

Validation of 3-Dimensional Speckle Tracking Imaging to Quantify Regional Myocardial Deformation

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Background-Three-dimensional speckle tracking imaging (3D-STI) has been introduced to assess regional left ventricular (LV) myocardial function. This study was designed to validate LV strain measurements by 3D-STI against data obtained by sonomicrometry.

Methods and Results-In each of 10 anesthetized sheep, sonomicrometry crystals were implanted on the endo- and epicardium at the LV basal, mid, and apical anterior and lateral walls. LV 3D-STI data sets were obtained from the apical approach at a frame rate of approximately 30 frames/s. Segmental longitudinal (LS), radial (RS), and circumferential strain (CS) measurements by 3D-STI were compared with those by sonomicrometry at baseline and during pharmacological stress tests (dobutamine and propranolol infusion) and acute myocardial ischemia induced by coronary artery occlusion. Data were available from 136 LS, 108 CS, and 175 RS measurements. Good correlations were observed between strain measurements by 3D-STI and those by sonomicrometry (LS: $r=0.89$, $p<0.001$; RS: $r=0.84$, $p<0.001$; CS: $r=0.90$, $p<0.001$). In each segmental study, significant correlations of the three strain components were observed (LS: $r=0.65$ to 0.68 , $p<0.001$; RS: $r=0.59$ to 0.70 , $p<0.001$; CS: $r=0.71$ to 0.78 , $p<0.001$).

Conclusions- The newly developed 3D-STI technique can estimate LV regional circumferential, longitudinal, and radial strain components with reasonable correlation to sonomicrometry data. This methodology could be applied clinically to assess alteration of myocardial function by accurately measuring strain in basal, mid, and apical LV segments, even during pharmacological and ischemic interventions. Therefore, 3D-STI appears to be a reliable tool to assess LV regional wall function.

Key Words: 3-dimensional imaging, echocardiography, speckle tracking, myocardial contraction, strain

Left ventricular (LV) regional wall motion analysis is one focus of cardiac ultrasound imaging technology. Technology to measure regional myocardial deformation has been applied as an objective methodology for the assessment of regional myocardial function. Regional strain and strain-rate estimation with tissue Doppler imaging based on tissue velocity was first applied to assess regional wall deformation.¹⁻³ However, strain and strain-rate can be accurately estimated only at regions parallel to the ultrasound beam because one limitation of tissue Doppler imaging is angle dependence owing to the use of the Doppler effect.⁴ Recently, speckle tracking imaging (STI), which is based on tracking of speckle patterns created by interference of ultrasound beams in the myocardium, has been available to assess regional myocardial deformation. This methodology is based on a B-mode image, so it is conceptually angle independent of the ultrasound beam, and it provides for measurement of three deformation components, radial, circumferential, and longitudinal strain, on a commercially available echocardiographic system. Reliability of STI in estimation of these three strain components has been confirmed in previous experimental and clinical studies.⁵⁻⁸ In the clinical setting, STI has been applied to assess LV function and regional wall motion abnormalities, i.e., reduced function of ischemic myocardium,⁹ diastolic function in patients with heart failure,¹⁰ and cardiac dyssynchrony to predict response for cardiac resynchronization therapy.^{11,12} However, currently available STI methodology has problems because of its two-dimensional (2D) imaging-based method. The out-of-plane problem inherent in short-axis imaging is caused by longitudinal heart motion during the cardiac cycle. In addition, circumferential rotation also contributes to three-dimensional (3D) wall deformations and affects tracking accuracy. Therefore, accurate calculation of Lagrangian strain in LV regional walls requires an advanced

technology that uses a 3D tracking system. 3D echocardiography has been applied to assess LV volume and mass¹³⁻¹⁷ and LV global and regional wall motion, which is based on time-volume analysis of global and segmental LV volumes.¹⁸ For 3D-STI, although spatial temporal resolution has been a technical challenge, a robust 3D-STI system has been developed and introduced on a commercially available ultrasound system (Artida™, Toshiba Medical Systems Co., Tochigi, Japan).¹⁹ The aim of this experimental study was to validate LV strain measurements by the 3D-STI system against data obtained by sonomicrometry.

Methods

Animal Preparation

Ten male hybrid Suffolk sheep (Japan Lamb Co., Ltd., Hiroshima, Japan) were used for this study. After receiving approval from the Institutional Animal Experiment Committee of the University of Tsukuba, we carried out all experiments in a humane manner and in accordance with the “Regulation for Animal Experiments” of our university and the “Fundamental Guideline for Proper Conduct of Animal Experiment and Related Activities in Academic Research Institutions” under the jurisdiction of the Ministry of Education, Culture, Sports, Science and Technology. Anesthesia was induced with thiopental sodium (10-15 mg/kg intravenously), and the animals were intubated. Anesthesia was maintained with isoflurane (1.5%-2%) and oxygen. All animals underwent left thoracotomy under aseptic conditions. Polypropylene snares were loosely placed around the appropriate coronary arteries. A fluid-filled catheter was inserted via a femoral artery for continuous monitoring of systemic arterial pressure and heart rate.

