Analysis of Left Ventricular Volumes and Function: A Multicenter Comparison of Cardiac Magnetic Resonance Imaging, Cine Ventriculography, and Unenhanced and Contrast-Enhanced Two-Dimensional and Three-Dimensional Echocardiography

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Background: Contrast echocardiography improves accuracy and reduces interreader variability on left ventricular (LV) functional analyses in the setting of two-dimensional (2D) echocardiography. The need for contrast imaging using three-dimensional (3D) echocardiography is less defined. The aim of this multicenter study was to define the accuracy and interreader agreement of unenhanced and contrast-enhanced 2D and 3D echocardiography for the assessment of LV volumes and ejection fraction (EF).

Methods: A multicenter, open-label study was conducted including 63 patients, using intrasubject comparisons to assess the agreement of unenhanced and contrast-enhanced 2D and 3D echocardiography as well as calibrated biplane cine ventriculography with cardiac magnetic resonance for the determination of LV volumes and EF. Each of the imaging techniques used to define LV function was assessed by two independent, off-site readers unaware of the results of the other imaging techniques.

Results: LV end-systolic and end-diastolic volumes were underestimated by 2D and 3D unenhanced echocardiography compared with cardiac magnetic resonance. Contrast enhancement resulted in similar significant increases in LV volumes on 2D and 3D echocardiography. The mean percentage of interreader variability for LV EF was reduced from 14.3% (95% confidence interval [CI], 11.7%–16.8%) for unenhanced 2D echocardiography and 14.3% (95% CI, 9.7%–18.9%) for unenhanced 3D echocardiography to 8.0% (95% CI, 6.3%–9.7%; $P < .001$) for contrast-enhanced 2D echocardiography and 7.4% (95% CI, 5.7%–9.1%; $P < .01$) for contrast-enhanced 3D echocardiography and thus to a similar level as for cardiac magnetic resonance (7.9%; 95% CI, 5.4%–10.5%). A similar effect was observed for interreader variability for LV volumes.

Conclusions: Contrast administration on 3D echocardiography results in improved determination of LV volumes and reduced interreader variability. The use of 3D echocardiography requires contrast application as much as 2D echocardiography to reduce interreader variability for volumes and EF. (J Am Soc Echocardiogr 2014;27:292-301.)

Keywords: Cardiac magnetic resonance, Cine ventriculography, Contrast echocardiography, 3D echocardiography, Left ventricular function

Left ventricular (LV) volumes and ejection fraction (EF) are major clinical parameters with respect to diagnosis and prognosis in patients with cardiac diseases. Important treatment decisions and the evaluation of therapeutic effects are based on these parameters.[1-3] Several techniques have been used for the analysis of LV volumes and EF, among them cine ventriculography, echocardiography, cardiac

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Abbreviations
CI = Confidence interval
CMR = Cardiac magnetic resonance
EF = Ejection fraction
LV = Left ventricular
3D = Three-dimensional
2D = Two-dimensional

magnetic resonance (CMR) and computed tomography.[4-7] CMR has evolved into the preferred reference technique because of its high spatial resolution and ability to obtain complete volumetric data sets, allowing very accurate determinations of regional and global LV function.[7,8] Echocardiography has been limited by moderate reproducibility and accuracy due to poor acoustic windows as well as inadequate discrimination of the endocardial border. In addition, limited accuracy has been related to geometric assumptions resulting from the two-dimensional (2D) approach. Recent innovations in contrast and three-dimensional (3D) echocardiography have enabled significant improvements in endocardial border definition and have made it possible to overcome the geometric assumptions of native 2D echocardiography.[9-12] There is only limited knowledge of the impact of contrast administration in 3D echocardiography.[13]

The objective of this multicenter study was to determine the accuracy and interreader agreement of unenhanced and contrast-enhanced 2D and 3D echocardiography for the assessment of LV volumes and EF in comparison with CMR. In addition, cine ventriculography was performed in all patients. All echocardiographic techniques as well as cine ventriculography and CMR were performed in all patients to allow intraindividual comparative effectiveness assessment. Acquisition of cardiac images was performed at four sites. Blinded off-site reads using independent core laboratories were performed for each imaging technique according to well-defined standards.

METHODS

This was a multicenter, open label study using intraindividual comparisons to assess the agreement of unenhanced and contrast-enhanced 2D and 3D echocardiography with CMR for the determination of LV volumes and EF performed between January and October 2009. In addition, calibrated biplane cine ventriculography was performed in all patients. To undergo cine ventriculography, patients had to have indications for coronary angiography due to stable chest pain but no acute myocardial infarction and no coronary intervention during the procedure. All imaging studies were performed within 48 hours.

To provide uniform and interpretable image data sets, recommendations on the performance of image acquisition were predefined for all imaging modalities and provided to all participating institutions. Adherence to the predefined imaging protocols was monitored during the enrollment period of this multicenter trial.

Each of the imaging techniques used to define LV function was assessed by two independent experienced (at least 5 years of experience in the evaluated imaging modality) off-site readers (reader 1 and reader 2) at independent core laboratories unaware of the results of the other imaging techniques. For a uniform evaluation, the evaluation procedures were predefined and provided as guidelines.

The primary objective of this study was to determine interreader variability in the assessment of LV volumes and EF using unenhanced and contrast-enhanced echocardiography, CMR, and cine ventriculography. For the primary objective, the analysis was prospectively planned considering the results of readers 1 and 2 in each modality. Correlation coefficients were compared using single-sample tests of correlation coefficients.[14] The research protocol was approved by the local institutional ethics committees. All patients gave written informed consent to participate in the study.

Patients

Sixty-three patients were enrolled at four European centers, with balanced contributions. Patient enrollment was stratified at each center on the basis of results from angiographic ventriculography to achieve a balanced distribution within three predefined EF groups (>55%, 35%–55%, and <35%). An interpretable cine ventriculogram with the availability of at least two consecutive nonextrasystolic cardiac cycles during ventriculographic contrast administration was a prerequisite for inclusion in the study.

