aims 1 and 2, demographic and echocardiographic variables were summarized using 25th, 50th, and 75th quantiles. Wilcoxon rank-sum statistic was used to test differences between sexes or methods. Spearman rank correlation was used to analyze the relationships
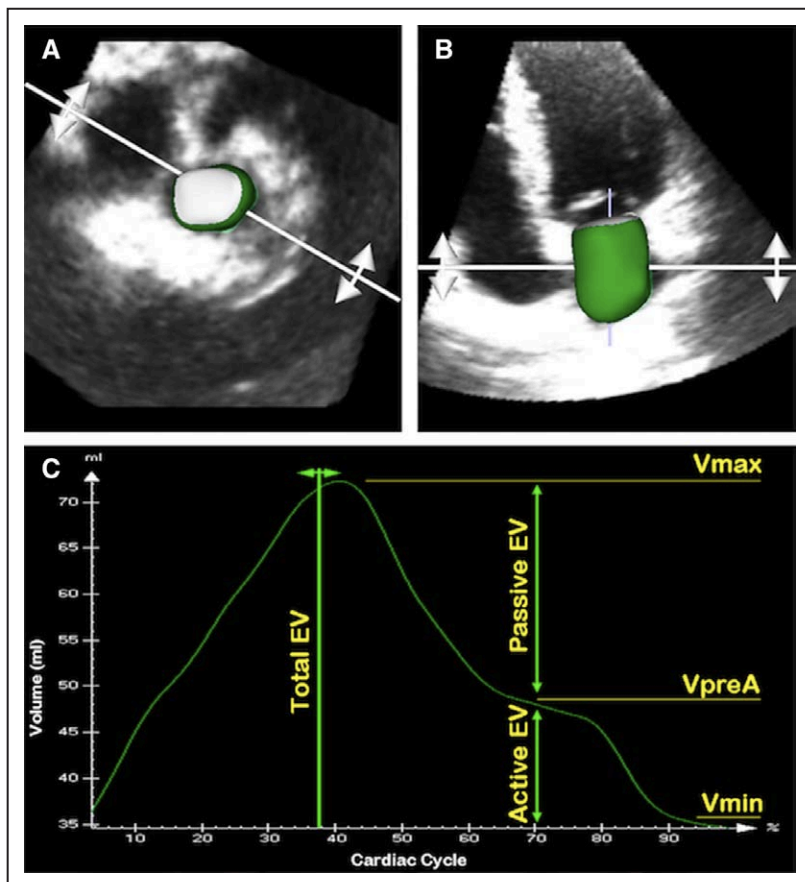
Figure 3. Dynamic changes of left atrial (LA) size and function during the cardiac cycle. Transversal (**A**) and longitudinal (**B**) visualization of three-dimensional echocardiography (3DE) LA volume, and (**C**) time–volume curve depicting LA volume changes during the cardiac cycle, with the computation of phasic emptying volumes (EV).

between 3DE LA volumes and phasic function indices with demographic and LV function variables. The study sample was divided into 10-year age subgroups according to sex, in order to develop reference limits for 3DE LA volumes and phasic function indices. Linear quantile regression was used to estimate the 2.5th, 50th, and 97.5th quantiles for each LA measure. Upper and lower limits of normalcy were defined as 97.5th and 2.5th percentiles, respectively. Differences between the age groups were tested using the nonparametric Kruskal–Wallis test.

Multivariable linear regression analyses were performed to identify the physiological correlates of 3DE LA indices. The multivariate model included the following covariates: weight, E, A, E/A, e' average, E/e' average, S septal, LV volumes, and GLS (having P values <0.10 on bivariate analysis) plus age and sex, with backward selection using likelihood ratio test ($P<0.10$ for selected variables).

For aims 3 and 4, agreement between 2DE and 3DE versus CMR volumes and reproducibility of LA 2DE and 3DE volumes were analyzed using Pearson correlation and Bland–Altman method.

Analyses were performed using SPSS 21.0 (SPSS, Inc, Chicago, IL). A P value <0.05 was considered statistically significant.

Results

The final study sample included 276 healthy subjects; feasibility of 3DE LA volumes quantification was 92% (Figure 1). Temporal resolution of 3DE LA data sets was 42±18 volumes per second. The image quality of the LA data sets was interpreted as optimal in 75% (excellent=21% and good=54%), whereas the remaining data sets had a suboptimal image quality (fair=20% and poor=5%). Mean duration of 3DE LA volumes analysis ranged from 4±1 minute in optimal quality data sets to 7±2 minutes in suboptimal quality data sets (requiring manual editing in multiple frames).

Reference Values of LA 3DE Measurements (Aim 1)

The cohort of healthy volunteers (age range: 18–79 years) had a fairly uniform distribution between sexes (Table 1) and across age groups (Table 2). Age and heart rate were similar in men and women, whereas body size and blood pressure were smaller in women than in men (Table 1).

Reference values for LA volumes and phasic function indices are summarized by sex and age group in Table 2. Men had larger LA volumes than women, but these differences disappeared after indexation by BSA. Overall, women had larger LA total EF, LA passive EV and EF, and LA expansion index than men, albeit these differences were small and not consistently seen in all age groups.

All LA 3DE volumes and function variables, except LA passive EV, were significantly different across age groups. Irrespective of sex, all indexed LA volumes increased significantly with age, with VpreA (+47%) increasing more than Vmax (+28%) and Vmin (+20%). Although LA passive EV did not change with ageing, LA active EV increased, resulting in an age-related decrease of passive EF counterbalanced

Table 1.   Demographic and Echocardiographic Characteristics of the Healthy Subjects

Variable	Overall (n=276) Median (p25; p75)	Men (n=119) Median (p25; p75)	Women (n=157) Median (p25; p75)
Age, y	44 (32; 56)	44 (32; 58)	44 (33; 57)
Height, cm	170 (163; 177)	178 (172; 181)	165 (172; 181)*
Weight, kg	67 (59; 75)	75 (72; 81)	60 (55; 67)*
Body mass index, kg/m²	23 (21; 25)	24 (22; 26)	22 (20; 24)*
Body surface area, m²	1.8 (1.6; 1.9)	1.9 (1.8; 2.0)	1.7 (1.6; 1.8)*
Heart rate, bpm	66 (60; 75)	66 (60; 75)	66 (60; 74)
Systolic blood pressure, mm Hg	120 (110; 130)	130 (120; 140)	118 (110; 125)*
Diastolic blood pressure, mm Hg	70 (70; 80)	80 (70; 80)	70 (65; 80)*
3DE LV diastolic volume, mL/m²	58 (52; 65)	62 (55; 70)	56 (51; 62)*
3DE LV systolic volume, mL/m²	21 (19; 24)	23 (20; 27)	20 (18; 22)*
3DE LV stroke volume, mL/m²	37 (33; 42)	38 (34; 43)	36 (33; 40)†
3DE LV ejection fraction, %	63 (60; 66)	62 (59; 65)	64 (62; 67)*
E, cm/s	79 (69; 91)	74 (65; 89)	85 (71; 92)†
A, cm/s	60 (51; 71)	59 (51; 68)	60 (51; 62)
E/A	1.3 (1.1; 1.6)	1.3 (0.92; 1.6)	1.4 (1.1; 1.6)
E/e' average	6.0 (5.2; 7.1)	6.0 (5.1; 6.8)	6.1 (5.3; 7.3)
s' septal, cm/s	8 (7; 9)	9 (8; 9)	8 (7; 9)†
s' lateral, cm/s	11 (9; 13)	12 (10; 13)	10 (9; 12)†
s' average, cm/s	9.5 (8.5; 11)	10 (9; 12)	10 (8; 11)†

Values are reported as median (p25: 25th percentile; p75: 75th percentile). A indicates transmitral peak velocity during late diastole; 3DE, 3-dimensional echocardiography; E, transmitral peak velocity during early diastole; e', peak myocardial velocity during early diastole; LV, left ventricular; and S, peak myocardial velocity during systole.