Sonomicrometry

In all sheep, 6 sonomicrometry crystals (2 mm in diameter, Sonometrics Corp., London, Ontario, Canada) were implanted in LV basal, mid, and apical anterior and lateral subendocardium. Before implantation of the crystals in the subendocardium, we carefully observed and determined the 6 placement positions with 2-D echocardiographic imaging. Basal crystals were implanted at the inferior border between the basal and mid segments, mid crystals were implanted at the inferior border between the mid and apical segments, and apical crystals were implanted in the mid-portion of the LV apical segment. Anterior crystals were implanted at the septal side of the anterior LV wall, and lateral crystals were implanted close to the border with the anterior wall. The crystals were introduced in an oblique way to avoid damage to the myocardium to be studied. Other sonomicrometry crystals were implanted in the LV epicardium just radially so as to be paired with crystals in the subendocardium to calculate radial strain. In the 10 sheep, 6 crystals were implanted at the basal, mid, and apical anterior and lateral epicardial sites in 7 sheep; 4 crystals were implanted at basal and apical anterior and lateral epicardial sites in one sheep; and 2 crystals only were implanted in the mid anterior and lateral epicardial sites in 2 sheep. After each implant procedure, appropriate positioning of each crystal was confirmed by 2-D echocardiographic imaging. Crystals in the apical regions were implanted in the segments corresponding to territories supplied by the distal LAD, which would subsequently become ischemic segments following coronary artery ligation, and basal crystals were implanted in the nonischemic segments. Implant locations for mid crystals were in borderline areas at risk of ischemia, which was dependent on individual differences in bifurcation patterns of the diagonal artery branch.

Sonomicrometry recordings were made with the CardioSOFT Pro (Sonometrics Corp.). Strain was calculated as: $\text{strain} = L(t) - L_0 / L_0$, where $L(t)$ is the segment length at time t , and L_0 is the segment length at the onset of the QRS. Longitudinal strain (LS) of the LV mid segment was measured between the basal and mid endocardial crystals, and LS of the LV apical segment was measured between mid and apical endocardial crystals in each anterior and lateral wall. Circumferential strain (CS) of the LV basal, mid, and apical anterior segments was measured between each anterior and lateral endomyocardial crystal pair. Radial strain (RS) of the LV basal, mid, and apical segments was measured between each pair of endo- and epicardial crystals at the basal, mid, and apical anterior and lateral walls. All strain data were calculated by averaging data from 10 consecutive heart beats.

Echocardiography

Echocardiographic examinations were performed with an ArtidaTM ultrasound system (Toshiba Medical Systems Co., Tochigi, Japan). Full-volume ECG-gated 3D data sets were acquired from apical positions using a matrix array 2.5 MHz transducer, which was fixed in an ultrasound gel-filled latex bag and placed on the apical epicardium. To obtain these data sets, 4 or 6 sectors were scanned and automatically integrated into a wide-angle (70x70 degrees) pyramidal data image covering the entire LV. Frame rate of each image was set at approximately 30 Hz.

The data were stored and transferred to a computer (INSPIRON 1300, Dell Inc., Round Rock, TX) for off-line analysis. The images were analyzed with software (3D Wall Motion Tracking, Toshiba Medical Systems Co.) specific for the analysis of data acquired by the ArtidaTM. 3D data sets were displayed as multiplanar reconstruction

(MPR) images corresponding to apical 2-chamber and 4-chamber views and three short-axis levels (Figure 1). First, the endocardial border of the 4-chamber image at end-diastole was traced manually, followed by manual tracing of the epicardial border. Second, the same tracing processes were repeated in the 2-chamber image. After these long-axis tracings were complete, 3D myocardial surfaces were automatically reconstructed and fine adjustments were made to the traced borders on the short-axis images.

Wall motion tracking method

First, the tracking points are distributed on the 3D curved surfaces, which are estimated by traced lines on MPR images (Figure 1). Alternatively, motion estimation points where the image has features appropriate for tracking are located automatically in a region of myocardium in each volume frame. Each tracking point is moved based on the motion information obtained for nearby motion estimation points. The motion vector for each motion estimation point between consecutive volume frames is detected by template matching technique. In the template matching process, the template volume in the current frame is generated from an approximately 10×10×10-mm cube in which the motion estimation point is centered (Figure 2).²⁰ The most similar point in the next volume is searched for by comparing a template volume with the cube in the next volume. We used the 3D sum of squared differences method to test image similarity. Finally, interpolation of the motion vectors is performed with a 3D interpolation algorithm. After these steps, arbitrary points of interest on the cardiac wall can be tracked by integrating the interpolated motions over all frames during one cardiac cycle. We identified the tracking quality by eye based on the tracking quality of both end- and epimyocardial trace lines on MPR images.

