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ORIGINAL PAPER

Usefulness of layer-specific strain in diagnosis of coronary artery disease in patients with stable angina pectoris

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Abstract
Novel software allows for layer-specific evaluation of myocardial strain by speckle tracking echocardiography (2DSTE). However, the potential of layer-specific strain at rest for diagnosing coronary artery disease (CAD) in patients with suspected stable angina pectoris (SAP) remains unknown. Our objective was to evaluate the usefulness of layer-specific 2DSTE at rest for diagnosis of CAD in patients with SAP. In total, 285 patients referred with clinically suspected SAP, normal ejection fraction, and no previous cardiac history were prospectively enrolled. All patients were examined by echocardiography, including 2DSTE, exercise ECG, and coronary angiography (CAG). Layer-specific 2DSTE was performed in three apical views to provide longitudinal peak systolic strains. Stenosis ≥ 70% in ≥ 1 major coronary artery on CAG was considered as significant CAD. Of 285 patients included, 104 had significant CAD (36%). Endocardial, epicardial, and mid-myocardial GLS were all significantly impaired in CAD patients ($P < 0.001$). Multivariable analysis including baseline clinical parameters, conventional echocardiographic measurements, Duke score, and layer-specific strain measurements revealed epicardial [odds ratio 1.19 ($P = 0.048$)] and mid-myocardial [odds ratio 1.16 ($P = 0.047$)] global longitudinal strain (GLS) as the only independent predictors of CAD. In direct comparison, epicardial and mid-myocardial GLS had a significantly higher diagnostic performance compared to endocardial GLS ($P = 0.038$ and $P = 0.031$, respectively). In conclusion, layer-specific GLS from 2DSTE at rest was significantly impaired in patients with significant CAD. In addition, epicardial and mid-myocardial GLS were independent predictors of CAD.

Keywords Angina · Coronary artery disease · Echocardiography · Speckle tracking

Introduction

Non-invasive imaging modalities are necessary for optimal decision on diagnosis and therapy in patients with CAD to improve patient outcome and healthcare costs. Most cardiac imaging techniques evaluate the entire wall thickness in the analysis of left ventricle (LV) function without further distinction, even though, the myocardium is non-homogenous and composed of three different layers of myocardial fibers ranging from the endocardium to the epicardium [1]. Thus, it would be encouraging if a careful layer-specific evaluation of myocardial deformation could improve the diagnostics of CAD in patients referred with suspected SAP.

Echocardiography is the widely-used imaging technique in patients with SAP for assessing regional wall motion abnormalities with level 1.B recommendation from the ESC guidelines on management of stable CAD [2]. It is possible to assess myocardial function and deformation by strain obtained from 2DSTE, which is a semi-automated quantitative technique based on echocardiographic grayscale images [3, 4]. Previous studies [5–8] have demonstrated 2DSTE at rest to be a more sensitive and objective marker of CAD than the conventional echocardiographic evaluation of LV systolic function. Recent software allows a layer-specific

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evaluation of myocardial strain by speckle tracking the endocardial and epicardial part of the myocardial wall separately, and the method has already been introduced in several clinical studies with promising results [9–12].

Basic first-line testing in patients suspected of having SAP includes biochemical testing, an ECG, and a resting echocardiography [2]. Further investigation and decision on subsequent revascularization therapy can include exercise testing [13, 14], although the diagnostic capability has been criticized [2, 15]. We are in need of a fast and simple diagnostic tool to facilitate risk stratification of patients with stable chest pain and to improve the rate of significant CAD in patients referred to CAG [16]. Hence, it would be appealing if the promising 2DSTE evaluation of layer-specific strain performed at rest could add incremental value to conventional echocardiography in first-line CAD diagnostics and, thereby, facilitate decision on treatment strategy in patients with suspected SAP.

Our group has previously shown that both longitudinal 2DSTE and color TDI at rest provides a thorough assessment of CAD in patients referred with suspected SAP [6, 17]. An independent evaluation of the myocardial layers from the newly developed technique allowing a layer-specific evaluation of myocardial function is likely to increase the pathophysiologic understanding of non-homogenous cardiac pathologies, such as ischemic injuries, and may improve the diagnostic accuracy of CAD. Consequently, the aim of this study was to evaluate the potential of layer-specific 2DSTE at rest for diagnosing CAD in patients with SAP.

Methods

Study population

This study prospectively included 285 consecutive patients referred with clinically suspected SAP during a 30-month period from September 2008 to March 2011. The study population has previously been described in detail elsewhere [17, 6]. In brief, patients were excluded if they had: (i) LVEF < 50%; (ii) arrhythmias; (iii) intraventricular conduction disturbance; (iv) pathological Q-waves; (v) congestive heart failure; (vi) heart valve disease; or (vii) known ischemic heart disease. All patients were examined by echocardiography, including 2DSTE, and exercise test followed by CAG. CAG was carried out regardless of the results of echocardiography and exercise test.