*$P<0.0001$ for sex differences.

†$P≤0.01$ for sex differences.

Table 2. Reference Ranges of Left Atrial Volumes and Function by 3-Dimensional Echocardiography Presented by Sex and Age Groups

	All (W=157; M=119)	18–29 y (W=24; M=20)	30–39 y (W=33; M=25)	40–49 y (W=40; M=27)	50–59 y (W=29; M=21)	60–69 y (W=26; M=20)	≥70 y (W=5; M=6)	P Value
Vmax, mL/m²								
Women	31 (27; 45)	28 (26; 31)	31 (26; 35)*	30 (26; 35)	33 (27; 36)	32 (28; 36)†	37 (31; 41)	0.074
Men	31 (19; 52)	30 (25; 32)	25 (21; 30)	30 (26; 37)	31 (26; 35)	39 (35; 43)	36 (28; 42)	<0.0001
Overall	31 (26; 35)	29 (26; 32)	28 (24; 33)	30 (26; 35)	32 (27; 35)	35 (32; 39)	37 (29; 41)	<0.0001
VpreA, mL/m²								
Women	18 (10; 30)	15 (12; 17)	15 (13; 18)	16 (14; 19)	19 (17; 22)	19 (17; 23)†	23 (21; 27)	<0.0001
Men	18 (9; 32)	15 (12; 18)	14 (12; 17)	18 (16; 21)	19 (16; 23)	27 (22; 29)	21 (20; 23)	<0.0001
Overall	17 (14; 21)	15 (12; 17)	14 (13; 18)	16 (14; 20)	19 (17; 22)	22 (18; 27)	22 (19; 27)	<0.0001
Vmin, mL/m²								
Women	10 (5; 18)	9 (8; 12)	9 (8; 11)	9 (8; 11)	10 (9; 13)	11 (9; 13)†	14 (10; 15)	0.029
Men	11 (4; 21)	11 (8; 12)	9 (7; 10)	10 (9; 12)	10 (9; 13)	15 (13; 17)	11 (10; 15)	<0.0001
Overall	10 (8; 12)	10 (8; 12)	9 (7; 10)	10 (8; 12)	10 (9; 13)	12 (10; 15)	12 (11; 15)	<0.0001
Total EV, mL/m²								
Women	21 (14; 30)	19 (18; 21)	21 (18; 23)*	21 (17; 23)	22 (17; 24)	21(17; 24)‡	25 (20; 25)	0.156
Men	20 (13; 35)	19 (18; 19)	16 (14; 20)	19 (17; 24)	21 (19; 23)	24 (19; 26)	24 (17; 29)	<0.0001
Overall	20 (17; 23)	19 (17; 20)	19 (16; 23)	20 (17; 23)	21 (18; 23)	22 (18; 24)	25 (19; 27)	0.001
Passive EV, mL/m²								
Women	14 (7; 21)‡	14 (12; 16)	14 (12; 17)*	14 (11; 17)	14 (10; 15)	12 (9; 14)	14 (7; 16)	0.023
Men	13 (8; 25)	13 (12; 15)	11 (9; 14)	12 (10; 15)	13 (11; 14)	13 (10; 15)	12 (7; 15)	0.605
Overall	13 (11; 15)	14 (12; 16)	13 (11; 16)	13 (11; 16)	13 (11; 14)	12 (10; 14)	13 (8; 14)	0.248
Active EV, mL/m²								
Women	7 (3; 15)	5 (4; 6)	6 (5; 7)	7 (5; 8)	9 (9; 11)	9 (7; 10)	11 (9; 13)	<0.0001
Men	7 (2; 15)	4 (3; 6)	5 (4; 7)	7 (5; 9)	10 (9; 11)	9 (8; 14)	9 (7; 17)	<0.0001
Overall	5 (4; 7)	5 (4; 6)	6 (4; 7)	9 (7; 10)	9 (6; 10)	9 (8; 12)	10 (8; 14)	<0.0001
Total EF, %								
Women	68 (53; 79)*	68 (63; 71)‡	70 (66; 73)	69 (66; 72)	66 (63; 71)	65 (61; 69)	63 (59; 71)	0.096
Men	66 (51; 80)	64 (61; 67)	66 (64; 69)	66 (60; 71)	65 (62; 70)	62 (58; 66)	61 (59; 74)	0.189
Overall	66 (62; 71)	66 (62; 70)	68 (65; 72)	68 (62; 72)	66 (62; 71)	63 (60; 67)	61 (59; 73)	0.012
Passive EF, %								
Women	45 (22; 60)‡	50 (46; 55)	48 (42; 54)	46 (42; 50)‡	41 (35; 45)	37 (31; 41)	35 (23; 42)	<0.0001
Men	43 (23; 61)	50 (42; 54)	44 (40; 52)	41 (36; 46)	40 (33; 46)	34 (26; 41)	34 (25; 48)	<0.0001
Overall	43 (37; 99)	49 (44; 54)	46 (41; 54)	44 (40; 49)	40 (34; 45)	37 (30; 41)	34 (29; 45)	<0.0001
Active EF, %								
Women	40 (18; 61)	36 (28; 40)	36 (33; 43)	43 (33; 48)	46 (38; 50)	44 (37; 49)	47 (37; 57)	0.001
Men	41 (20; 60)	34 (24; 38)	39 (28; 44)	41 (32; 45)	44 (34; 51)	40 (37; 47)	48 (35; 54)	0.006
Overall	40 (33; 47)	35 (25; 40)	37 (32; 44)	41 (33; 48)	46 (35; 50)	43 (37; 48)	47 (38; 53)	<0.0001
Expansion index, %								
Women	207 (171; 248)*	208 (171; 240)‡	231 (192; 268)	218 (190; 263)	195 (169; 249)	188 (159; 227)	171 (147, 247)	0.096
Men	184 (153; 221)	180 (155; 200)	194 (174; 223)	198 (151; 250)	182 (163; 236)	165 (136; 103)	154 (144, 285)	0.189
Overall	197 (165; 239)	190 (165; 342)	211 (183; 252)	212 (165; 259)	191 (166; 244)	173 (151; 208)	157 (146, 268)	0.012

Data are summarized as median (p25: 25th percentile; p75: 75th percentile). P values refer to age group differences. EF indicates emptying fraction; EV, emptying volume; M, men; Vmax, maximum volume; Vmin, minimum volume; VpreA, pre A volume; and W, women.

*P<0.006 women vs men.

†P<0.0001 women vs men.

‡P<0.05 women vs men.

by an increase in active EF. The LA expansion index also decreased in the elderly subgroups (Table 2).

Comparison With LA Reference Values by 2DE

Upper limits of normality for LA Vmax, VpreA, and Vmin measured by 3DE were 43, 31, and 18 mL/m^2, respectively (Table 3). Lower limit of normality for LA total EF measured by 3DE was 53%. For 2DE, limits of normality for LA Vmax, preA, and Vmin were significantly smaller (35, 25, and 14 mL/m^2, respectively; $P<0.001$) than that for 3DE. Overall, LA volumes measured by 3DE were 22% to 30% larger than the corresponding volumes calculated with 2DE. The difference between 3DE and 2DE LA Vmax was positively correlated with LA size measured by 3DE ($r=0.36$; $P<0.0001$). LA total EF and passive EF measured by 3DE were also larger than those measured by 2DE. Conversely, expansion index was similar when measured by either 3DE or 2DE (Table 3).