Finally, 6 LV basal, 6 mid, and 4 apical segment measurements (total 16) of LS, CS, and RS were made. Strain was calculated as: $\text{strain} = L(t) - L_0 / L_0$, where $L(t)$ is the segment length at time t , and L_0 is the segment length at the peak of the R wave of the QRS complex. LS is defined as the percentage change in regional length in the direction of the longitudinal axis of the endocardium, CS as the percentage change in regional length in the circumferential direction of the endocardium, and RS as the percentage change in wall thickness in the direction perpendicular to the endocardium and away from the lumen. All strain data were measured by the data from one heart beat with good tracking quality. Temporal change of strain data was displayed as a color-coded “plastic bag” image, MPR images, and time-strain curves (Figure 3). LV volume was obtained directly from the tracked 3D endocardial surface information obtained by 3D-STI, and volumes were obtained from a single cardiac cycle with no assumptions made about LV shape. LV end-diastolic volume (LVEDV) was defined as the LV volume at end-diastole, and LV end-systolic volume (LVESV) was defined as the minimum LV volume measured during the cardiac cycle. LV ejection fraction (LVEF, %) was calculated by the formula: $(LVEDV - LVESV) * 100 / LVEDV$.

Comparison of Data Between 3D-STI and Sonomicrometry

LS at the LV mid and apical walls measured by 3D-STI was compared with LS measured by sonomicrometry between basal and mid endocardial crystal, and LS measured between mid and apical endocardial crystals, respectively, in each anterior and lateral wall. Basal, mid, and apical anterior CS values measured by 3D-STI were compared with CS measured by sonomicrometry between basal anterior and lateral crystals, CS measured between mid anterior and lateral crystals, and CS measured between apical anterior and lateral crystals, respectively. RS measured by 3D-STI at the

LV basal, mid, and apical walls was compared with RS measured by sonomicrometry between each endo- and epicardial crystal at the basal, mid, and apical segment in each anterior and lateral wall.

Experimental Protocol

After recording baseline measurements in the 10 sheep, sonomicrometric and echocardiographic measurements were repeated under continuous intravenous dobutamine infusion (2-3 $\mu\text{g}/\text{kg}/\text{min}$). In 8 sheep, after the end of dobutamine infusion and when vital signs had returned to resting conditions, recordings were repeated under intravenous infusion of propranolol (6-10 mg). After baseline recordings and pharmacological stress tests, ligation of the left anterior descending (LAD) coronary artery and its second diagonal branch was performed at 40% of the distance from the apex to the base of the heart. Recordings were repeated at 10 minutes after LAD occlusion. The mechanical ventilator was stopped for no more than 30 seconds to minimize the effects of variation in heart position caused by breathing while acquiring the 3D data set because tracking quality can be affected by stitching artifacts appearing at the sector borders.

Reproducibility

Six studies composed of 2 studies each of baseline, dobutamine infusion, and ischemic procedures were selected for the assessment of intra- and inter-observer reproducibility of CS, LS, and RS measurements. To test intra-observer variability, a single observer analyzed the data twice on occasions separated by an interval of 1 month. To test inter-observer variability, a second observer analyzed the data without knowledge of the first observer's measurements. Reproducibility was assessed as the mean percent error (absolute difference divided by the mean of the two observations).

Statistical Analysis

Results are expressed as the mean value \pm SD. A mixed model analysis was used to compare results among the variables obtained under the various conditions and among regions. When significant differences between groups were present, Bonferroni's test was used to compare individual groups. Agreement between sonomicrometry and 3D-STI-derived strain data was assessed by linear regression analysis with the Bland-Altman method.²¹ Comparisons between STI and corresponding sonomicrometry data were performed with a paired t-test. Reproducibility was assessed as the mean percent error (absolute difference divided by the mean of the two observations). A P value <0.05 was considered to indicate statistical significance. All calculations were performed with the SPSS 17 for Windows statistical program (SPSS Inc., Chicago, IL).

The authors had full access to and take full responsibility for the integrity of the data. All authors have read and agree to the manuscript as written.

Results

During off-line analysis, the endo- and epicardial borders on the MPR images corresponding to the apical 2- and 4-chamber views could be adequately traced within few minutes in all cases. Subsequently, the 3D-STI system provided strain measurements and LV volumetric data within approximately 10 seconds after manual tracing was completed. The entire time required to acquire and analyze 3D data sets was within 5 minutes for each image.

Hemodynamic and Echocardiographic Data

Hemodynamic and echocardiographic data obtained at baseline and during

pharmacological stress and coronary artery occlusion studies for the 10 sheep (body weight 36.4 ± 11.5 kg, range 27 to 57 kg) are summarized in Table 1. During dobutamine infusion tests, heart rate was significantly higher than during other test conditions, and systolic blood pressure and LVEF were significantly higher than the values recorded during propranolol infusion tests. During coronary artery occlusion tests, systolic blood pressure and LVEF were reduced compared to values recorded during the other test conditions.