Echocardiography

At all sites, 2D echocardiography was performed using a commercially available ultrasound scanner (iE33; Philips Medical Systems, Andover, MA) using tissue harmonic imaging for unenhanced and contrast-specific imaging for contrast-enhanced echocardiography. Two-dimensional apical four-chamber, two-chamber, and three-chamber views as well as 3D full-volume data sets from the apical position were acquired without and with contrast enhancement. Five consecutive cardiac cycles of each view were acquired during breath hold and digitally stored. Great care was taken to avoid apical foreshortening and to maximize the length from base to apex. A 3D full-volume data set of the ventricle was obtained with gated (five beats) acquisition. Sector size and depth were optimized to obtain the highest possible volume rates, reaching 17 to 20 frames/sec in the contrast 3D full-volume mode.

For contrast-enhanced assessment of LV function, a 20-gauge intravenous catheter was introduced into the right antecubital vein. SonoVue (Bracco Imaging, Milan, Italy) was administered using a dedicated infusion pump (VueJect; Bracco Imaging) with continuous mixing of the contrast agent suspension at a starting infusion rate of 1 mL/min and subsequent adjustment to reach homogenous LV cavity opacification without attenuation. SonoVue is a commercially available ultrasound contrast agent consisting of sulfur hexafluoride microbubbles stabilized by a highly flexible phospholipid monolayer shell.

Ultrasound machine settings were optimized for contrast specific imaging. Transmit power was set to be low (mechanical index < 0.4), and dynamic range was adjusted to achieve optimal contrast between cardiac walls and the LV cavity.

Analysis of unenhanced and enhanced echocardiograms as well as 2D and 3D echocardiography was performed in random order. All acquired 2D and 3D data sets were transferred to a dedicated workstation (TomTec 4D; TomTec Imaging Systems, Munich, Germany). Considering 2D data sets, end-diastolic and end-systolic LV volumes and EF were determined by semiautomatic tracing of end-systolic and end-diastolic endocardial borders using apical four-chamber and two-chamber views, using Simpson's method. According to the recommendations of the American Society of Echocardiography,[15] the tracings were performed with the papillary muscles and trabeculations allocated to the LV cavity. The mitral annulus was to be traced as deeply as possible. Considering the 3D data sets, reconstructed 2D views of the four-chamber, two-chamber, and long-axis views were obtained using the TomTec system. Within the reconstructed views, endocardial border tracing of the end-diastolic and end-systolic images was performed to obtain the corresponding LV volumes.

TomTec 4D LV-Analysis software was also used for semiautomatic tracing of the endocardial borders in the full-volume data sets. For this purpose, endocardial border tracings were semiautomatically performed in three long-axis images at end-systole and end-diastole. The contouring was verified on long-axis and short-axis cine images and modified as necessary to ensure optimal endocardial tracking including analysis of the valve plane.

Cine Ventriculography

Standard biplane cine ventriculography was performed using a 30° right anterior oblique projection and a 60° left anterior oblique projection with injection of ≥30 cm^3 contrast medium at a flow rate of 12 to 14 mL/sec using 5-Fr to 6-Fr pigtail catheters. The frame rate was set at 30 Hz. Semiautomatic border tracking was used to define the end-diastolic image on the basis of the R wave on electrocardiography and the end-systolic image on the basis of the frame with the smallest ventricular silhouette. Image calibration was performed with the use of a metal ball with a diameter of 5.0 cm with identical positions of the x-ray tubes. LV end-diastolic and end-systolic volumes were determined using Simpson's method, according to well-defined standards and after formal training for biplane analyses, using CAAS II software with the LV biplane analysis module (Pie Medical Imaging, Maastricht, The Netherlands).

CMR

Electrocardiographically triggered CMR investigations at a field strength of 1.5 T during breath hold were performed using a special volume-adapted surface coil. To assess LV function, standard steady-state free precession cine imaging (spatial resolution, 1.4 × 1.4 × 8 mm; 35 phases per cardiac cycle; repetition time, 3.1 msec; echo time, 1.6 msec; flip angle, 55°) was performed during short repetitive end-expiratory breath holding. Four-chamber, two-chamber, three-chamber, and short-axis views with a slice thickness of 10 mm were acquired in the basal-apical direction.

Evaluations were performed using Siemens Argus software (syngoMMWP VE27A and syngo VE31H; Siemens Healthcare, Erlangen, Germany). Endocardial border tracings were performed automatically by the system, with manual correction if needed for each short-axis slice separately at end-diastole and end-systole to derive LV volumes and EF. The definition of the most basal slice required continuously visible myocardium, including its transition into the LV outflow tract. Tracings were performed with the papillary muscles and trabeculations allocated to the LV cavity as performed on echocardiographic images.

Statistical Analysis

Statistical analysis was performed using Medidata (Medidata Solutions, Konstanz, Germany). Continuous variables are presented as mean ± SD and were compared using Student's *t* tests. The limits of agreement (defined as ±2 SDs from the mean difference) between readers 1 and 2 on echocardiographic analysis of global LV function without and with contrast administration as well as between echocardiographic and CMR measurements of global LV function were determined using Bland-Altman analysis.[16] Linear regression analysis was performed to determine the correlations between readers 1 and 2 and between echocardiography and CMR in the assessment of volumes and EF. In addition, the interreader variability in the assessment of LV volumes and EF between the two readers was determined as a percentage of variability. The percentage of variability was calculated as the standard deviation between two measurements divided by their mean multiplied by 100. *P* values ≤ .05 were considered to indicate statistical significance.

RESULTS

Baseline Characteristics

Forty-nine male and 14 female patients (mean age, 63.8 ± 10.4 years) were included in this study. Twenty patients (31%) had histories of myocardial infarction. Prior coronary revascularization procedures included percutaneous coronary intervention in 33 patients (52%) and coronary bypass surgery in seven patients (11%). The patients' mean height was 171 ± 8 cm (range, 150–186 cm), and their mean weight was 76 ± 13 kg (range, 46–115 kg). The SonoVue infusion rate to achieve optimal image quality (Figure 1) was 1.13 ± 0.19 mL/min. In two patients, CMR was not performed. These patients were excluded from the analysis. All other patients were included irrespective of image quality. Table 1 displays the patient characteristics of all 63 patients.