Relationships With Body Size, Blood Pressure, and LV Function

On bivariate analysis, all LA volumes showed moderate positive correlations with body size (Vmax: $r=0.41$, 0.48, 0.51, and 0.3 for height, weight, BSA, and body mass index, respectively; $P<0.0001$ for all). Both systolic and diastolic blood pressure showed weak correlations with LA volumes (Vmax $r=0.23$, $P<0.0001$; $r=0.19$, $P=0.002$; Vmin $r=0.22$, $P<0.0001$; $r=0.16$, $P=0.002$). LA passive EF and total and passive EV showed a weak negative correlation with LV GLS ($\rho=-0.15$, -0.21, and -0.12, respectively; $P\leq0.046$). LA active EV correlated with A-wave velocity ($\rho=0.27$; $P<0.001$) and E/e' ($\rho=0.13$; $P=0.046$). LA active EV showed negative correlations with E-wave velocity ($\rho=-0.31$), septal e' ($\rho=-0.35$), lateral e' ($\rho=-0.32$; $P<0.001$ for all). Positive correlations were also found between LA volumes with LV end-diastolic, end-systolic, and stroke volumes (LAV max: $\rho=0.50$, $\rho=0.42$, and $\rho=0.51$, respectively; $P<0.0001$).

On multivariable analysis, age, weight, LV end-systolic volume, GLS, and diastolic function indices (E, A, E/A, e' average, S' average, and E/e') resulted as correlates of LA 3DE volumes and function, accounting for the variance of Vmax=48%, VpreA=50%, Vmin=38%, total EV=34%, passive EV=22%, active EV=35%, total EF=13%, passive EF=29%, active EF=15%, and expansion index=11% (Table 4; see the Data Supplement).

Comparison With CMR (Aim 3)

The validation cohort included 22 consecutive patients (age: 52±19 years; range: 16–79 years; 15 men; BSA: 1.8±0.2 m^2) with acute myocardial infarction (n=8); pericarditis/myocarditis (n=5); cardiomyopathies (n=3), valvular heart disease (n=3), and other (n=3). The temporal resolution of 3DE LA data sets was 54±18 volumes per second. Both 2DE and 3DE LA volumes correlated tightly with CMR measurements (Figures 4 and 5). Bland–Altman analysis showed more underestimation of LA size when measured by 2DE than by 3DE (negative biases of −17 mL and −13 mL for 2DE Vmax and Vmin, and −7 mL and −8 mL for 3DE Vmax and Vmin) and similar limits of agreement (2SD from 30–32 mL). The underestimation by both 3DE and 2DE methods was positively correlated with the LA size at CMR ($r=0.73$ for 3DE Vmax and $r=0.84$ for 2DE Vmax; $P<0.001$ for both).

Reproducibility (Aim 4)

The intra- and interobserver reproducibility of semiautomated LA 3DE quantification was excellent, when analyses were repeated on the same good-quality images by experienced observers (Table 5). At repeated measurements (including test–retest on different image quality data sets and expert versus trainee comparison), LA Vmax and total EF showed the lowest variability among all LA measures. Experience with LA 3DE analysis and image quality impacted mostly on VpreA and Vmin reproducibility ($P<0.01$).

Table 3.   Comparison of Left Atrial Volumes and Function Indices Obtained Using 3DE and 2DE

	3DE		2DE			
	median (p25; p75)	LN	median (p25; p75)	LN	P Value*	Δ% (95% CI)
Vmax, mL/m^2	32 (28; 36)	43	24 (21; 28)	35	<0.001	26.7 (23.6 to 29.8)
VpreA, mL/m^2	18 (14; 21)	31	14 (12; 18)	25	<0.001	21.7 (17.3 to 26.2)
Vmin, mL/m^2	10 (8; 12)	18	8 (6; 10)	14	<0.001	29.9 (24.5 to 35.4)
Total EV, mL/m^2	21 (18; 24)	13	16 (14; 18)	10	<0.001	26.4 (23.3 to 29.6)
Passive EV, mL/m^2	14 (11; 16)	7	10 (7; 12)	4	<0.001	35.9 (31.2 to 40.7)
Active EV, mL/m^2	7 (5; 9)	3	7 (5; 8)	3	0.64	9.4 (3.6 to 15.3)
Total EF, %	67 (63; 71)	53	67 (62; 74)	48	0.03	−0.3 (−2.3 to 1.6)
Passive EF, %	44 (38; 49)	24	41 (32; 48)	19	<0.001	9.9 (5.5 to 14.3)
Active EF, %	41 (35; 48)	21	46 (39; 53)	24	0.09	−12.2 (−16.2 to 8.3)
Expansion index, %	208 (171; 250)	103	204 (165; 289)	110	0.29	−3.8 (−9.5 to 1.8)

Limits of normality (LN) are reported as 2.5 percentile or 97.5 percentile as appropriate. Values are reported as median (p25: 25th percentile; p75: 75th percentile). Δ%, average difference of agreement assessed by Bland–Altman method. CI indicates confidence interval; 2DE, two-dimensional echocardiography; 3DE, 3-dimensional echocardiography; EF, emptying fraction; EV, emptying volume; Vmax, maximum volume; Vmin, minimum volume; and VpreA, pre A volume.

*P values refer to 2DE and 3DE differences by Wilcoxon signed-rank test.

Table 4.   Multivariable Linear Regression Analysis Showing the Correlates of the Left Atrial Phasic and Emptying Volumes

|	LA Vmax, mL		LA VpreA, mL		LA Vmin, mL		LA total EV, mL		LA passive EV, mL		LA active EV, mL	
	r^2	P Value	r^2	P Value	r^2	P Value	r^2	P Value	r^2	P Value	r^2	P Value
Model influence	0.48	<0.0001	0.498	<0.0001	0.384	<0.0001	0.34	<0.0001	0.22	<0.0001	0.352	<0.0001
Variables evaluated	B*	P Value	B*	P Value	B*	P Value	B*	P Value	B*	P Value	B*	P Value
Age, y	0.575	<0.0001	0.564	<0.0001	0.371	<0.0001	0.600	<0.0001	0.257	0.011	0.618	<0.0001
Weight, kg	0.211	0.016	0.158	0.056	…	…	0.194	0.037	…	…	…	…
E, cm/s	…	…	…	…	…	…	0.290	0.002	0.338	0.001	…	…
A, cm/s	0.169	0.062	…	…	…	…	…	…	…	…	…	…
E/A	0.24	0.022	…	…	…	…	…	…	0.240	0.007	…	…
e' average, cm/s	…	…	…	…	…	…	−0.300	0.002	…	…	…	…
E/e' average	−0.18	0.02	…	…	…	…	…	…	−0.966	0.18	−0.14	0.05
LV end-systolic volume, mL	0.296	0.009	0.228	0.035	0.295	0.004	0.378	<0.0001	0.406	<0.0001	…	...
LV stroke volume, mL	0.242	0.015	0.301	…	0.360	<0.0001	…	…	…	…	0.300	<0.0001
LV global longitudinal strain, %	−0.195	0.004	−0.099	0.077	…	…	−0.215	0.004	−0.177	0.028	…	…

A indicates transmitral peak velocity during late diastole; E, transmitral peak velocity during early diastole; e', peak myocardial velocity during early diastole; LA, left atrium; LV, left ventricle; S, peak myocardial velocity during systole; Vmax, maximum volume; Vmin, minimum volume; and VpreA, pre A volume.

*Standardized β value.