Comparison of Strain Data

We obtained 152 LS, 118 CS, and 190 RS data sets in the 3D-STI studies and 136 LS, 108 CS, and 181 RS data sets in the sonomicrometry studies. Ultimately, 136 LS, 108 CS, and 175 RS data sets were completely comparable between the 3D-STI and sonomicrometry studies, and these data were used to compare strain measurements between the two methods. Both 3D-STI- and sonomicrometry-derived strain data obtained during baseline, pharmacological stress, and coronary artery occlusion studies are summarized in Table 2. During coronary artery occlusion studies, 2D echocardiographic examinations revealed that apical anterior and apical lateral wall motions were dramatically reduced due to akinesis or dyskinesis, where apical sonomicrometry crystals were placed. The 3D LV colored “plastic bag” images and polar map images of a representative case during coronary artery occlusion are shown in Figure 4. Apical wall motion abnormalities are clearly defined. Corresponding to these wall motion abnormalities, both STI- and sonomicrometry-derived LS and RS of the apical anterior and apical lateral walls and apical CS were reduced significantly during coronary artery occlusion studies compared to the other studies. In contrast, mid CS

derived from STI was reduced significantly during coronary artery occlusion studies, although mid CS derived from sonomicrometry during coronary artery occlusion studies did not differ. The RS derived from STI was greater than that derived from sonomicrometry at many regions: anterior-basal, lateral-mid, lateral-apical walls at baseline, anterior-apical, lateral-basal, and lateral-mid walls during dobutamine infusion tests, and the lateral-basal wall during propranolol infusion studies.

Correlations Between STI- and Sonomicrometry-Derived Strain

The correlation of all combined strain data under baseline, pharmacological stress, and coronary artery occlusion conditions between 3D-STI and sonomicrometry are summarized in panel A, and Bland-Altman analysis is shown in panel B of Figures 5-7, respectively. Strong correlations and good agreements between 3D-STI- and sonomicrometry-derived CS, LS, and RS were observed. Regional correlation between both methods is shown in Figure 8. Significant correlations are present between 3D-STI- and sonomicrometry-derived CS, LS, and RS at all levels assessed.

Reproducibility

Intra- and interobserver variability was 8.9% and 9.8% for CS measurements, 7.8% and 8.2% for LS measurements, and 13.5% and 15.4% for RS measurements, respectively.

Discussion

This experimental study is the first validation of the reliability of 3D-STI in assessing LV regional strain, and it shows that 3D-STI provides reasonable measurement of the lateral, circumferential, and radial strain components when compared with simultaneously recorded sonomicrometry data. The important finding of the present study is that on the basis of the three strain components calculated, 3D-STI has the potential to assess altered wall contractility induced by pharmacological stress test and coronary artery occlusion. In addition, the entire acquisition and analysis time for 3D data sets on this newly developed 3D-STI system may be acceptable to allow its use in the clinical setting.

The present study shows the accuracy of 3D-speckle tracking in animal experimental studies. Among affecters of strain measurements, the lower frame rate of 3D-STI is the most problematic factor in obtaining accurate strain data; this lower frame rate may cause large differences in speckle patterns between sequential frames. In this system's algorithm, differences in the speckle pattern of a cubic template are matched between sequential frames with minimization by the sum of squared differences.²⁰ The algorithm makes it possible to set a frame rate for each image of approximately 30 Hz, which is recommended as the minimum frame rate to limit frame-by-frame changes in the speckle pattern.¹¹ In addition, for 3D-STI, displacement of a target point is tracked as displacement of a volume of interest (cubic template) around the point. The size of this volume is another important factor that can affect tracking accuracy of the 3D-STI method.²² The size of the cubic template (approximately 10 mm on a side) in this 3D-STI method was chosen to achieve a balance between tracking accuracy and robustness at an ultrasound frequency of around 2.5 MHz. These developed algorithms

contribute to the reasonable agreement with sonomicrometry data, although higher heart rates in this experimental study were assumed to affect 3D tracking accuracy of regional myocardial motion.

The present study showed good correlation between the 3D-STI- and sonomicrometry-derived measurements of LS, CS, and RS. The correlation between 3D-STI and sonomicrometry was increased by changing the range of strain data by using the acute ischemic tests because combined data analysis has been used in previous studies to validate the correlation of strain data between 2D-STI and sonomicrometry.⁶⁻⁸ As in the previous validation studies of the reliability of 2D-STI-derived strain data,⁵⁻⁸ strain data measured with sonomicrometry has been used as control data. Myocardial strain data measured by sonomicrometry is associated with 3D myocardial deformation rather than with 2D deformation. Because sonomicrometry measures 3D strain, we considered it to be a better method to validate the accuracy of 3D-STI-derived strain data than that of 2D-STI, which has limitations such as misalignment between the ultrasound plane and the crystals, and out-of-plane motion.

In the present study, a maximum of 6 sonomicrometry crystal pairs (total of 12 crystals), which is more than in previous validation studies for 2D-STI,⁶⁻⁸ was implanted to assess regional deformation. Despite the relatively complex procedures used, the present study confirmed that 3D-STI could estimate regional CS and RS with acceptable accuracy in apical, mid, and basal regions, and LS in the apical and mid regions, respectively, of the left ventricle. Independence from regional variance in the ability to estimate myocardial deformation is an advantage in visualizing 3D regional wall function. However, in the apical region, worse correlations of CS and RS were observed compared to those in the basal and mid regions. The image quality of the

apical region was poorer than that of the mid or basal region because of its proximity to the near field of the ultrasound transducer. Therefore, image quality may be responsible for the worse correlations seen in the apical regions.