LV Volumes and EF

Table 2 displays end-diastolic and end-systolic volumes as well as EF determined using the three echocardiographic techniques with and without contrast enhancement, by CMR, and by cine ventriculography as measured by reader 1 for each imaging modality. End-diastolic and end-systolic volumes by CMR were significantly larger than those obtained by any of the echocardiographic modalities. There were no significant differences in EF between any of the applied imaging modalities. Thus, neither 3D echocardiography compared with 2D echocardiography nor contrast-enhanced echocardiography compared with unenhanced echocardiography resulted in significantly different measurements of LV EF.

LV end-diastolic and end-systolic volumes defined by reconstructed 2D planes from 3D echocardiographic data sets as well as 3D full-volume data sets were not significantly different from LV volumes determined by 2D echocardiography (Table 2). There were also no differences in EF between 2D and 3D echocardiography.

End-diastolic volumes determined by contrast-enhanced echocardiography were significantly larger than those defined by unenhanced echocardiography, irrespective of the use of 2D or 3D echocardiography (Table 2). The differences between end-diastolic volumes determined by contrast echocardiography and those determined by CMR were significantly smaller than those determined by unenhanced echocardiography.

Interreader Variability in the Determination of LV Volumes and EF

The correlation between readers 1 and 2 for EF was improved for 2D contrast-enhanced echocardiography ($r = 0.88$; 95% confidence interval [CI], 0.82–0.94) compared with 2D unenhanced echocardiography ($r = 0.79$; 95% CI, 0.68–0.86; $P = .045$). The correlation between readers 1 and 2 for EF with 3D contrast-enhanced echocardiography ($r = 0.90$; 95% CI, 0.84–0.94) was improved compared with 3D unenhanced echocardiography ($r = 0.74$; 95% CI, 0.61–0.84; $P = .001$).

The 95% limits of agreement between both readers for EF with 2D echocardiography were reduced from −12.6% to 26.8% to −10.1% to 17.8% with contrast administration. For 3D full-volume data sets, the limits of agreement for EF between both readers were reduced

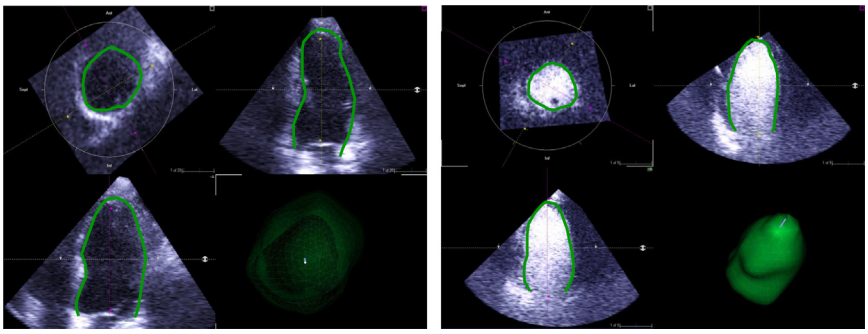

Figure 1 Apical transthoracic 3D echocardiography display of the left ventricle obtained without (*left*) and with (*right*) administration of contrast agent using contrast-specific low–mechanical index imaging techniques. The *green line* indicates the endocardial tracking.

Table 1 Patients' baseline characteristics (n = 63)

Variable	Value
Age (y)	63.8 ± 10.4
History of myocardial infarction	20 (32%)
Prior coronary angioplasty	33 (52%)
Prior coronary bypass surgery	7 (11%)
Significant coronary artery disease	48 (76%)
Coronary stenosis in LAD	35 (56%)
Coronary stenosis in LCX/RCA	40 (63%)
Diabetes mellitus	6 (10%)
Hypertension	44 (70%)
Hypercholesterolemia	21 (33%)
EF by cine ventriculography (%)	
<35	10 (16%)
35–55	16 (25%)
>55	37 (59%)

LAD, Left anterior descending coronary artery; *LCX*, left circumflex coronary artery; *RCA*, right coronary artery.
Data are expressed as mean ± SD or number (percentage).

Table 2 LV volumes and EF determined by the different imaging techniques in 63 patients

Technique	ESV (mL)	EDV (mL)	EF (%)
2D echocardiography	48 ± 41	107 ± 51	56 ± 15
2D contrast echocardiography	59 ± 45*	129 ± 53*	54 ± 15
3D reconstructed views	45 ± 38	99 ± 48	55 ± 14
3D reconstructed views with contrast	59 ± 46*	127 ± 52*	54 ± 15
3D full-volume echocardiography	53 ± 43	107 ± 48	50 ± 15
3D full-volume contrast echocardiography	59 ± 44	123 ± 48*	52 ± 14
CMR	97 ± 68	175 ± 73	46 ± 14
Cine ventriculography	59 ± 48	167 ± 82	64 ± 17

EDV, End-diastolic volume; *ESV*, end-systolic volume.
Data were determined by reader 1 for each method.
*P < .05 for comparison of LV volumes determined by contrast-enhanced versus unenhanced echocardiography.

from −25.2% to 15.5% to −11.2% to 16.2% with contrast administration (Figure 2).

Interreader variability between readers 1 and 2, expressed as the mean percentage of variability, ranged from 4.5% to 28% for end-diastolic volume for the different imaging modalities (Table 3). It was only 4.5% for CMR, whereas it reached 28% for unenhanced 3D full-volume echocardiography. The mean percentage of variability for end-diastolic volume was reduced using contrast enhancement compared with unenhanced imaging for 2D ($P < .001$) as well as both 3D echocardiographic modalities ($P < .001$ for both) (Figure 3). The mean percentage of variability for cine ventriculography was significantly higher compared with contrast echocardiography. The findings for the mean percentage of variability for end-systolic volume were similar to those for end-diastolic volume, with overall higher variability (Table 3, Figure 3).