Conventional echocardiography and tissue doppler imaging

Echocardiograms were acquired using Vivid7 Dimension (GE Healthcare, Horten, Norway) with a 3.5-MHz transducer. All individuals were examined at rest using conventional 2-dimensional echocardiography by an experienced investigator blinded to the results of the exercise test, CAG, and clinical information.

LV diameter and wall thickness were obtained from the parasternal long-axis view at the level of the mitral valve leaflet tips, and LVMI was calculated using the LV linear dimensions as recommended by the American Society of Echocardiography [18]. LVEF was calculated using the modified biplane Simpson method. Left atrial volume was measured by the area-length method and divided by the body surface area to calculate the left atrial volume index [18]. Mitral inflow velocities were obtained using pulsed-wave Doppler at the apical position, and peak velocity of early (E) and atrial (A) diastolic filling and deceleration time (DT) of the E-wave were measured with the sample volume between the tips of mitral leaflets. The E/A-ratio was calculated. Lateral and septal mitral annular velocities in early diastole were obtained from pulsed-wave TDI in the 4-chamber view and averaged (e′) to calculate E/e′.

Two-dimensional speckle tracking echocardiography

2DSTE was performed off-line with commercially available software (EchoPac BT13, GE Healthcare, Horten, Norway) by a single investigator blinded to all other information (Fig. 1). Analysis of layer-specific GLS was performed in the apical long-axis, 4-chamber, and 2-chamber views by manually tracking the endocardial border in end-systole. The software then (1) automatically traced the entire endocardial border for the LV myocardium; (2) divided the myocardial wall into two separate layers (endocardial and epicardial) for a layer-specific deformation analysis, and 3) calculated endocardial GLS, epicardial GLS and mid-myocardial GLS (strain average through the whole width of the region of interest) by tracking of speckles throughout the cardiac circle frame by frame (Fig. 1). Analysis of GCS was performed in the parasternal short-axis view at mid-papillary level with same procedure as stated above. Frame rates were > 70 Hz for all grayscale images. Manual readjustment of tracking was performed if necessary to ensure accurate tracking and, subsequently, validated by the software. Any segment with persistently inadequate tracking was excluded. Eleven patients were excluded because of poor image quality and no possibility

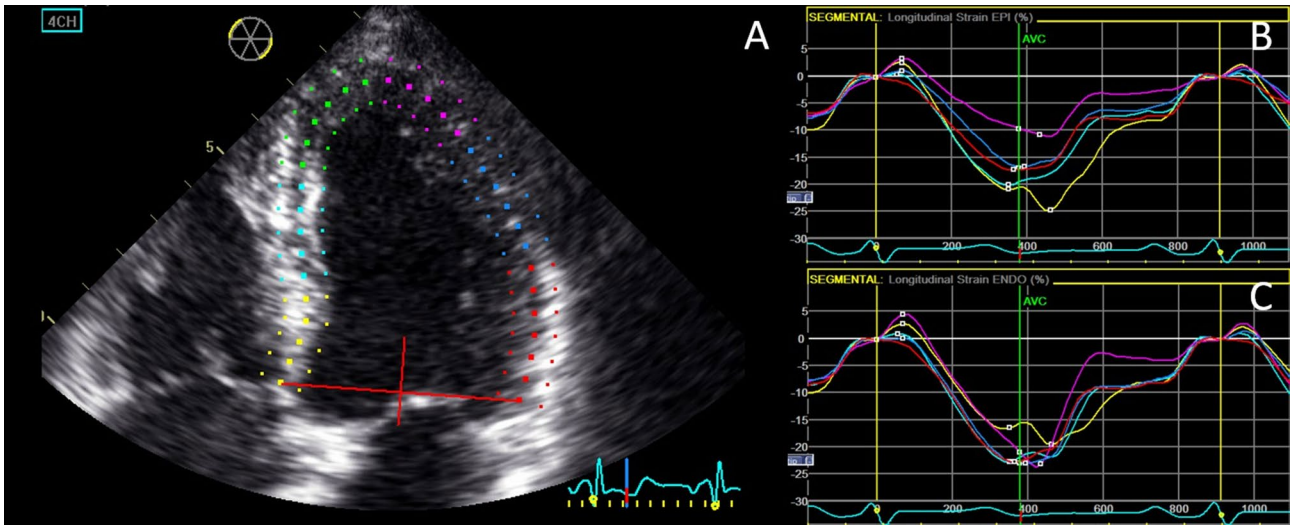

Fig. 1 Layer-specific analysis of echocardiographic image using 2-dimensional speckle tracking with EchoPac BT13. The echocardiographic example illustrates: (**A**) Layer-specific myocardial speckle tracking in 4-champer view; (**B**) analysis of epicardial global longitudinal strain; and (**C**) analysis of endocardial global longitudinal strain

of layer-specific speckle tracking. For improvement of objectivity and inter-operator reproducibility, strain analysis was performed according to previous established guidelines [19].