Among the strong correlations between 3D-STI and sonomicrometry, in particular, the CS and LS components showed better accuracy for estimation of regional deformation than did that of RS. In contrast, 3D-STI slightly underestimated CS compared to sonomicrometry. In the present study, the CS component is measured as the circumferential deformation of endomyocardium. The 3D-STI method estimated strain using cubic templates so that a cubic volume of interest at more epicardial sites may be tracked to estimate endocardial CS.²² Because strain at the endocardium is larger than that at the epicardium,^{23,24} 3D-STI may underestimate the CS component compared to sonomicrometry. However, these correlations and agreements suggest clinically acceptable accuracy for regional LS and CS components as compared to the previous studies for 2D-STI.

For RS measurements, poorer data agreement was observed compared to that of the CS and LS components. One reason for this poor agreement may be that RS is estimated by both endo- and epicardial speckle tracking so that RS measurements may be highly dependent on image quality compared to those of the LS and CS components, which are estimated only by endocardial speckle patterns. In addition, because the reference baseline length of the RS component is shorter than that of the CS or LS component, tracking accuracy between frames may affect the measurement of RS more greatly than that of CS or LS. In addition, the use of the open-chest model in this experimental study may also affect RS measurements. However, agreement of RS measurements between STI and sonomicrometry are also poor in previous studies for

2D-STI.^{6,8} Despite these limitations in the estimation of the RS component, the reasonable correlation between both techniques supports the fact that this new ultrasound method has the potential to estimate RS as well as sonomicrometry can.

In the present study, because ischemic areas were created by coronary ligation, myocardial wall motion in these areas was dramatically reduced mainly in the apical region. 3D-STI clearly could detect the reduced deformation in the 3 strain components. The good correlation of 3D-STI measurements with the regional reduction in strain measured by sonomicrometry was comforting to validate the ability of 3D-STI to detect regional change in myocardial deformation. The color-coded 3D-STI images (“plastic bag” and polar map images) were useful for monitoring the extent of the ischemic area.

Limitations

In our series, each study was performed at relatively high heart rate. Thus, the low frame rate of 3D-STI could cause miscorrelation between frames and possibly may have affected tracking quality and strain data.

Full-volume LV data sets for 3D-STI comprised 4 or 6 sectors. The artifact occurring around the border between sectors may affect speckle tracking quality. The main reason for this artifact is breathing. In this study, the mechanical ventilator was stopped to maintain the same heart position of the animal while acquiring the 3D data set.

STI depends on good image quality. In this experimental study, all 3D data were acquired via thoracotomy. Therefore, it was possible to obtain adequate 3D image quality to assess 3D-STI. In the clinical setting, however, the quality of the 3D images may be inferior to those in our experimental series, thus possibly affecting speckle

tracking accuracy.

In this study, only the anterior and lateral ventricular walls were evaluated for LS and RS, and only the anterior wall was evaluated for CS. Therefore, evaluation of these limited regions may be insufficient to estimate regional deformations in other regions.

3D-STI-derived strain measurements were not compared with 2D-STI-derived data. Therefore, further studies are needed to confirm the superiority of 3D-STI data over that of 2D-STI in the assessment of regional strain.

Conclusions

The newly developed technique of 3D-STI can estimate LV regional circumferential, longitudinal, and radial strain components with reasonable correlation to sonomicrometry data. 3D-STI methodology may be applied to accurately assess changes in myocardial function during pharmacological and ischemic interventions in LV segments between the LV apex and the base. In addition, these data can be acquired within a comfortable amount of acquisition and analysis time. Thus, 3D-STI may become a powerful tool to assess LV regional function in the clinical setting. In the future, the reliability of 3D-STI to assess regional myocardial function must be confirmed by clinical data.

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Disclosures

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References

1. Støylen A, Heimdal A, Bjørnstad K, Wiseth R, Vik-Mo H, Torp H, Angelsen B, Skjaerpe T. Strain rate imaging by ultrasonography in the diagnosis of coronary artery disease. *J Am Soc Echocardiogr.* 2000;13:1053-1064.
2. Urheim S, Edvardsen T, Torp H, Angelsen B, Smiseth OA. Myocardial strain by Doppler echocardiography. Validation of a new method to quantify regional myocardial function. *Circulation.* 2000;102:1158-1164.
3. Edvardsen T, Gerber BL, Garot J, Bluemke DA, Lima JA, Smiseth OA. Quantitative assessment of intrinsic regional myocardial deformation by Doppler strain rate echocardiography in humans: validation against three-dimensional tagged magnetic resonance imaging. *Circulation.* 2002;106:50-56.
4. Sutherland GR, Di Salvo G, Claus P, D'hooge J, Bijnens B. Strain and strain rate imaging: a new clinical approach to quantifying regional myocardial function. *J Am Soc Echocardiogr.* 2004;17:788-802.
5. Korinek J, Wang J, Sengupta PP, Miyazaki C, Kjaergaard J, McMahon E, Abraham TP, Belohlavek M. Two-dimensional strain--a Doppler-independent ultrasound method for quantitation of regional deformation: validation in vitro and in vivo. *J Am Soc Echocardiogr.* 2005;18:1247-1253.
6. Langeland S, D'hooge J, Wouters PF, Leather HA, Claus P, Bijnens B, Sutherland GR. Experimental validation of a new ultrasound method for the simultaneous assessment of radial and longitudinal myocardial deformation independent of insonation angle. *Circulation.* 2005;112:2157-2162
7. Amundsen BH, Helle-Valle T, Edvardsen T, Torp H, Crosby J, Lyseggen E, Støylen A, Ihlen H, Lima JA, Smiseth OA, Slørdahl SA. Noninvasive myocardial strain