The mean percentage of variability for EF between readers 1 and 2 for the different imaging modalities was in the range of 7.4% to 14.3% (Figure 3). It was low for CMR, at 7.9% (95% CI, 5.4%–10.5%). It was also low for the 2D and 3D contrast-enhanced echocardiographic modalities (Table 3). It was higher for cine ventriculography, at 13.8% (95% CI, 10.9%–16.7%) as well as for unenhanced 2D and 3D echocardiography. Using contrast-enhanced echocardiography, the mean percentage of variability between readers 1 and 2 for EF was significantly ($P < .001$) reduced, with much smaller CIs compared with unenhanced echocardiography considering 2D as well as 3D echocardiography. The mean percentage of variability on contrast-enhanced echocardiography was comparable with that obtained for CMR. Furthermore, the mean percentage of variability for EF was significantly ($P < .001$) lower between the two readers using contrast-enhanced echocardiography compared with cine ventriculography.

Agreement between the Echocardiographic Imaging Modalities and CMR in the Determination of EF

The mean difference between EF defined by unenhanced 2D echocardiography (reader 1) and CMR (reader 1) was changed from 11.0% (95% limits of agreement, −9.6% to 31.7%) to 9.2% (95% limits of agreement, −5.3% to 23.8%) using contrast enhancement (Table 4, Figure 4). Considering full-volume 3D echocardiography, the mean difference between EFs by unenhanced and contrast-enhanced echocardiography (reader 1) and CMR (reader 1) was slightly lower compared with 2D echocardiography. The mean difference between EFs defined by unenhanced 3D full-volume echocardiography (reader 1) and CMR (reader 1) was not significantly different from that of 3D full-volume echocardiography with contrast enhancement (4.8% vs 6.3%). The 95% limits of agreement with CMR were reduced from −19.1% to 28.6% for unenhanced 3D echocardiography to

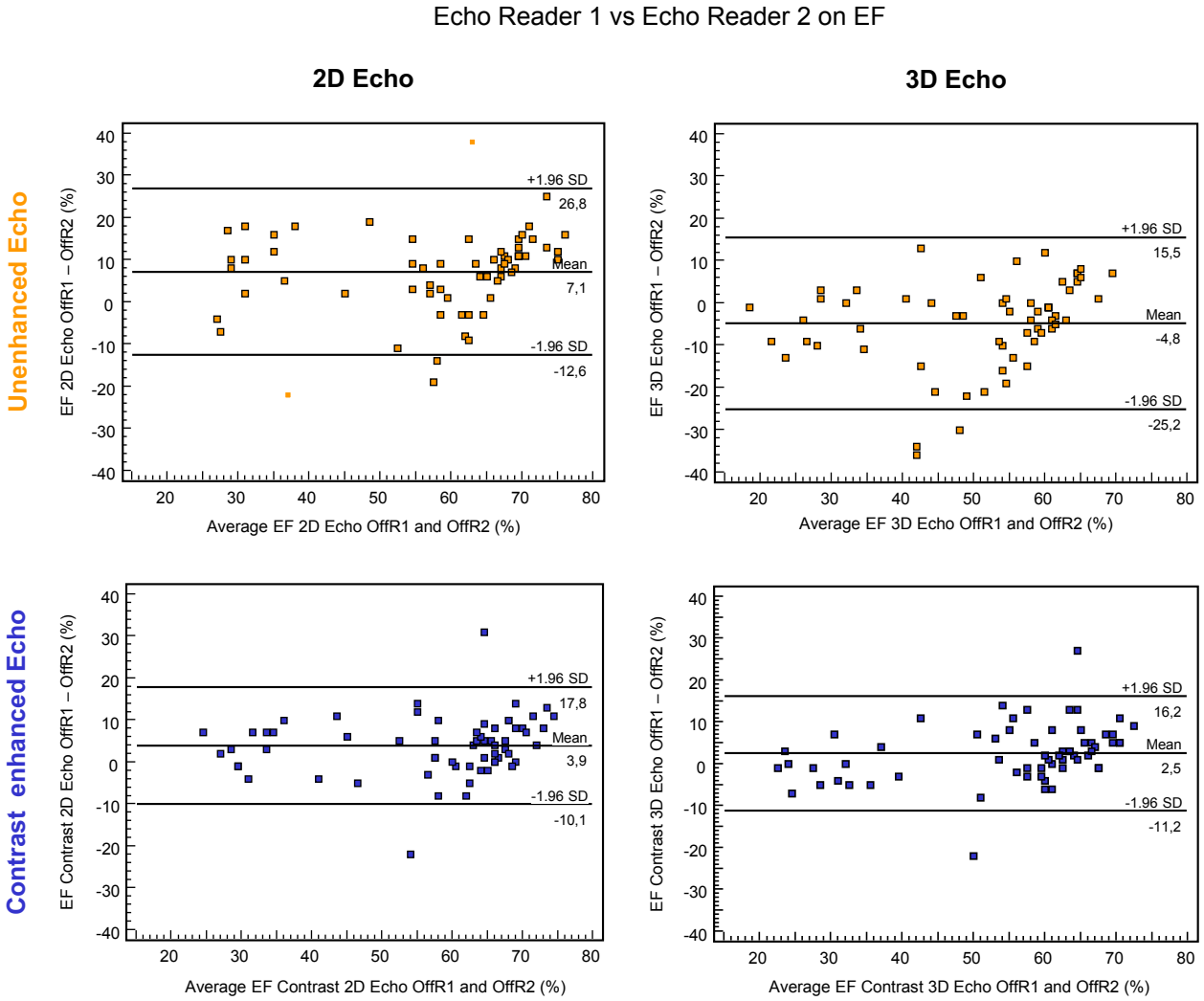

Figure 2 Bland-Altman plots for intramethod agreement. The plots show the mean difference (*solid lines*) and the limits of agreement (*dashed lines*) between measurements by off-site reader 1 and off-site reader 2 on unenhanced (*top row*) and contrast-enhanced (*bottom row*) echocardiographic measurements of EF using 2D echocardiography (*left*) and 3D full-volume echocardiography (*right*).