Exercise electrocardiography

All patients were examined by symptom-limited bicycle exercise testing in accordance with recommended standard bicycle exercise protocol [20]. Blood pressure, heart rate, and 12-lead ECGs were recorded at rest and at exercise every second minute. The ECGs were categorized as either normal or abnormal upon analysis from three investigators blinded to all other information. An abnormal exercise ST response was defined as ≥ 1 mm of horizontal or down sloping ST-depression (J-point + 80 ms) or ≥ 1 mm ST-segment elevation. In accordance with guidelines, Duke Score (DS) was used as outcome of the exercise test, and is considered as both a strong diagnostic and prognostic index combining information on ECG-alterations, exercise capacity, and symptoms during the test. DS was calculated as: exercise capacity − (5 × ST deviation) − (4 × exercise angina). Exercise angina had a value of 0 if the patients did not have any angina during the test, 1 if the patient had non-limiting angina, and 2 if the patient had to stop the test because of angina. ST-deviation was the maximum ST-deviation during or after exercise measured in millimeters [15, 20, 21].

Coronary angiography

All patients underwent CAG, and the procedure was performed by the percutaneous femoral approach. Coronary angiograms were gained for each coronary vessel in ≥ 2 projections, and stenosis with ≥ 70% reduction of the arterial lumen area in at least one major coronary artery was considered significant. Analysis of the coronary angiograms was performed visually by an experienced operator blinded to all other results.

Reproducibility

Intra- and inter-observer variability was assessed in 20 randomly selected patients by CV of repeated analyses and by Bland–Altman analysis deriving the absolute bias including 95% limits of agreement. For intra-observer variability, the original investigator reanalyzed the same echocardiographic images of the 20 patients selected at random. For inter-observer variability, the same echocardiographic images from the same cardiac cycles were analyzed by another experienced investigator also blinded to all other results.

Statistical analysis

Statistical analysis was performed using STATA Statistics/Data analysis, SE 12.0 (StataCorp, TX). A P value < 0.05 was considered statistically significant. Continuous data was expressed as means ± standard deviation and compared by Student's *t* test. Categorical data was expressed as absolute numbers and percentages and compared by the χ^2test. Logistic regressions were performed to adjust for baseline characteristics (age, sex, diabetes mellitus, hypercholesterolemia, heart rate, diastolic blood pressure, and angina type), conventional echocardiographic parameters (LVMI, DT, E/A, and e′), and exercise test. ROC curves were constructed

for GLS measures, and the optimal cut-off values with the greatest sensitivity and specificity in the prediction of CAD were selected. AUC was calculated and compared for the above parameters using the roccomp function [22]. Continuous net reclassification index (NRI) was performed to investigate the additive value of layer-specific GLS to clinical and conventional echocardiographic parameters in diagnosis of CAD. Multiple linear regressions were built to test significant stenosis in LAD, LCX, or RCA as independent predictors of RLS in all 18 myocardial segments. ANOVA was carried out to test whether layer-specific GLS developed with increasing severity of CAD.

Ethics

The regional Committee on Biomedical Research Ethics (j.no. H-C-2008–044) approved the study, and informed consent was obtained from all individual participants included in the study.

Results

Out of the 285 patients included in this study, 104 had significant CAD and 181 had nonsignificant or no CAD. Table 1 presents all clinical characteristics and data on coronary angiography, ECG exercise testing, and echocardiography including 2DSTE.

The exercise performance of patients with significant CAD were significantly impaired in all aspects of the test including exercise capacity, ST deviation, angina during the test, and the composite index expressed as DS (Table 1.C). The extent of disease in patients with CAD varied from 1-vessel disease to 3-vessel disease (3VD) with 31% having 3VD, and the most frequently diseased territory was found to be the LAD (74% of CAD patients) (Table 1.B). Conventional echocardiographic measures were significantly impaired in patients with significant CAD in terms of diastolic function (A, E/A, DT, e′, and E/e′) and LVMI (Table 1.D). Endocardial, epicardial, and mid-myocardial GLS were all significantly impaired in CAD patients. No significant differences were found regarding the GCS measures (Table 1.E).

Multivariable logistic regression models including GLS (either epicardial, endocardial or mid-myocardial) or GCS (either epicardial, endocardial or mid-myocardial), baseline characteristics (age, sex, diabetes mellitus, angina type), Duke Score, and conventional echocardiographic variables (LVMI, E/A ratio, e′, DT) revealed epicardial GLS [odds ratio (OR) 1.19, 95% CI 1.00–1.41, P = 0.048] and mid-myocardial GLS (OR 1.16, 95% CI 1.00–1.35, P = 0.047) as the only echocardiographic parameters independently associated with significant CAD (Table 2).