- measurement by speckle tracking echocardiography: validation against sonomicrometry and tagged magnetic resonance imaging. *J Am Coll Cardiol.* 2006;47:789-793.
8. Reant P, Labrousse L, Lafitte S, Bordachar P, Pillois X, Tariosse L, Bonoron-Adele S, Padois P, Deville C, Roudaut R, Dos Santos P. Experimental validation of circumferential, longitudinal, and radial 2-dimensional strain during dobutamine stress echocardiography in ischemic conditions. *J Am Coll Cardiol.* 2008;51:149-157.
 9. Becker M, Hoffmann R, Kühl HP, Grawe H, Katoh R, Kramann R, Bücker A, Hanrath P, Heussen N. Analysis of myocardial deformation based on ultrasonic pixel tracking to determine transmuralty in chronic myocardial infarction. *Eur. Heart J.* 2006;27:2560-2566.
 10. Wang J, Khoury DS, Yue Y, Torre-Amione G, Nagueh SF. Preserved left ventricular twist and circumferential deformation, but depressed longitudinal and radial deformation in patients with diastolic heart failure. *Eur Heart J.* 2008;29:1283-1289.
 11. Suffoletto MS, Dohi K, Cannesson M, Saba S, Gorcsan J 3rd. Novel speckle-tracking radial strain from routine black-and-white echocardiographic images to quantify dyssynchrony and predict response to cardiac resynchronization therapy. *Circulation.* 2006;113:960-968.
 12. Delgado V, Ypenburg C, van Bommel RJ, Tops LF, Mollema SA, Marsan NA, Bleeker GB, Schalij MJ, Bax JJ. Assessment of left ventricular dyssynchrony by speckle tracking strain imaging comparison between longitudinal, circumferential, and radial strain in cardiac resynchronization therapy. *J Am Coll Cardiol.* 2008;51:1944-52.

13. Jenkins C, Bricknell K, Hanekom L, Marwick TH. Reproducibility and accuracy of echocardiographic measurements of left ventricular parameters using real-time three-dimensional echocardiography. *J Am Coll Cardiol.* 2004;44:878-886.
14. Kühl HP, Schreckenber M, Rulands D, Katoh M, Schäfer W, Schummers G, Bücken A, Hanrath P, Franke A. High-resolution transthoracic real-time three-dimensional echocardiography: quantitation of cardiac volumes and function using semi-automatic border detection and comparison with cardiac magnetic resonance imaging. *J Am Coll Cardiol.* 2004;43:2083-2090.
15. Sugeng L, Mor-Avi V, Weinert L, Niel J, Ebner C, Steringer-Mascherbauer R, Schmidt F, Galuschky C, Schummers G, Lang RM, Nesser HJ. Quantitative assessment of left ventricular size and function: side-by-side comparison of real-time three-dimensional echocardiography and computed tomography with magnetic resonance reference. *Circulation.* 2006;114:654-661.
16. Mor-Avi V, Sugeng L, Weinert L, MacEneaney P, Caiani EG, Koch R, Salgo IS, Lang RM. Fast measurement of left ventricular mass with real-time three-dimensional echocardiography: comparison with magnetic resonance imaging. *Circulation.* 2004;110:1814-1818.
17. Takeuchi M, Nishikage T, Mor-Avi V, Sugeng L, Weinert L, Nakai H, Salgo IS, Gerard O, Lang RM. Measurement of left ventricular mass by real-time three-dimensional echocardiography: validation against magnetic resonance and comparison with two-dimensional and m-mode measurements. *J Am Soc Echocardiogr.* 2008;21:1001-1005.
18. Kapetanakis S, Kearney MT, Siva A, Gall N, Cooklin M, Monaghan MJ. Real-time three-dimensional echocardiography: a novel technique to quantify global left