−8.1% to 20.7% for contrast-enhanced 3D full-volume echocardiography (Table 4).

The correlation between echocardiography and CMR for measurements of EF increased significantly compared with unenhanced echocardiography if contrast was administered for both 2D ($r = 0.76$ vs 0.87, respectively, $P = .032$) and full-volume 3D ($r = 0.75$ vs 0.89, respectively, $P = .007$) echocardiography (Figure 5, Table 4).

DISCUSSION

The present study demonstrates that (1) unenhanced 2D and 3D echocardiography significantly underestimates LV volumes compared with CMR, (2) contrast enhancement improves accuracy in the determination of LV volumes irrespective of the use of 2D or 3D echocardiography, (3) contrast enhancement reduces interreader variability in the determination of LV volumes and EF with both 2D and 3D echocardiographic techniques, (4) the interreader variability of contrast-enhanced 2D and 3D echocardiography for EF is on a level observed with CMR, and (5) intermethod agreement of 2D and 3D echocardiography with CMR for EF is increased by contrast enhancement.

Multiple studies have compared cine ventriculography, echocardiography, CMR, and cardiac computed tomography for the definition of LV volumes and EF.[4-7,17-19] Most studies have been performed in single-center settings with single readers. The majority of previous studies have used 2D echocardiography for the analysis of LV volumes and EF. Recently, a meta-analysis was performed to obtain a better understanding of the diagnostic value and accuracy of the different imaging techniques.[17] Three-dimensional echocardiography was found to result in slightly larger LV volumes than 2D echocardiography, whereas EF was similar between both methods.

Microbubble administration in combination with recent advances in contrast-specific LV imaging has been shown to result in significant improvement in endocardial border definition and reader confidence in regional and global LV function assessment.[20,21] Improved accuracy and reliability of LV volume and functional measurements were demonstrated in single-center as well as multicenter 2D

Table 3 Interreader variability in 63 patients on assessments of EDV, ESV, and EF for the different imaging techniques

Variable	Mean percentage of variability	95% CI
EDV		
Cine ventriculography	17.8	12.9–22.8
CMR	4.5	3.2–5.9
Unenhanced echocardiography		
2D	19.7	16.3–23.1
3D reconstructed views	26.6	22.6–30.6
3D full volume	28.0	23.6–32.5
Contrast-enhanced echocardiography		
2D	9.7*,‡	8.0–11.5
3D reconstructed views	10.9*	8.5–13.2
3D full volume	9.6*,‡	7.4–11.8
ESV		
Cine ventriculography	35.3	27.6–43.0
CMR	8.5	6.5–10.4
Unenhanced echocardiography		
2D	34.5	28.8–40.3
3D reconstructed views	39.5	33.7–45.3
3D full volume	24.6	20.3–28.9
Contrast-enhanced echocardiography		
2D	15.6*,‡	12.1–19.1
3D reconstructed views	18.3*,‡	14.3–22.4
3D full volume	15.0*,‡	11.3–18.6
EF		
Cine ventriculography	13.8	10.9–16.7
CMR	7.9	5.4–10.5
Unenhanced echocardiography		
2D	14.3	11.7–16.8
3D reconstructed views	13.6	10.8–16.4
3D full volume	14.3	9.7–18.9
Contrast-enhanced echocardiography		
2D	8.0*,†,‡	6.3–9.7
3D reconstructed views	8.5*,†,‡	6.9–10.1
3D full volume	7.4*,†,‡	5.7–9.1

EDV, End-diastolic volume; *ESV*, end-systolic volume.
*$P < .01$, contrast-enhanced versus unenhanced echocardiography.
†P = NS versus CMR.
‡$P < .05$ versus cine ventriculography.

echocardiography–based studies.[10,21] Although LV EF defined by echocardiography was found to be very similar compared with CMR for unenhanced as well as contrast-enhanced imaging, LV volumes were found to be too small compared with CMR and cine ventriculography, even with contrast administration.[22] Geometric assumptions and foreshortening of the left ventricle have been considered to be the reasons for such an underestimation by 2D techniques. Three-dimensional echocardiography has been reported to improve accuracy in the assessment of LV volumes.[8,11,23] Recent echocardiographic developments have allowed the full-volume capture of 3D data sets within one cardiac cycle.[24] However, 3D echocardiography is frequently affected by a low definition of the endocardial border. There is only limited knowledge on contrast administration during 3D echocardiography for LV volume analysis.[13] A major advantage of the present study in comparison with previous single-center studies is its multicenter design, with the acquisition of imaging data at different sites and subsequent off-site reading by independent blinded core centers.