There was a larger systematic interobserver bias by 2DE than by 3DE: intraobserver (bias%±SD) 2.6±5% for Vmax, 1±13.6% for Vmin, and 3.1±8.7% for total EF; interobserver 6.1±7.2% for Vmax, 20±12% for Vmin, and 8.1±7.1% for total EF. When compared with multibeat acquisition, LA measurements obtained from single-beat 3DE data sets yielded smaller Vmax (bias −8 mL), larger VpreA (bias 13 mL), similar Vmin and total EF, an overestimation of active EF (by 21%), and underestimation of passive EF (by −23%, Table II in the Data Supplement).
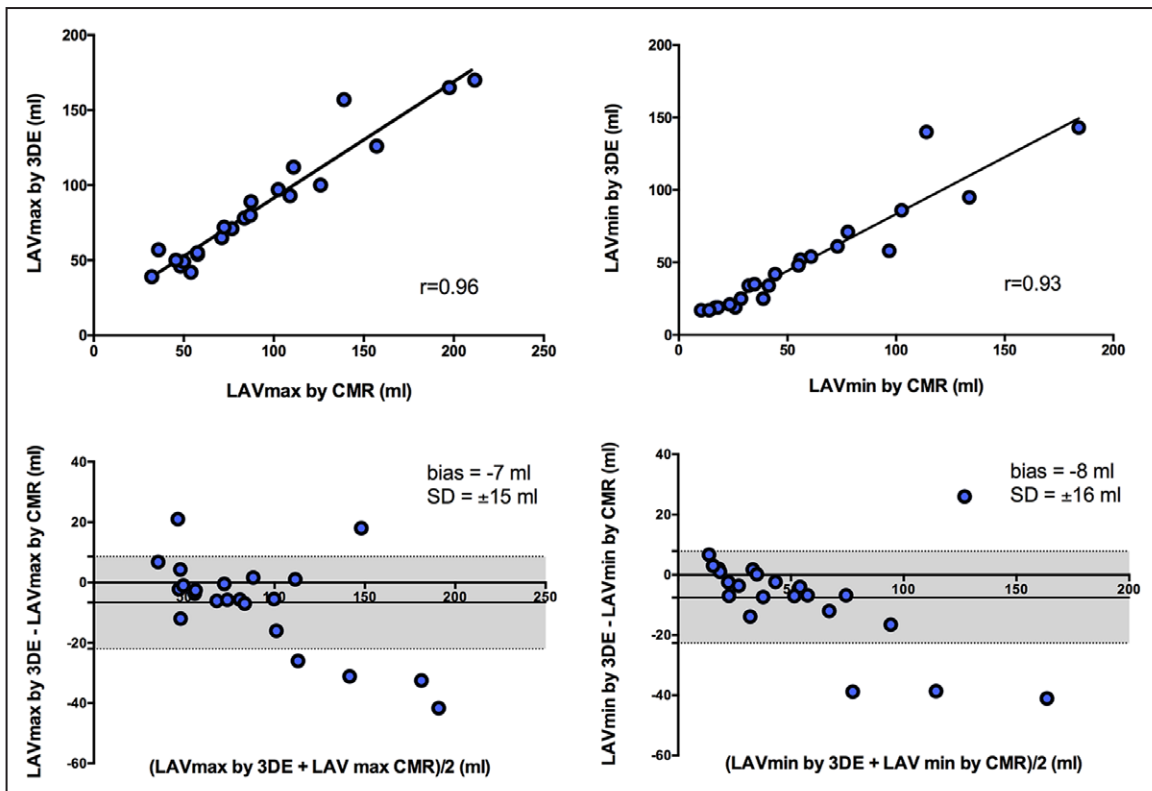
Figure 4. Comparisons of maximum and minimal left atrial (LA) volumes by 3-dimensional echocardiography (3DE) with cardiac magnetic resonance (CMR) reference measurements using Pearson correlation (**top**) and Bland–Altman (**bottom**) analyses.
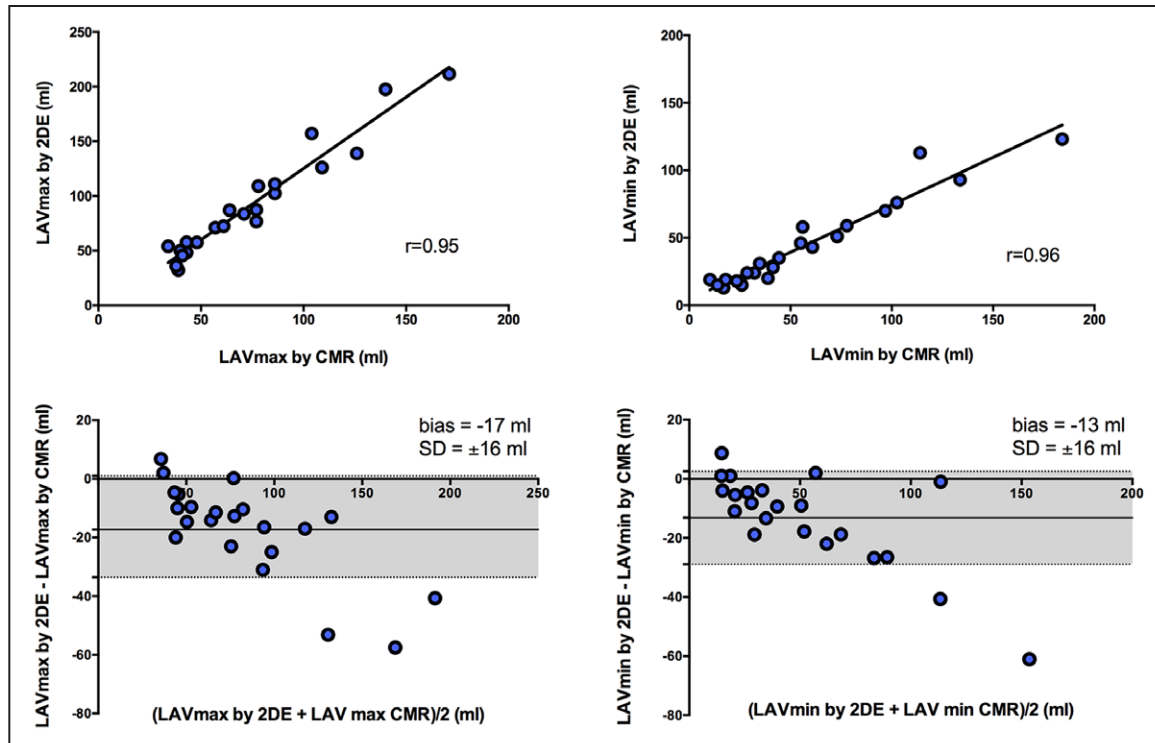
Figure 5. Comparisons of maximum and minimal left atrial (LA) volumes by 2-dimensional echocardiography (2DE) with cardiac magnetic resonance (CMR) reference measurements using Pearson correlation (**top**) and Bland–Altman (**bottom**) analyses.