- ventricular mechanical dyssynchrony. *Circulation*. 2005;112:992-1000.
19. Kawagishi T. Speckle tracking for assessment of cardiac motion and dyssynchrony. *Echocardiography*. 2008;25:1167-1171.
 20. Ogawa K, Hozumi T, Sugioka K, Matsumura Y, Nishiura M, Kanda R, Abe Y, Takemoto Y, Yoshiyama M, Yoshikawa J. Usefulness of automated quantitation of regional left ventricular wall motion by a novel method of two-dimensional echocardiographic tracking. *Am J Cardiol*. 2006;98:1531-1537.
 21. Bland JM, Altman DG. Statistical methods for assessing agreement between two methods of clinical measurement. *Lancet*. 1986;1:307-310.
 22. Crosby J, Amundsen BH, Hergum T, Remme EW, Langeland S, Torp H. 3-D speckle tracking for assessment of regional left ventricular function. *Ultrasound Med Biol*. 2009;35:458-471.
 23. Bogaert J, Rademakers FE. Regional nonuniformity of normal adult human left ventricle. *Am J Physiol Heart Circ Physiol*. 2001;280:H610-20.
 24. Cheng A, Nguyen TC, Malinowski M, Daughters GT, Miller DC, Ingels NB Jr. Heterogeneity of left ventricular wall thickening mechanisms. *Circulation*. 2008;118:713-721.

Table 1. Hemodynamic and Echocardiographic Parameters

	Baseline	Dobutamine	Propranolol (n=8)	CA occlusion	P value
SBP, mmHg	102±14 ^{†#}	112±14 [¶]	95.8±13*	82.8±13	<0.001
DBP, mmHg	75.2±10 [†]	72.4±12 [†]	68.6±9.3 [†]	58.0±12	<0.001
HR, bpm	107±13	133±9.0 [¶]	106±8.7	109±19	<0.001
LVEDV, ml	38.0±7.5	35.0±4.7	39.1±8.5	39.8±5.6	1.0
LVESV, ml	18.4±3.7 [‡]	14.4±1.9	20.9±4.9 [‡]	25.0±3.2 [¶]	<0.001
LVEF, %	51.5±2.4 ^{†#}	57.1±5.1 [¶]	48.0±4.6 [†]	37.5±3.4	<0.001

Data are presented as the mean ± SD. P value means the overall P-value from the mixed model analysis.

CA, coronary artery; SBP, systolic blood pressure; DBP, diastolic blood pressure; HR, heart rate; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; LVEF, left ventricular ejection fraction.

*P<0.05 vs. CA occlusion, †P<0.01 vs. CA occlusion, #P<0.01 vs. propranolol, ‡P<0.01 vs. dobutamine,

¶ P<0.01 vs. other groups

Table 2. Comparisons of Strain Between STI and Sonomicrometry in Each Procedure

		Baseline	Dobutamine	Propranolol (n=8)	CA occlusion	P value
Longitudinal strain						
Total	STI	-12.6±3.7 [†]	-13.2±4.5 [†]	-9.7±5.1 [†]	-1.3±9.8	<0.001
	Sono	-15.6±5.0 ^{†¶}	-15.9±4.5 ^{†¶}	-11.4±5.0 [†]	0.2±14	<0.001
Ant-Mid	STI	-12.0±4.3	-11.5±4.1	-11.2±5.1	-7.4±9.5 [‡]	0.175
	Sono	-17.0±6.5 [¶]	-15.1±4.6 [¶]	-11.9±2.4	-7.2±15 [‡]	0.123
Ant-Apex	STI	-12.5±4.1 [†]	-14.0±3.1 [†]	-9.0±5.0 [†]	7.2±5.9	<0.001
	Sono	-14.9±2.1 [†]	-16.0±5.2 [†]	-11.3±7.2 [†]	9.3±5.4	<0.001
Lat-Mid	STI	-12.4±3.2	-14.0±4.9	-9.1±5.8	-8.9±8.0 [‡]	0.070
	Sono	-14.9±5.4	-17.1±4.4	-10.4±5.0	-11.4±14 [‡]	0.275
Lat-Apex	STI	-13.1±4.9 [†]	-14.4±4.7 [†]	-10.0±5.0 [†]	3.8±6.2	<0.001
	Sono	-15.2±4.4 [†]	-15.6±5.6 [†]	-11.5±5.2 [†]	7.9±10	<0.001
Circumferential strain						
Total	STI	-28.0±4.7 ^{†¶}	-31.8±6.0 ^{†¶#¶}	-21.0±6.9 [†]	-8.2±18	<0.001
	Sono	-25.2±6.5 [†]	-25.9±8.0 [†]	-19.7±9.4 [*]	-8.8±22	<0.001
Base	STI	-26.5±6.0	-27.0±5.9	-20.5±6.4	-23.8±7.8 [‡]	0.176
	Sono	-26.0±8.2	-26.9±9.4	-20.4±9.6	-25.8±16 [‡]	0.314
Mid	STI	-25.0±7.1	-27.2±4.1 [*]	-21.2±6.8	-15.1±11 [‡]	0.040
	Sono	-21.9±4.3	-23.7±5.4	-16.9±5.3	-16.5±15 [‡]	0.254
Apex	STI	-29.1±5.2 [†]	-32.9±4.9 [†]	-23.5±8.9 [†]	12.9±11	<0.001
	Sono	-27.1±7.2 [†]	-29.0±8.1 [†]	-23.8±11 [†]	12.3±15	<0.001
Radial strain						
Total	STI	24.1±4.0 ^{†¶}	28.1±8.4 ^{†¶#¶}	19.0±6.6 ^{†¶}	8.9±11	<0.001
	Sono	20.1±9.2 [†]	22.6±9.9 [†]	16.1±7.3 [†]	4.9±11	<0.001
Ant-Base	STI	21.5±4.1	23.5±8.8	13.9±9.0	17.1±9.5 [‡]	0.138
	Sono	16.7±9.7	22.0±6.0	14.2±9.7	14.1±7.3 [‡]	0.127
Ant-Mid	STI	26.0±7.4 [*]	30.5±7.7 [†]	18.9±6.5	6.4±10 [‡]	0.003
	Sono	23.2±9.1 [†]	27.7±4.2 [†]	17.6±6.6 [†]	3.2±7.1	<0.001
Ant-Apex	STI	24.8±6.9 [†]	26.0±8.4 [†]	20.1±2.3 [†]	-6.8±4.9	<0.001
	Sono	21.7±12 [†]	23.5±5.9 [†]	18.3±8.6 [†]	-6.1±3.1	<0.001
Lat-Base	STI	22.0±7.8	28.0±8.1	20.1±2.9	22.8±5.9 [‡]	0.204
	Sono	17.8±10	21.1±9.0	19.3±5.0	18.7±4.8 [‡]	0.325
Lat-Mid	STI	25.4±6.9 ^{*¶}	31.9±7.1 ^{†¶#¶}	18.5±3.5	17.7±8.5 [‡]	<0.001
	Sono	18.9±4.9	22.2±8.9	16.0±7.5	11.9±5.7 [‡]	0.143