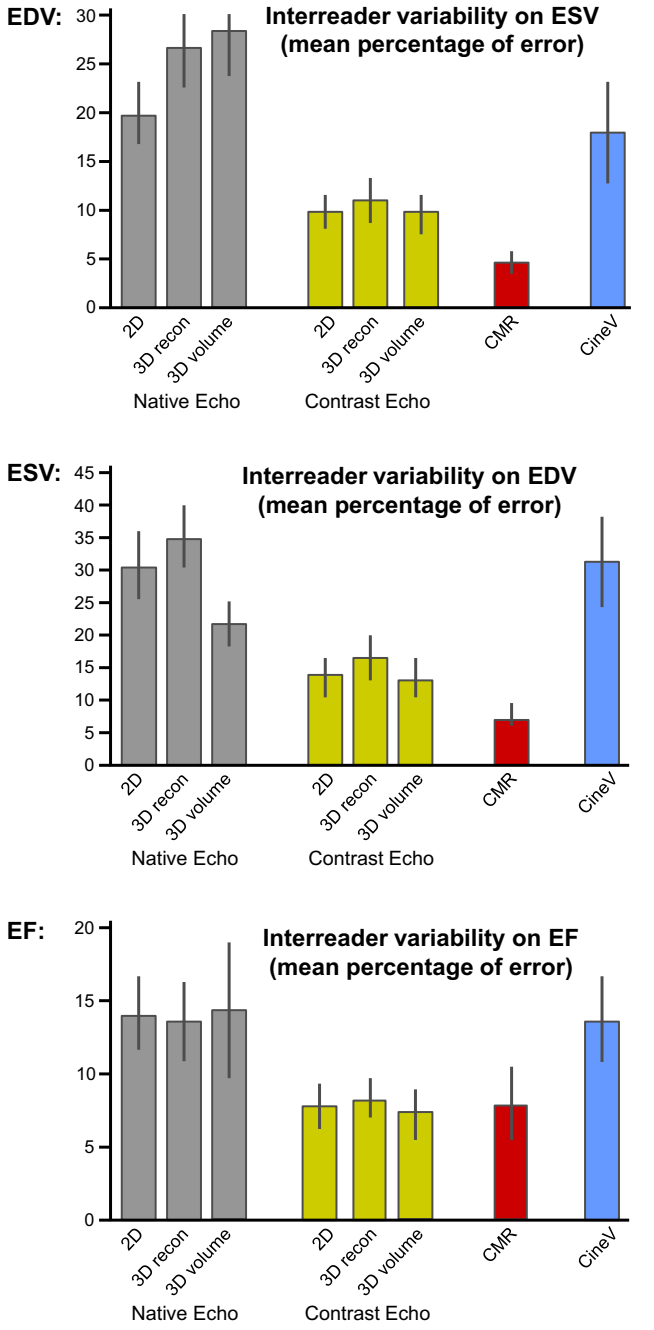

Figure 3 Interreader variability in the assessment of LV end-diastolic volume (EDV), LV end-systolic volume (ESV), and EF between the two readers for unenhanced echocardiography, contrast-enhanced echocardiography, CMR, and cine ventriculography expressed as mean percentage of error. The interreader variability is given for all three evaluated echocardiographic modalities: 2D echocardiography, reconstructed views from the 3D data set (3D recon), and 3D full-volume (3D volume).

LV Volumes

LV volumes by 2D as well as 3D echocardiography were significantly underestimated using unenhanced echocardiography with state-of-the-art harmonic imaging compared with CMR. Underestimation of LV volumes by up to 50% using echocardiography in comparison

Table 4 Intermethod agreement in 63 patients for EF, described as the mean difference between methods and correlation between methods

Comparison	Unenhanced echocardiography			Contrast enhanced echocardiography			*P* value for correlation coefficient
	Mean difference	Limits of agreement	*r*	Mean difference	Limits of agreement	*r*	
2D echocardiography vs CMR	11.0	−9.6 to 31.7	0.76	9.2	−5.3 to 23.8	0.87	.032
3D reconstructed vs CMR	9.3	−9.2 to 27.4	0.79	6.2	−8.4 to 20.4	0.89	.027
3D full volume vs CMR	4.8	−19.1 to 28.6	0.75	6.3	−8.1 to 20.7	0.89	.007

Results are given for unenhanced echocardiography and contrast-enhanced echocardiography (related to reader 1 echocardiography vs reader 1 CMR).