Value of global longitudinal strain to diagnose coronary artery disease

ROC curves were created to determine the diagnostic performance of GLS parameters (Fig. 2). In direct comparison, both epicardial GLS and mid-myocardial GLS had a significantly higher diagnostic performance compared to endocardial GLS (P = 0.038 and P = 0.031, respectively), determined by AUCs. The optimal cut-off value leading to the highest sensitivity and specificity for diagnosing CAD was − 18.0% for endocardial GLS, with a sensitivity of 57% and a specificity of 73%; − 15.1% for epicardial GLS, with a sensitivity of 67% and a specificity of 62%; and − 16.0% for mid-myocardial GLS, with a sensitivity of 59% and a specificity of 71%. All layer-specific GLS parameters improved NRI when added to baseline characteristics, reclassifying 47–58% (continuous increase in NRI: endocardial GLS: 0.47, 95% CI 0.16–0.81; epicardial GLS: 0.58, 95% CI 0.16–0.91; mid-myocardial GLS: 0.55 95% CI 0.19–0.86). In comparison, adding GLS parameters to both baseline characteristics and conventional echocardiographic parameters did not result in a better model (continuous increase in NRI: endocardial GLS: 0.49, 95% CI − 0.12–0.87; epicardial GLS: 0.51, 95% CI − 0.10–0.91; mid-myocardial GLS: 0.52 95% CI − 0.10–0.93).

Disease severity, regional coronary artery disease, and regional longitudinal strain

Layer-specific GLS and the severity of significant CAD followed a significant trend from no CAD to 3VD in terms of a progressively impaired GLS (P < 0.001) (Table 3). Analysis of significant CAD in the LAD, RCA, and LCX and RLS from multiple linear regression models are shown in Fig. 3. Significant association between RLS measures and CAD was seen in about half of the myocardial segments mimicking the typical anatomic distribution of the major coronary arteries (Fig. 3a).

Reproducibility of two-dimensional strain echocardiography

Reproducibility of GLS was as follows: Intra-observer Bland–Altman analysis showed a bias of (mean ± SD difference) 0.502 ± 0.944 and a CV of 5.2% for endocardial GLS; a bias of 0.379 ± 0.967 and CV of 6.9% for epicardial GLS; and a bias 0.444 ± 0.823 and CV of 5.2% for mid-myocardial GLS. The inter-observer Bland–Altman analysis showed a bias of (mean ± SD difference) 0.636 ± 1.89 and CV of 10.2% for endocardial GLS; a bias of 0.351 ± 1.34 and CV of 9.3% for epicardial GLS; and a bias of 0.472 ± 1.57 and CV of 9.6% for mid-myocardial GLS. Reproducibility of GCS was as follows:

Table 1 Clinical and paraclinical characteristics

N	Patients with no significant CAD	Patients with significant CAD	P value
	181	104	
A. Clinical characteristics			
Age, years	59.3 ± 9.7	63.8 ± 10.0	**< 0.001**
Female, n (%)	116 (64%)	27 (26%)	**< 0.001**
Hypertension, n (%)	96 (53%)	48 (46%)	0.26
Diabetes mellitus, n (%)	18 (10%)	20 (19%)	**0.026**
BMI, kg/m^2	26.1 ± 4.3	26.9 ± 4.3	0.15
Hypercholesterolemia, n (%)	61 (34%)	42 (40%)	0.26
Smoker, n (%)			
Current	35 (19%)	25 (24%)	0.52
Previous	71 (39%)	42 (40%)	
Family history of CAD, n (%)	89 (49%)	62 (60%)	0.09
Anticoagulant therapy, n (%)	0	1 (1%)	0.19
Platelet inhibitors, n (%)	27 (15%)	23 (22%)	0.25
B-Blockers, n (%)	28 (16%)	20 (19%)	0.41
Calcium channel blockers, n (%)	31 (17%)	19 (18%)	0.81
Nitrates, n (%)	13 (7%)	6 (6%)	0.65
Angina, n (%)			
Typical	66 (36%)	83 (80%)	**< 0.001**
Atypical	73 (40%)	14 (13%)	
Nonanginal chest pain	42 (23%)	7 (7%)	
ECG heart rate, bpm	69 ± 11	69 ± 12	0.77
Diastolic blood pressure, mmHg	81 ± 10	80 ± 11	0.42
B. Coronary angiography			
1—vessel disease	–	44 (42%)	–
2—vessel disease	–	28 (27%)	–
3—vessel disease	–	32 (31%)	–
LAD, n (%)	–	75 (72%)	–
LCX, n (%)	–	58 (56%)	–
RCA, n (%)	–	63 (61%)	–
C. ECG exercise test			
Exercise capacity (METs)	7.0 ± 1.8	6.2 ± 1.6	**< 0.001**
ST deviation > 1 mm	40 (22%)	60 (58%)	**< 0.001**
Angina pectoris			
Occurred	49 (27%)	39 (38%)	**< 0.001**
Stopped the test	1 (1%)	14 (14%)	
Duke score	4.4 ± 3.6	− 2.1 ± 7.2	**< 0.001**
D. Echocardiographic parameters			
LVEF, %	59 ± 5	58 ± 4	0.08
LVld, cm	4.5 ± 0.5	4.6 ± 0.5	0.45
LVMI, g/m^2	76.2 ± 16.1	86.7 ± 16.6	**< 0.001**
LAVI, mL/m^2	27.5 ± 8.4	29.4 ± 8.9	0.07
E, m/s	0.7 ± 0.2	0.7 ± 0.2	0.06
A, m/s	0.7 ± 0.2	0.8 ± 0.2	**0.027**
E/A	1.1 ± 0.3	1.0 ± 0.2	**< 0.001**
DT, ms	226.4 ± 50.3	242.4 ± 58.4	**0.019**
e′, cm/s	8.5 ± 2.0	7.4 ± 1.9	**< 0.001**
E/e′	11.6 ± 2.9	10.7 ± 3.1	**0.02**
E. 2DSTE parameters			
Endocardial GLS, %	− 19.5 ± 3.1	− 17.8 ± 3.2	**< 0.001**