Discussion

This study provides reference values of LA volumes and phasic function indices measured with 3DE obtained from a relatively large sample of healthy volunteers. Our main results can be summarized as follows: (1) reference values of LA volumes obtained by 3DE were significantly larger than those obtained by 2DE; (2) in healthy subjects, LA volumes indexed by BSA were similar in men and women and increased across age groups; (3) LV end-systolic volume, GLS, diastolic function, and body weight were correlated with LA volumes and phasic function indices; (4) although accuracy of 3DE LA

Table 5. Reproducibility of Left Atrial Volumes by Three-Dimensional Echocardiography

	Intraobserver	Interobserver	Test–Retest	Expert vs Trainee
LA Vmax	1.5±7	1.4±7	0.01±10	1.7±10
LA VpreA	1.1±9	1.7±10	3.6±25	7.9±17
LA Vmin	2±8	2.2±9	8.0±19	4.5±17
LA total EV	2.2±14	3.6±12	4.4±14	0.9±19
LA passive EV	2.4±10	6±10	0.5±38	10.6±31
LA active EV	5±11	8±8	6.1±53	18.6±38
LA total EF	0.6±7	1.1±6	4.4±10	0.8±12
LA passive EF	1.4±14	2.7±10	0.9±39	9.1±25
LA active EF	0.6±11	4.7±6	10±38	10.8±25

Values are reported as bias%±SD. EF indicates emptying fraction; EV, emptying volume; LA, left atrial; Vmax, maximum volume; Vmin, minimum volume; and VpreA, pre A volume.

volume measurement was related to its size and reproducibility varied with image quality, temporal resolution, and operator's experience, overall 3DE LA volume measurements were more accurate and reproducible than 2DE calculations.

It is well known that the LA modulates LV filling through various mechanical functions, acting as (1) a reservoir during LV systole; (2) a conduit for the blood flow from the pulmonary veins to the LV during early diastole; (3) an active contractile chamber that augments LV ventricular filling in late diastole; and (4) a suction source that refills itself in early systole.[1] Yet, the only echocardiographic measure recommended to be routinely quantified is the LA Vmax by 2DE.[2] This stems from to the fact that LA Vmax is an established predictor of adverse outcomes both in the general population and in patients with various heart diseases[15,16] and that tracing the endocardial border of the LA at 3 time points on both the apical 4- and the 2-chamber 2DE views is quite unpractical to perform in routine patients. However, both LA Vmin[8,9,17] and total EF[17,18] showed incremental prognostic power compared with Vmax.

With 3DE, LA volumes at multiple time points during cardiac cycle and several LA function indices are automatically calculated from a single LA data set in just few minutes. In line with previous studies,[5,7,19] our data confirmed that LA volumes measured by 3DE are more accurate and reproducible than those calculated by 2DE.

Reference Values for LA Volumes

Availability of reference values for LA size and function indices is a prerequisite for their routine clinical application. Two studies have previously reported reference values for LA geometry and function in healthy subjects using 3DE. Aune et al[10] studied

166 healthy Norwegian subjects (52% women, 29–80 years) and found very similar lower limits of normality for the LA Vmax (41 versus 43 mL/m², respectively) and Vmin (19 versus 18 mL/m²) but lower normality limit for total EF (45% versus 53%). Wu et al[8] studied 124 healthy Japanese subjects (55% women; 18–85 years) and reported significantly smaller limits of normality for Vmax (33 versus 43 mL/m²) and similar limits of normality for Vmin (18 mL/m²). Interestingly, in their study, Wu et al[8] did not found any difference between 2DE and 3DE LA volumes, and their 3DE normal limit for Vmax is actually smaller than the 2DE one (33 versus 36 mL). This is in contrast with all the other studies that have compared 2DE versus 3DE LA volumes and reported a 20% underestimation of LA volumes by 2DE in comparison to computed tomography and CMR.[3,4]

We think that there may be several reasons for the differences between our study and earlier reports. In both previous studies, a software package designed for LV volume analysis was adapted to measure the LA volume by 3DE, whereas we have used a software specifically designed for LA analysis. The use of semiautomated software packages dedicated for LV, designed to provide an ellipsoidal 3D cast after placing few reference points on the endocardial border, may lead to an underestimation of LA volume, unless the semiautomated 3D LA model is heavily modified by the operator.[20] Cardiac computed tomography and anatomic studies have shown that the LA has a complex shape, with an irregular barrel-like free-wall and an oblique, flat interatrial septal wall,[21,22] which hardly resembles the ellipsoidal LV shape. The LA-specific software used in our study requires a substantial manual input for initialization of endocardial contours, in order to capture more closely the complex LA shape. Moreover, both previous studies used a different ultrasound system and a wide-angle 3D acquisition for the LA, which resulted in a significantly lower temporal resolution (18 versus 42 volumes per second in our study). We have found that lower temporal resolution of 3DE data sets may lead to significantly smaller LA Vmax and a trend toward lower total EF in comparison with measurements from high volume rate data sets (Table II in the Data Supplement). Therefore, a high temporal resolution from dedicated 3DE LA acquisitions seems important for an accurate evaluation of LA maximal size and phasic function.

The accuracy of the LA 3DE software used in this study has been demonstrated by comparison with CMR in a multicenter setting.[5] In our single-center validation cohort, LA 3DE volume measurements were tightly correlated and slightly underestimated in comparison with CMR, in line with other validation studies comparing 3DE volumes of LV and RV with CMR.[23,24] The bias was significantly larger in patients with enlarged atria (Figure 4). Our limits of agreement with CMR are similar those reported by Mor-Avi et al,[5] whereas the systematic bias is larger (−8 versus −1 mL). The latter could be related to a slightly different way of endocardial border initialization (more outward, as depicted in the paper by Mor-Avi et al,[5] with respect to our way of border initialization, which was performed on the black-white interface, Figure 2). Notably, in our study, the upper normal limit for 2DE LA Vmax was 35 mL/m², which is practically similar with the 34 mL/m² cutoff value indicated by current guidelines.[2]

Comparison Between 3DE and 2DE Reference Values

Our reference values for 3DE volumes were significantly larger than those for 2DE. The average bias for LA Vmax measured
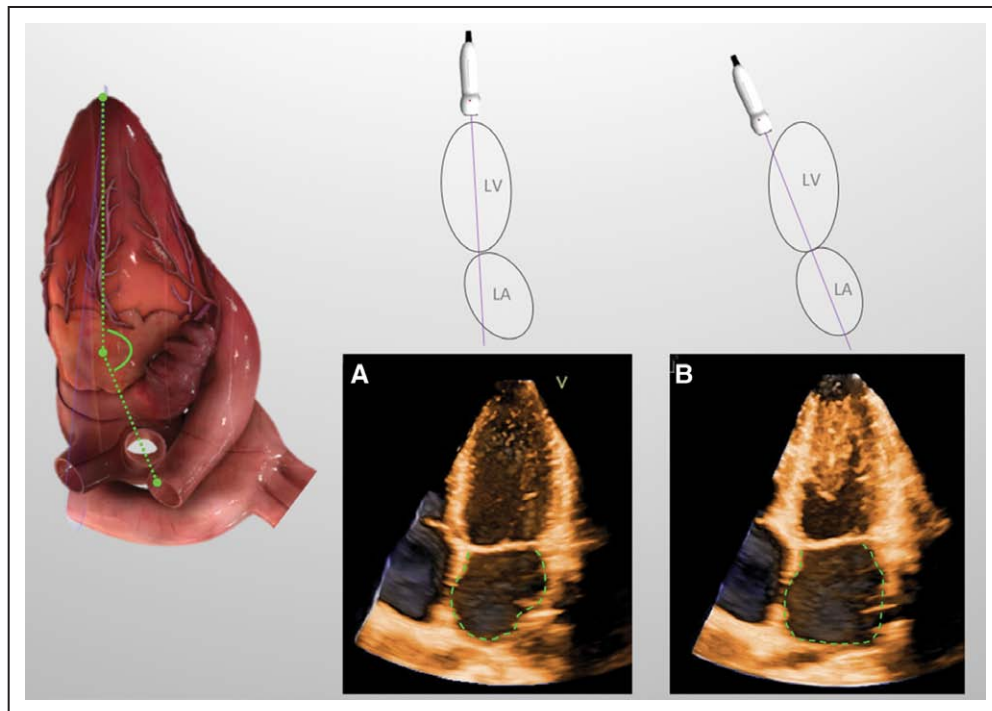
Figure 6. Three-dimensional anatomic heart model and 3-dimensional echocardiography (3DE) rendering, illustrating the fact that the long axis of the left ventricle (LV) and left atrium (LA) do not lie in the same plane. LA size displayed in the 4-chamber view optimized for the LV (**A**) is smaller than the LA size displayed in the 4-chamber view in which LA has been maximized (**B**). Notice that the LV cavity is almost closed in (**B**). HeartWorks anatomy image reprinted with permission of the publisher. Copyright ©2016, Inventive Medical, Ltd.

by 3DE versus 2DE was 10 mL, and it was similar in healthy subjects and in CMR patients. Larger differences between 3DE and 2DE were seen in larger atria, suggesting that 2DE underestimates more the LA volume when the LA is larger.