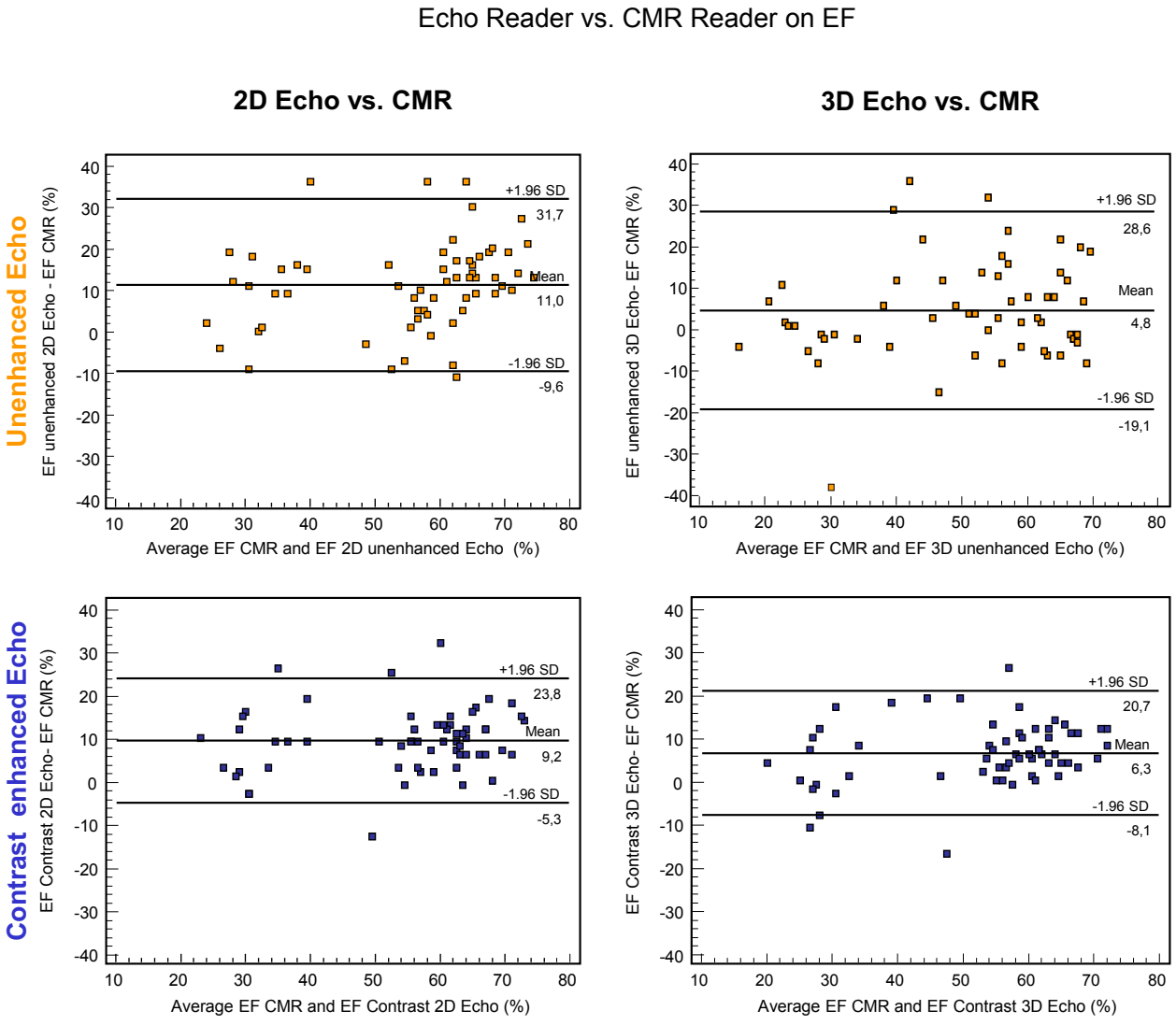

Figure 4 Bland-Altman plots for intermethod agreement. The plots show the mean difference (*solid lines*) and the limits of agreement (*dashed lines*) between measurements of EF by unenhanced echocardiography and CMR (*top row*) and between measurements of EF by contrast-enhanced echocardiography and CMR (*bottom row*) using 2D echocardiography (*left*) and 3D full-volume echocardiography (*right*).

with CMR and cine ventriculography has been reported for 2D echocardiography.[4,18-20,25] It reflects the inability to visualize the endocardial border contours, to define the real LV apex by 2D echocardiography, and the need for geometric assumptions.[26] Contrast enhancement resulted in significantly higher volumes and better correlation and agreement with the reference methods, as has been demonstrated in single-center studies as well as in a previous multicenter study.[9,10,22] In this study, LV volumes defined by 3D

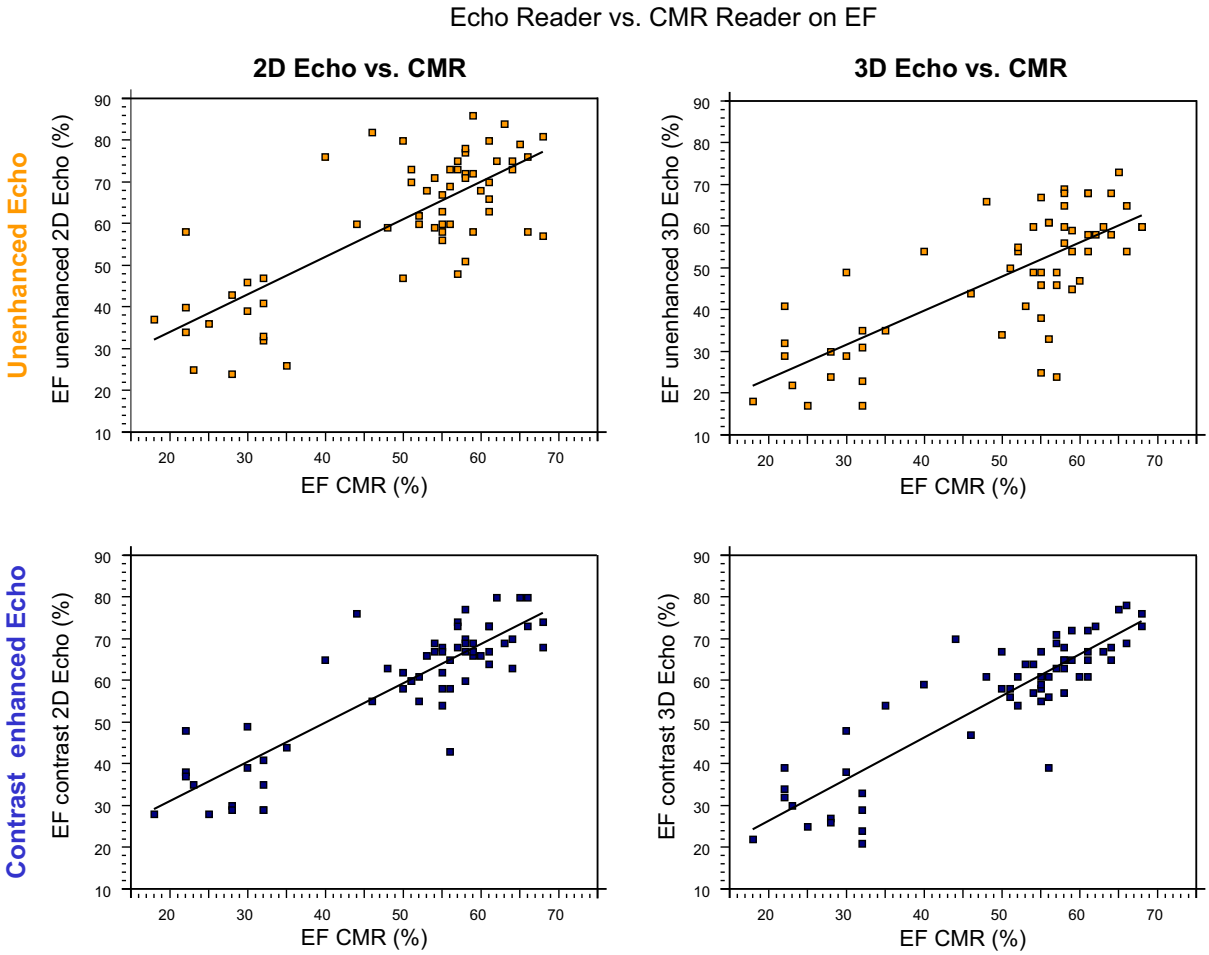

Figure 5 Linear regression plots for intermethod agreement. The plots show measurements of EF by unenhanced echocardiography and CMR (*top row*) and by contrast-enhanced echocardiography and CMR (*bottom row*) using 2D echocardiography (*left*) and 3D full-volume echocardiography (*right*).