Table 1 (continued)

N	Patients with no significant CAD	Patients with significant CAD	P value
	181	104	
Epicardial GLS, %	− 15.7 ± 2.6	− 14.2 ± 2.3	**< 0.001**
Mid-myocardial GLS, %	− 17.4 ± 2.8	− 15.7 ± 2.7	**< 0.001**
GLS absolute difference	− 3.8 ± 1.3	− 3.6 ± 1.5	0.40
Endocardial GCS, %	− 28.6 ± 6.9	− 26.7 ± 7.9	0.07
Epicardial GCS, %	− 12.2 ± 5.9	− 10.9 ± 5.4	0.13
Mid-myocardial GCS, %	− 18.7 ± 6.0	− 17.1 ± 6.1	0.08

Continuous data are expressed as mean ± SD and categorical data are presented as number (%)

BMI body mass index, *CAD* coronary artery disease, *ECG* electrocardiography, *LAD* left anterior descending artery, *LCX* left circumflex artery, *RCA* right coronary artery, *Duke score* exercise capacity − (5 × ST deviation)– − 4 × angina score), *LVEF* left ventricular ejection fraction, *LVId* left ventricular internal diameter in diastole, *LVMI* left ventricular mass index, *LAVI* left atrial volume index, *E* peak transmitral early diastolic inflow velocity, *A* peak transmitral late diastolic inflow velocity, *DT* deceleration time of early diastolic transmitral inflow, *e′* average peak early diastolic longitudinal mitral annular velocity, *GLS* global longitudinal strain, *GCS* global circumferential strain

Values significantly different from patients with no significant CAD are marked in bold ($P \leq 0.05$)

Table 2 Multivariable stepwise logistic regression including GLS and GCS parameters, baseline characteristics, Duke Score, and conventional echocardiographic variables for assessment of the association with significant CAD

	Endocardial GLS	Epicardial GLS	Mid-myocardial GLS	Endocardial GCS	Epicardial GCS	Mid-myocardial GCS
Adjustment for baseline characteristics (age, sex, diabetes mellitus, and angina type)						
OR (95% CI)	1.11 (1.01–1.21)	1.13 (1.00–1.27)	1.13 (1.01–1.26)	1.03 (0.98–1.08)	1.05 (0.99–1.11)	1.05 (0.99–1.11)
P value	0.032	0.045	0.032	0.229	0.133	0.134
Adjustment for baseline characteristics (age, sex, diabetes mellitus, and angina type) and Duke score						
OR (95% CI)	1.16 (1.04–1.29)	1.23 (1.07–1.42)	1.21 (1.06–1.37)	1.05 (0.99–1.11)	1.06 (0.99–1.14)	1.07 (0.99–1.14)
P value	0.007	0.004	0.004	0.095	0.102	0.070
Adjustment for baseline characteristics (age, sex, diabetes mellitus, and angina type), Duke score, and conventional echocardiographic measures (LVMI, E/A ratio, e′, DT)						
OR (95% CI)	1.13 (0.99–1.28)	1.19 (1.00–1.41)	1.16 (1.00–1.35)	1.04 (0.97–1.12)	1.07 (0.98–1.17)	1.07 (0.98–1.17)
P value	0.062	0.048	0.047	0.233	0.156	0.143

Data are expressed as odds ratio (OR) including 95% CI

In addition to epicardial and mid-myocardial GLS, Duke score (OR 0.77, 95% CI 0.70–0.85, P < 0.001), gender (OR 0.22, 95% CI 0.09–0.54, P = 0.001), and angina type (OR 0.25, 95% CI 0.1–0.64, P = 0.004) were also independent predictors of significant CAD

GLS global longitudinal strain, *GCS* global circumferential strain, *CAD* coronary artery disease, *BMI* body mass index, *Duke score* exercise capacity − (5 × ST deviation) − (4 × angina score), *LVMI* left ventricular mass index, *E* peak transmitral early diastolic inflow velocity, *A* peak transmitral late diastolic inflow velocity, *DT* deceleration time of early diastolic transmitral inflow, *e′* average peak early diastolic longitudinal mitral annular velocity

Intra-observer Bland–Altman analysis showed a bias of (mean ± SD difference) − 1.95 ± 2.97 and a CV of 10.1% for endocardial GCS; a bias of 0.871 ± 4.95 and CV of 36.9% for epicardial GCS; and a bias − 0.153 ± 3.50 and CV of 17.7% for mid-myocardial GCS. The inter-observer Bland–Altman analysis showed a bias of (mean ± SD difference) 2.97 ± 4.47 and CV of 15.0% for endocardial GCS; a bias of 3.72 ± 4.42 and CV of 28.2% for epicardial GCS; and a bias of 3.16 ± 4.03 and CV of 19.0% for mid-myocardial GCS.