LA foreshortening, geometric assumptions, and manual tracing errors may contribute to LA volume underestimation by 2DE versus 3DE. Measuring the LA in the conventional 4- and 2-chamber views (commonly optimized to display the maximal LV length) is a common source of LA size underestimation, because LV and LA axes do not lie in the same plane[2] (Figure 6). Our 2DE imaging protocol included dedicated apical views for LA, in which efforts were made to maximize the LA size (Figure 7).[2] However, as it is the case for LV volumes,[25] dedicated views may still underestimate the true LA size because of constrains related to acoustic access and also because there is no reliable way to verify and exclude the LA foreshortening by 2DE only (unless by comparison with 3DE nonforeshortened views, Figure 8). In addition, 2DE calculations are based on the geometric assumptions that the 4-chamber view and the 2-chamber view are orthogonal and that they are crossing the LA cavity through its center, assumptions which do not hold true (Figure 8). Finally, the manual tracing of the endocardium is operator dependent, and 2DE showed higher systematic interobserver bias than 3DE, suggesting that border tracing manner may also play a role in 2DE volume measurements.

Demographic and Anthropometric Correlates of LA Volumes

In agreement with previous studies, we found that body size was a major determinant of LA volumes.[26,27] To adjust for this effect and allow meaningful comparisons, LA size should be indexed by BSA.[2] In our healthy study sample, from which obese subjects have been excluded, sex differences were almost completely accounted for after correcting LA volumes for BSA.

The effect of age on LA size and function remains controversial.[28] It has been reported that the normal ageing process is associated with an increase in LA size.[29] However, other studies of normal individuals over a wide range of age did not find any relationship between LA volumes and age.[30] In our study sample, all LA volumes increased significantly with age, also LA passive EF decreased, whereas LA active EF increased across age groups. Whether these changes reflect the age-related decrease in LV relaxation or are consequences of the chronologic ageing per se remains to be clarified.

Limitations

This is a single-center study on a cohort of white healthy subjects performed with a LA-specific software applied on dedicated transthoracic 3DE data sets. Our reference values might not apply for measurements obtained by adapting vendor-specific LV software packages for LA analysis or from single-beat or wide-angle 3DE data sets with lower temporal resolution. Despite this is the largest study to date to report reference values of LA 3DE volumes, the study sample could have been too small to support a precise estimation of normal limits or to detect sex differences. Because our enrollment criteria excluded the obese, the effect of indexation of LA volumes to BSA in this setting remains to be clarified. Larger multicenter and multiethnic studies will hopefully clarify the effects of sex, race, and body size on LA reference values.
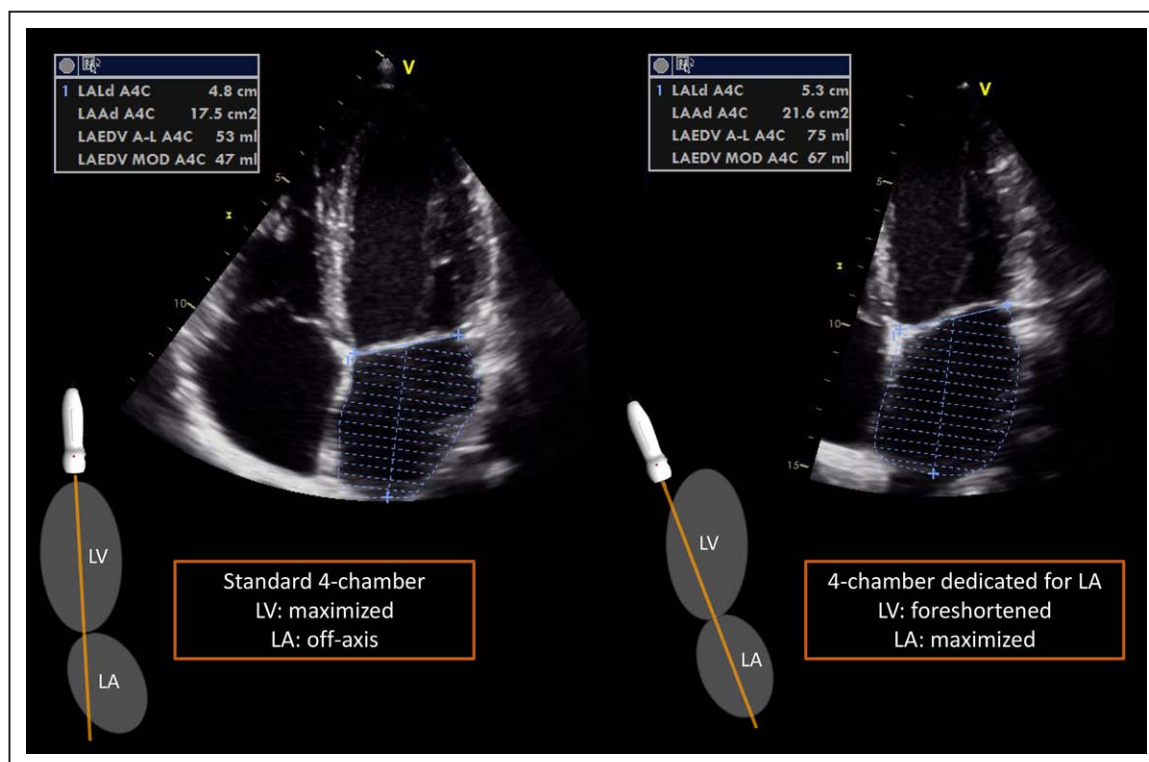
Figure 7. Example of difference in left atrial (LA) measurements by two-dimensional echocardiography (2DE) between the standard 4-chamber view and the 4-chamber view dedicated for LA. A-L indicates area-length method; LAAd, left atrial maximal area; LAEDV, left atrial maximal volume; LALd, left atrial maximal length; LV, left ventricle; and MOD, method of discs.
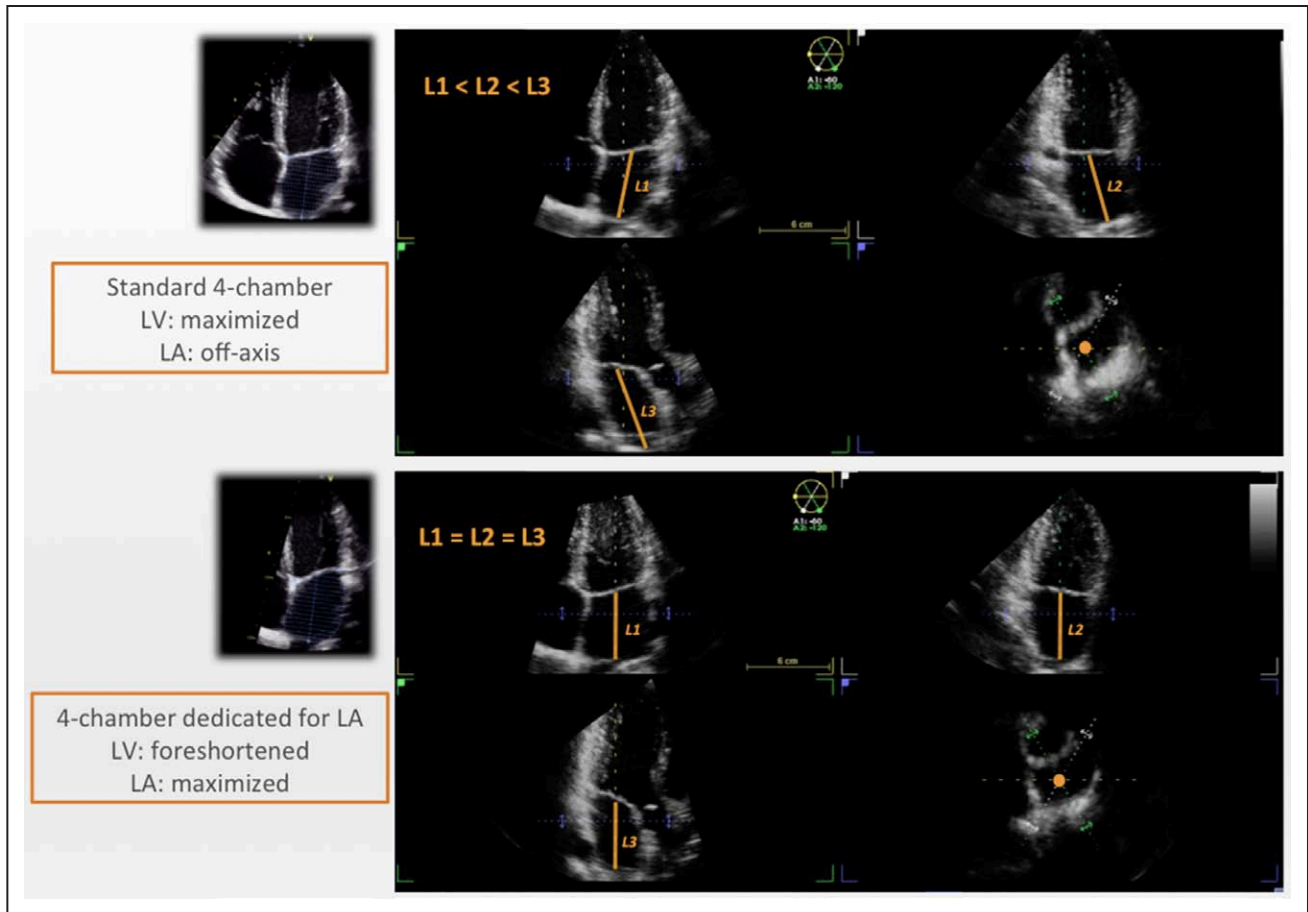
Figure 8. Comparison between the 2-dimensional echocardiography (2DE) views from Figure 7 vs 3-dimensional echocardiography (3DE) data set of the same subject before (**top right**) and after alignment (**bottom right**). In the standard 4-chamber view, there is a significant foreshortening of the left atrium (LA), which is almost resolved, albeit not completely, in the dedicated LA 4-chamber view. L1, L2, and L3 represent the LA lengths in the 3 apical views.