Lat-Apex	STI	23.8±4.0 ^{†¶}	28.6±9.5 [†]	21.2±9.6 [†]	-5.8±2.3	<0.001
	Sono	15.8±7.9 [†]	23.1±12 [†]	16.6±7.4 [†]	-5.9±4.5	<0.001

Data are presented as the mean ± SD. P value means the overall P-value from the mixed model analysis.

CA, coronary artery; Ant, anterior; Lat, lateral; STI, speckle tracking imaging; Sono, sonomicrometry.

*P<0.05 vs. CA occlusion, †P<0.01 vs. CA occlusion, #P<0.01 vs. propranolol, ‡P<0.05 vs. apex data of same site during CA occlusion, ¶ P<0.05 vs. corresponding Sono data.

Figure legends

Figure 1. Three-dimensional speckle tracking of the left ventricle at end-diastole. The multiplanar reconstruction images correspond to the apical 2-chamber (right panel: B) and 4-chamber views (central panel: A) and three short-axis views at different levels (left panel: C3, C5, C7).

Figure 2. Speckle tracking with 3-dimensional pattern matching. Tracking of the volume of interest (white cubic template) from one volume (starting volume) to the next volume (red cubic template).

Figure 3. A representative parametric image of regional strain-time curves and the “plastic bag” image at end-systole of 3-dimensional tracking results (circumferential strain). Each color of strain-time curves is corresponding to data in 16 regional segments of left ventricle.

Figure 4. “Plastic bag” and polar map images at end-systole showing longitudinal strain (left panel), circumferential strain (central panel), and radial strain (right panel) during a coronary artery occlusion study. The blue area in the apex in each panel corresponds to dyskinetic motion induced by coronary artery occlusion. ant = anterior wall; ant-sept = anteroseptal wall; sept = septal wall; inf = inferior wall; post = posterior wall; lat = lateral wall.

Figure 5. (Left panel) Scatter plot showing the relation between all measurements of circumferential strain (CS) by sonomicrometry and 3-dimensional speckle tracking imaging (3D-STI). The solid line shows a regression line of all measurements. The dashed dotted line shows a regression line at baseline (\times), during dobutamine infusion (\circ), and during propranolol infusion (\square). The dashed line shows a regression line during the coronary artery occlusion (\triangle).

(Right panel) Bland-Altman plots for the comparison of all measurements of CS as measured with the 2 methods showing the mean differences (solid line) and 95% limits of agreement (dashed lines).

Figure 6. (Left panel) Scatter plot showing the relation between all measurements of longitudinal strain (LS) by sonomicrometry and 3-dimensional speckle tracking imaging (3D-STI). (Right panel) Bland-Altman plots for the comparison of all measurements of LS. Symbols and lines are the same definitions as Figure 5.

showing the mean differences (solid line) and 95% limits of agreement (dashed lines).

Figure 7. (Left panel) Scatter plot showing the relation between all measurements of radial strain (RS) by sonomicrometry and 3-dimensional speckle tracking imaging (3D-STI). (Right panel) Bland-Altman plots for the comparison of all measurements of RS. Symbols and lines are the same definitions as Figure 5.

Figure 8. Scatter plots showing the relation between regional measurements combined baseline, during dobutamine, and during propranolol infusion test.