Discussion

The main findings of the present study can be summarized as follows: (1) global myocardial function, as assessed by layer-specific 2DSTE at rest, was significantly impaired in both myocardial layers in addition to mid-myocardial GLS in patients with significant CAD amongst patients with preserved LVEF and suspected of having SAP; (2) multivariable logistic regression revealed epicardial and

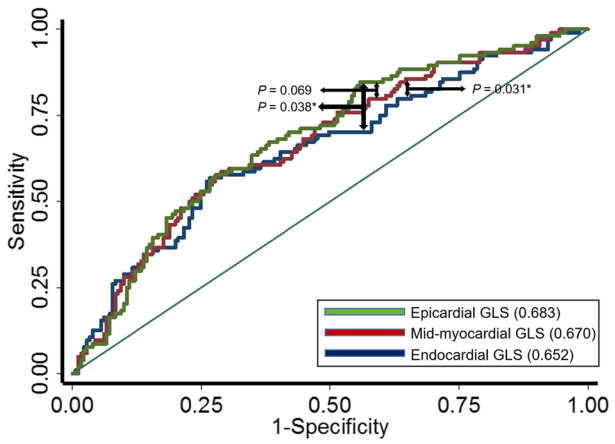

Fig. 2 Receiver operating characteristic curves and area under the curve of layer-specific and mid-myocardial global longitudinal strain for diagnosing coronary artery disease in patients with suspected stable angina pectoris

mid-myocardial GLS as the only independent and strongest echocardiographic predictors of significant CAD; (3) in direct comparison, epicardial and mid-myocardial GLS were significantly superior to endocardial GLS in diagnosis of CAD as determined by AUCs, however, layer-specific GLS only possessed a modest accuracy of CAD.

2DSTE and GLS have already been proven useful for detection of CAD and subclinical LV dysfunction in patients referred with SAP and a normal LVEF, solely from a resting echocardiographic examination [6, 5, 23, 24]. Recently, two studies have investigated the diagnostic capability of layer-specific 2DSTE at rest [9, 12] using the current edition of commercially available EchoPac software. However, the previous studies of layer-specific 2DSTE have included smaller populations and with various types of CAD ranging from patients with already known CAD and LV dysfunction [9] to patients admitted with acute coronary syndrome [12]. Similar structural limitations exist in related studies using layer-specific speckle tracking software from different manufactures [10, 11]. In the present study, the 285 consecutive patients referred only with suspected angina pectoris were included prospectively, had no previous cardiac history, a relatively low prevalence of cardiac risk factors, and a normal conventional echocardiography. Furthermore, none of the previous studies have included an exercise test, conventional echocardiography, and CAG in the diagnostic workup, which all together makes our population more homogenous, less confounded, and in greater resemblance to the challenging clinical reality. Overall disease prevalence was low (36% of patients had significant CAD), and the fact that layer-specific 2DSTE performed at rest was found to be an independent predictor of CAD even in a low-risk population with preserved LVEF could suggest that layer-specific 2DSTE may be a stronger predictor of CAD in a population with higher risk of CAD.

Our group has previously demonstrated both color TDI and 2DSTE performed at rest as sensitive markers of significant CAD in patients referred with suspected SAP [6, 17]. GLS was confirmed as an independent predictor of CAD with incremental value compared with exercise testing and conventional echocardiography in diagnosis of CAD [6]. However, recent innovation in speckle tracking software allows for a layer-specific evaluation of myocardial strain, challenging the already validated transmural deformation analysis. To our knowledge, this is the first study to investigate the value of layer-specific 2DSTE at rest and the relationship between GLS from separate myocardial layers in consecutive patients referred with suspected SAP, preserved LVEF, and no cardiac history.

To investigate the correlation between CAD and layer-specific RLS, significant stenosis in the LAD, RCA, and LCX were tested as independent predictors of reduced RLS in each segment by performing multiple linear regression models. RLS provides the clinician with an assessment of the local consequences of an ischemic injury and the possibility of identifying the occluded arterial branch responsible. Since coronary anatomy has variability [25], stenotic segments were not assigned to predefined vascular territories as they may not reflect the right coronary artery distribution. Significant correlations between CAD and RLS were seen in about half of the myocardial segments in a pattern mimicking the typical perfusion pattern of major coronary arteries. RLS in the non-significant segments could have been affected by stenotic arteries in adjacent segments [26]

Table 3 The severity of CAD and the corresponding longitudinal strain values. Patients are divided into groups with no CAD, with singlevessel disease (1VD), and with multivessel disease (2VD and 3VD)

	No CAD	1VD	2VD	3VD	P value for trend
Endocardial GLS, %	− 19.5 ± 3.1	− 18.7 ± 3.1	− 17.5 ± 3.4	− 16.9 ± 3.2	< 0.001
Epicardial GLS, %	− 15.7 ± 2.6	− 14.9 ± 2.3	− 14.2 ± 2.4	− 13.3 ± 2.1	< 0.001
Mid-myocardial GLS, %	− 17.4 ± 2.8	− 16.5 ± 2.5	− 15.6 ± 2.8	− 14.8 ± 2.6	< 0.001