Given the limited number of subjects ≥70 years, reference values in this age group may be less robust. However, the enrollment of truly healthy volunteers in the elderly subgroup is particularly challenging. Despite our strict enrollment criteria, we cannot exclude the possibility of subclinical coronary artery disease, especially in older participants.

Conclusions

This study provides reference values for LA 3DE volumes and function from a relatively large cohort of healthy subjects with a wide age range. Because LA volumes are significantly larger when measured by 3DE than by 2DE, the limits of normality for detecting LA remodeling and dysfunction by 2DE cannot be used interchangeably with those by 3DE. Availability of specific reference values for 3DE may help clinicians to identify LA remodeling and dysfunction and should facilitate the implementation of 3DE for LA assessment in clinical and research practice.

Sources of Funding

Disclosures

References

1. Leung DY, Boyd A, Ng AA, Chi C, Thomas L. Echocardiographic evaluation of left atrial size and function: current understanding, pathophysiologic correlates, and prognostic implications. *Am Heart J.* 2008;156:1056–1064. doi: 10.1016/j.ahj.2008.07.021.

2. Lang RM, Badano LP, Mor-Avi V, Afilalo J, Armstrong A, Ernande L, Flachskampf FA, Foster E, Goldstein SA, Kuznetsova T, Lancellotti P, Muraru D, Picard MH, Rietzschel ER, Rudski L, Spencer KT, Tsang W, Voigt JU. Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *Eur Heart J Cardiovasc Imaging.* 2015;16:233–270. doi: 10.1093/ehjci/jev014.

3. Avelar E, Durst R, Rosito GA, Thangaroopan M, Kumar S, Tournoux F, Chan RC, Hung J, Hoffmann U, Abbara S, Brady T, Cury RC. Comparison of the accuracy of multidetector computed tomography versus two-dimensional echocardiography to measure left atrial volume. *Am J Cardiol.* 2010;106:104–109. doi: 10.1016/j.amjcard.2010.02.021.

4. Kühl JT, Lønborg J, Fuchs A, Andersen MJ, Vejlstrup N, Kelbæk H, Engstrøm T, Møller JE, Kofoed KF. Assessment of left atrial volume and function: a comparative study between echocardiography, magnetic resonance imaging and multi slice computed tomography. *Int J Cardiovasc Imaging.* 2012;28:1061–1071. doi: 10.1007/s10554-011-9930-2.

5. Mor-Avi V, Yodwut C, Jenkins C, Kühl H, Nesser HJ, Marwick TH, Franke A, Weinert L, Niel J, Steringer-Mascherbauer R, Freed BH, Sugeng L, Lang RM. Real-time 3D echocardiographic quantification of left atrial volume: multicenter study for validation with CMR. *JACC Cardiovasc Imaging*. 2012;5:769–777. doi: 10.1016/j.jcmg.2012.05.011.

6. Keller AM, Gopal AS, King DL. Left and right atrial volume by freehand three-dimensional echocardiography: *in vivo* validation using magnetic resonance imaging. *Eur J Echocardiogr*. 2000;1:55–65. doi: 10.1053/euje.2000.0010.

7. Jenkins C, Bricknell K, Marwick TH. Use of real-time three-dimensional echocardiography to measure left atrial volume: comparison with other echocardiographic techniques. *J Am Soc Echocardiogr*. 2005;18:991–997. doi: 10.1016/j.echo.2005.03.027.

8. Wu VC, Takeuchi M, Kuwaki H, Iwataki M, Nagata Y, Otani K, Haruki N, Yoshitani H, Tamura M, Abe H, Negishi K, Lin FC, Otsuji Y. Prognostic value of LA volumes assessed by transthoracic 3D echocardiography: comparison with 2D echocardiography. *JACC Cardiovasc Imaging*. 2013;6:1025–1035. doi: 10.1016/j.jcmg.2013.08.002.

9. Caselli S, Canali E, Foschi ML, Santini D, Di Angelantonio E, Pandian NG, De Castro S. Long-term prognostic significance of three-dimensional echocardiographic parameters of the left ventricle and left atrium. *Eur J Echocardiogr*. 2010;11:250–256. doi: 10.1093/ejechocard/jep198.

10. Aune E, Baekkevar M, Roislien J, Rodevand O, Otterstad JE. Normal reference ranges for left and right atrial volume indexes and ejection fractions obtained with real-time three-dimensional echocardiography. *Eur J Echocardiogr*. 2009;10:738–744. doi: 10.1093/ejechocard/jep054.