echocardiography were not greater compared with those by 2D echocardiography. This is in contrast to most previous single-center studies. However, Although initial studies of 3D echocardiography demonstrated almost equivalence to CMR in the assessment of LV volumes, subsequent studies demonstrated significant underestimation compared with CMR even with 3D echocardiography.[17] Furthermore, a recent study of CMR, cardiac computed tomography, cine ventriculography, and 2D and 3D echocardiography also did not show greater volumes with 3D echocardiography compared with 2D echocardiography.[18] Thus, the findings of this multicenter study on LV volumes using 2D or 3D echocardiography do not stand alone. In this study, contrast administration was found to result in larger LV volumes compared with unenhanced echocardiography and less underestimation compared with CMR also using 3D echocardiography. To minimize the difference in volume measurements compared with CMR, contrast administration should therefore be considered also when 3D echocardiography is used. This finding is in agreement with a single-center study of 20 patients that demonstrated improved accuracy on volume measurements compared with CMR with contrast 3D echocardiography.[13]

EF

Unenhanced echocardiography resulted in only moderate agreement with CMR for EF, whereas contrast application increased the correlation and reduced the limits of agreement with CMR. Three-dimensional echocardiography compared with 2D echocardiography did not change the average assessment of EF. This is in agreement with a recent meta-analysis.[17] Three-dimensional compared with 2D echocardiography did not result in a reduction of the average bias for EF considering CMR as the standard. Contrast administration resulted in improved correlation as well as reduced bias of end-diastolic and end-systolic volumes compared with CMR when applied with 2D as well as 3D echocardiography.

Interobserver Variability in the Determination of LV Volumes and EF

For situations in which serial follow-up of LV function is clinically relevant, the reliability of volume and EF determination is crucial to clinical decision making. CMR has been commended for its high accuracy and reproducibility, making it possible to reduce sample sizes compared with 2D echocardiography.[27,28] In a recent study comparing 2D and 3D unenhanced and contrast-enhanced echocardiography for the sequential assessment of LV EF and volumes, noncontrast 3D echocardiography was the most reproducible technique for LV volume and LV EF measurements.[29] Although contrast enhancement resulted in a reduction of minimal detectable volume change for 2D echocardiography as well as triplane

echocardiography, this reduction was not seen for 3D echocardiography. However, patients selected for that study needed to have normal strain measurements, which suggests good image quality, a prerequisite to obtain normal strain measurements. In our study, a considerable proportion of patients had suboptimal image quality, thus enhancing the effect of contrast administration. Furthermore, semiautomatic contour detection was applied in the study by Thavendiranathan *et al.*[29] only with 3D unenhanced echocardiography. In contrast, manual contour detection was applied with 3D contrast-enhanced images because of limitations of the applied software for semiautomatic analysis of contrast-enhanced images. This difference in analytic technique is likely to have resulted in the inferiority of contrast 3D echocardiography compared with unenhanced 3D echocardiography. In our study, the same analytic modalities were applied with contrast-enhanced and unenhanced 3D echocardiography. Thus, differences observed in our study between the applied imaging modalities could be attributed to the imaging itself, not to the analytic technique. There was a remarkable reduction in interreader variability on the determination of LV volumes and EF when contrast-enhanced echocardiograms were compared with unenhanced echocardiograms irrespective of the use of 2D or 3D echocardiographic techniques. For LV EF, the interreader agreement of contrast echocardiography reached the same level as the interreader agreement of CMR and was better than that of cine ventriculography.

Data on interreader variability have been reported for echocardiography, CMR, and cine ventriculography.[20,22,28-30] In most studies, only readers from the same centers participated in the analysis, and only data on one or two imaging modalities were reported. In a previous multicenter study, contrast administration on 2D echocardiography was shown to result in a significant reduction in interreader variability for EF, to a level similar to that with CMR.[22] This study confirms the results of that previous study with similar findings. Interreader variability for 2D unenhanced echocardiography and cine ventriculography was high, while interreader variability for 2D contrast-enhanced echocardiography and CMR was significantly lower. However, this study extends previous findings to 3D echocardiography. Although interreader variability for LV volumes and EF tended to be greater with 3D compared with 2D echocardiography, contrast enhancement with 3D echocardiography also resulted in a significant reduction of interreader variability.

The low interreader variability of 2D and 3D contrast echocardiography indicates that it may be a very valid method for studies requiring serial assessments of LV systolic function.

Study Limitations

It is impossible to blind readers to the presence of contrast agents on echocardiographic images or the use of 2D vs 3D imaging techniques, and this may potentially induce bias. However, readers were totally blinded to the patients' identities and to the other imaging results of each patient. Training of off-site readers was similar for all imaging techniques.

Current 3D contrast echocardiographic imaging allows the acquisition of full-volume data sets with a frame rate of only up to 20 frames/sec if optimal sector size and depth selection are used. Thus, in case of high heart rates, the real end-diastolic and end-systolic volumes may be missed.

Although native 2D and 3D echocardiography and contrast 2D echocardiography have been in clinical practice for many years, there is much less experience in performing 3D contrast echocardiography and analyzing 3D contrast echocardiographic data sets. The applied 3D software was the only tool that allowed semiautomatic analysis of 3D contrast echocardiographic data sets at the time of study initiation, while other software tools applied in preparation of the study failed to allow a reproducible analysis.

CONCLUSIONS

Contrast administration on 3D echocardiography results in improved determination of LV volumes. The application of contrast on 3D echocardiography reduces interreader variability in LV volumes and function. For LV functional analysis, interreader variability can be achieved with 3D echocardiography using contrast administration that is similar to that of CMR. The use of 3D echocardiography requires contrast application as much as 2D echocardiography to reduce interreader variability in volumes and EF. Contrast administration should be fostered to improve the accuracy and reproducibility of LV volume and function measurements.

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APPENDIX

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