Continuous data are expressed as mean ± SD, and categorical data are presented as frequency (%)

CAD coronary artery disease, *GLS* global longitudinal strain, *1VD* 1-vessel disease, *2VD* 2-vessel disease, *3VD* 3-vessel disease

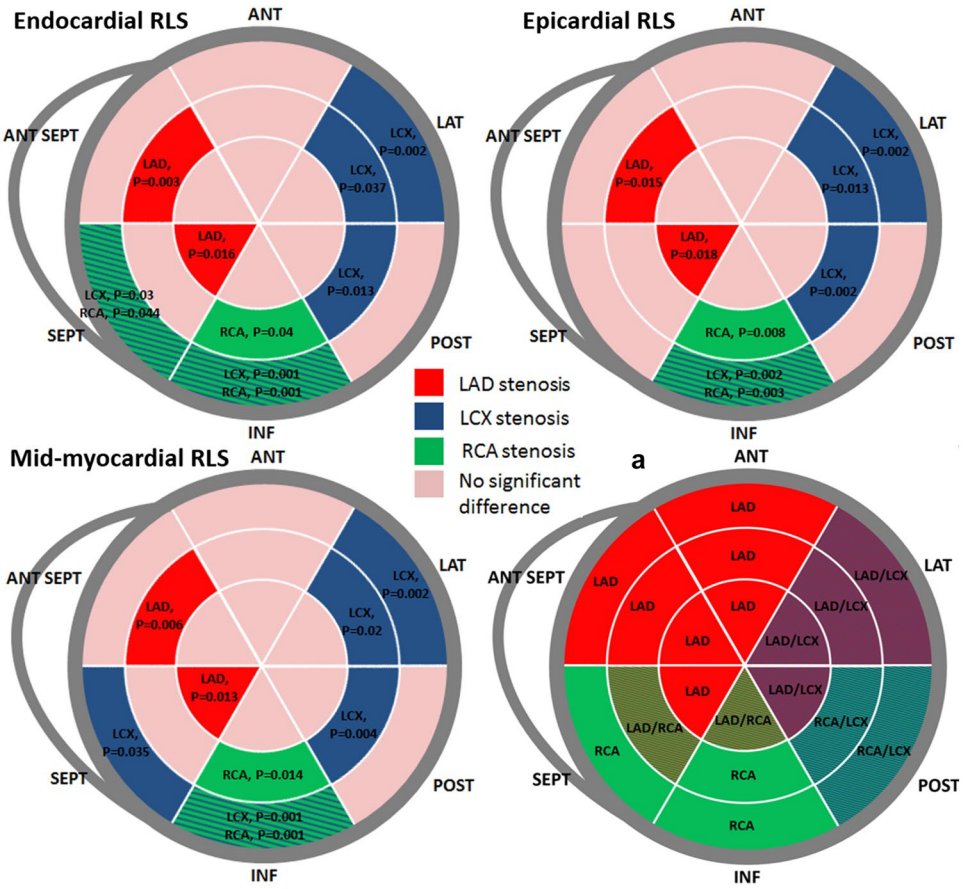

Fig. 3 Association between RLS and regional CAD as assessed by coronary angiography. Multiple linear regression models were constructed, and significant CAD (n = 104) in the LAD (72%), RCA (61%), and LCX (56%) were tested as independent predictors of RLS in each of the 18 segments, in order to illustrate the association between RLS and regional CAD. A P value < 0.05 when comparing segments supplied by diseased coronary arteries and non-diseased arteries was considered statistically significant and colored red, green, or blue. Similar analysis has been validated in a previous study of GLS done by our group [6]. (**a**) illustrates the typical perfusion patterns of LAD, LCX, and RCA [18]. *ANT* Anterior, *ANT SEPT* Anterior septal, *CAD* coronary artery disease, *INF* Inferior, *LAD* Left anterior descending, *LAT* Lateral, *LCX* Left circumflex coronary artery, *POST* Posterior, *RCA* Right coronary artery, *RLS* Regional longitudinal strain, *SEPT* Septal

or microvascular communications between territories, since the majority of included patients had multivessel CAD. In the present study, layer-specific GLS seemed to avoid this inaccuracy and has earlier been proven to be an excellent predictor of global infarct size [26]. Thus, a low GLS at rest in patients with suspected SAP and a normal conventional echocardiography must lead to further investigation, and analysis of RLS should only assist the global strain assessment. In addition, layer-specific GLS followed a significant trend in terms of a progressively impaired GLS from no CAD to 3VD. Thus, layer-specific GLS seemed capable of identifying significant CAD based on the severity, and the risk of multivessel disease increased with decreasing GLS.