11. Dubois D, Dubois EF. Clinical calorimetry. A formula to extimate the approximate surface area if height and weight are known. *Arch Int Med* 1916;17:863–71.

12. Muraru D, Cucchini U, Mihăilă S, Miglioranza MH, Aruta P, Cavalli G, Cecchetto A, Padayattil-Josè S, Peluso D, Iliceto S, Badano LP. Left ventricular myocardial strain by three-dimensional speckle-tracking echocardiography in healthy subjects: reference values and analysis of their physiologic and technical determinants. *J Am Soc Echocardiogr*. 2014;27:858–871.e1. doi: 10.1016/j.echo.2014.05.010.

13. To AC, Flamm SD, Marwick TH, Klein AL. Clinical utility of multimodality LA imaging: assessment of size, function, and structure. *JACC Cardiovasc Imaging*. 2011;4:788–798. doi: 10.1016/j.jcmg.2011.02.018.

14. Perez de Isla L, Feltes G, Moreno J, Martinez W, Saltijeral A, de Agustin JA, Gomez de Diego JJ, Marcos-Alberca P, Luaces M, Ferreiros J, Garcia Fernandez MA, Macaya C. Quantification of left atrial volumes using three-dimensional wall motion tracking echocardiographic technology: comparison with cardiac magnetic resonance. *Eur Heart J Cardiovasc Imaging* 2014;15:793–799.

15. Beinart R, Boyko V, Schwammenthal E, Kuperstein R, Sagie A, Hod H, Matetzky S, Behar S, Eldar M, Feinberg MS. Long-term prognostic significance of left atrial volume in acute myocardial infarction. *J Am Coll Cardiol*. 2004;44:327–334. doi: 10.1016/j.jacc.2004.03.062.

16. Pritchett AM, Mahoney DW, Jacobsen SJ, Rodeheffer RJ, Karon BL, Redfield MM. Diastolic dysfunction and left atrial volume: a population-based study. *J Am Coll Cardiol*. 2005;45:87–92. doi: 10.1016/j.jacc.2004.09.054.

17. Russo C, Jin Z, Homma S, Rundek T, Elkind MS, Sacco RL, Di Tullio MR. Left atrial minimum volume and reservoir function as correlates of left ventricular diastolic function: impact of left ventricular systolic function. *Heart*. 2012;98:813–820. doi: 10.1136/heartjnl-2011-301388.

18. Gupta S, Matulevicius SA, Ayers CR, Berry JD, Patel PC, Markham DW, Levine BD, Chin KM, de Lemos JA, Peshock RM, Drazner MH. Left atrial structure and function and clinical outcomes in the general population. *Eur Heart J*. 2013;34:278–285. doi: 10.1093/eurheartj/ehs188.

19. Artang R, Migrino RQ, Harmann L, Bowers M, Woods TD. Left atrial volume measurement with automated border detection by 3-dimensional echocardiography: comparison with magnetic resonance imaging. *Cardiovasc Ultrasound*. 2009;7:16. doi: 10.1186/1476-7120-7-16.

20. Muraru D, Peluso M, Bianco LD, Beraldo M, Solda' E, Tuveri M, Cucchini U, Mamary AA, Badano L, Iliceto S. Single-plane and biplane 2D algorithms and non-atrial specific 3D echo softwares underestimate left atrial volumes in comparison with specific 3D echo software tailored for left atrium. *Eur J Echocardiogr* 2011;12:1. Abstract.

21. O'Brien JP, Srichai MB, Hecht EM, Kim DC, Jacobs JE. Anatomy of the heart at multidetector CT: what the radiologist needs to know. *Radiographics*. 2007;27:1569–1582. doi: 10.1148/rg.276065747.

22. Ho SY, McCarthy KP, Faletra FF. Anatomy of the left atrium for interventional echocardiography. *Eur J Echocardiogr*. 2011;12:i11–i15. doi: 10.1093/ejechocard/jer093.

23. Muraru D, Onciul S, Peluso D, Soriani N, Cucchini U, Aruta P, Romeo G, Cavalli G, Iliceto S, Badano LP. Sex- and Method-specific reference values for right ventricular strain by 2-dimensional speckle-tracking echocardiography. *Circ Cardiovasc Imaging*. 2016;9:e003866. doi: 10.1161/CIRCIMAGING.115.003866.

24. Shimada YJ, Shiota T. A meta-analysis and investigation for the source of bias of left ventricular volumes and function by three-dimensional echocardiography in comparison with magnetic resonance imaging. *Am J Cardiol*. 2011;107:126–138. doi: 10.1016/j.amjcard.2010.08.058.

25. Muraru D, Badano LP, Peluso D, Dal Bianco L, Casablanca S, Kocabay G, Zoppellaro G, Iliceto S. Comprehensive analysis of left ventricular geometry and function by three-dimensional echocardiography in healthy adults. *J Am Soc Echocardiogr*. 2013;26:618–628. doi: 10.1016/j.echo.2013.03.014.

26. Poutanen T, Jokinen E, Sairanen H, Tikanoja T. Left atrial and left ventricular function in healthy children and young adults assessed by three dimensional echocardiography. *Heart*. 2003;89:544–549.

27. Abhayaratna WP, Seward JB, Appleton CP, Douglas PS, Oh JK, Tajik AJ, Tsang TS. Left atrial size: physiologic determinants and clinical applications. *J Am Coll Cardiol*. 2006;47:2357–2363. doi: 10.1016/j.jacc.2006.02.048.

28. Casaclang-Verzosa G, Gersh BJ, Tsang TS. Structural and functional remodeling of the left atrium: clinical and therapeutic implications for atrial fibrillation. *J Am Coll Cardiol*. 2008;51:1–11. doi: 10.1016/j.jacc.2007.09.026.

29. Nikitin NP, Witte KK, Thackray SD, Goodge LJ, Clark AL, Cleland JG. Effect of age and sex on left atrial morphology and function. *Eur J Echocardiogr*. 2003;4:36–42.

30. Thomas L, Levett K, Boyd A, Leung DY, Schiller NB, Ross DL. Compensatory changes in atrial volumes with normal aging: is atrial enlargement inevitable? *J Am Coll Cardiol*. 2002;40:1630–1635.

CLINICAL PERSPECTIVE

Left atrial (LA) size is an important predictor of outcome in various clinical conditions. LA analysis by 3-dimensional echocardiography (3DE) is more accurate and reproducible and offers incremental prognostic information in comparison with calculations by 2-dimensional echocardiography. However, the limited normative data available for 3DE has significantly limited its clinical use. This prospective study was designed to assess the reference values of LA volumes and phasic function indices by 3DE and the influence of age, sex, body size, and left ventricular function in 276 healthy adult volunteers. We found that LA volumes indexed by body surface area were similar in men and women and increased across age groups. Because LA volumes are significantly larger when measured by 3DE than by 2-dimensional echocardiography, the limits of normality for detecting LA remodeling and dysfunction by 2-dimensional echocardiography cannot be used interchangeably with those by 3DE. Body weight and LV function indices were also correlated with LA 3DE parameters. Thus, LA size and function measured by 3DE should be interpreted taking into account patient age, body surface area, and left ventricular systolic and diastolic function. Overall, 3DE LA volume measurements were more accurate and reproducible than 2-dimensional echocardiography calculations. Our reference values may help clinicians to identify LA remodeling and dysfunction and should facilitate the implementation of 3DE for LA assessment in clinical and research practice. Given the prognostic implication of LA minimum volume and emptying fraction measured by 3DE, the availability of specific reference values will allow more routine evaluation of these parameters.