In the present study, layer-specific GCS failed to show any significant association with CAD. This contrasts with previous studies [12, 11], although GLS seemed to be stronger associated with CAD than GCS [12]. It has previously been shown that longitudinal myocardial fibers are most susceptible to ischemia [10–12], and that circumferential strain can remain relatively unaffected or even increase as a compensation to a decreased GLS in the effort to maintain LVEF within the normal range [27, 28], which may explain our finding of a preserved GCS in patients with CAD. The division of myocardial layers is not absolute, and strain is a result of an active contraction within the layer and a passive motion from neighboring myocardium [29, 11]. Thus, the complex co-dependent contractile function of myocardial layers may benefit from a layer-specific assessment of both regional and global longitudinal strain, adding essential information to the assessment of LVEF in patients with suspected SAP.

The present study demonstrates impaired deformation in both myocardial layers including the entire wall in patients with significant CAD and a decreasing absolute strain from the endocardium to epicardium in both patients with and without significant CAD in line with previous studies [10, 29, 11, 12, 30]. The decreasing absolute strain from the endocardium to epicardium is a result of a gradient of thickening across the myocardial wall. The endomyocardial region undergoes the largest deformation of the healthy myocardium, contributing to the largest part of LV contraction [31, 32]. Therefore, inhomogeneous pathologic changes are expected in patients with CAD, and previous studies have shown that the endomyocardial region consisting of longitudinal fibers was most susceptible to the ischemic injury [10–12]. In the present study, epicardial GLS and mid-myocardial GLS appeared slightly superior to endocardial GLS in diagnosing CAD, however, the AUCs and related

cut-off values demonstrated only a modest accuracy of CAD diagnosis. Furthermore, both endocardial, epicardial, and mid-myocardial GLS increased the NRI when added to a model with baseline characteristics. Our population differs significantly from previous studies investigating layer-specific 2DSTE [10–12], which might explain the different results to some extent. However, a recently published study of layer-specific GLS from our group including 80 patients with SAP, preserved LVEF, and reversible ischemia assessed by single-photon emission computed tomography showed the same notable finding, indicating epicardial GLS as a better predictor of significant CAD than endocardial GLS [33]. Another recently published study from our group demonstrated epicardial and mid-myocardial GLS as superior prognostic predictors of heart failure and cardiovascular death following acute coronary syndrome [34]. The reason several new studies found epicardial GLS as a superior diagnostic and prognostic parameter may be due to technical aspects such as: (1) superior tracking of the epicardial region; (2) less variance of epicardial GLS from base to apex as compared to endocardial GLS [29]; (3) better reproducibility of the epicardial layer [33, 34]. Yet, the notable finding will require further investigation since it contradicts the pathophysiological understanding of ischemic injuries. In line with previous studies [14, 11, 12], the present study demonstrated a lower absolute difference between endocardial and epicardial GLS in patients with significant CAD, reflecting a decreased function of the longitudinal fibers in the endocardial layer, even though the difference was not significant. In patients with SAP, cumulative stunning and recurrent ischemia can lead to permanent LV dysfunction [35], detectable from solely a resting echocardiography [6, 23, 24, 5]. Resting layer-specific GLS is a fast and simple echocardiographic tool, easily applicable in a clinical setting, and offers additional information to conventional echocardiographic parameters in diagnosis of CAD.

Limitations

Wall motion abnormalities is not unique for CAD and may be due to other conditions such as heart valve disease, interventricular conduction disturbance, arterial hypertension, diabetes mellitus, age, and sex. However, to avoid confounding from co-existence of these conditions, patients with heart valve disease and interventricular conduction disturbance were excluded, and adjusted analysis for significant confounders were carried out. Selection bias may have occurred, even though patients were enrolled consecutively. Since patients were excluded if having a history of heart disease, an abnormal ECG, or a LVEF < 50%, the enrolled patients were at relatively low risk of CAD. The strict exclusion criteria may, therefore, restrict extrapolation of the present findings to the general patient population referred with suspected SAP.

Longitudinal and circumferential, but not radial, strain was included in this study. We chose to exclude radial strain from the analysis since it has been proven inferior to the two other parameters in identifying CAD and possess several methodological limitations [36].

We used CAG as the reference standard for assessment of layer-specific 2DSTE's ability in CAD diagnostics since data on fractional flow reserve was not available. A comparison of a physiological and an anatomical method is not ideal, since the presence of an anatomical stenosis is not necessarily associated with myocardial ischemia and vice versa. Considerations on whether layer-specific 2DSTE would have an improved diagnostic performance compared to physiological test is therefore relevant. Despite this limitation, the fraction of patients with non-flow limiting stenosis may have been reduced since a high cut-off for significant CAD was used. Analysis of CAG were performed visually, and not quantitative, by an experienced investigator as was clinical practice at the time of the study enrollment period.

Conclusion

In patients with clinically suspected SAP and preserved LVEF, layer-specific GLS as assessed by 2DSTE at rest was significantly impaired in all myocardial layers amongst patients with significant CAD. In addition, epicardial and mid-myocardial GLS were independently associated with the presence of significant CAD.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Maciver DH (2012) The relative impact of circumferential and longitudinal shortening on left ventricular ejection fraction and stroke volume. Exp Clin Cardiol 17(1):5–11
2. Task Force M, Montalescot G, Sechtem U, Achenbach S, Andreotti F, Arden C, Budaj A, Bugiardini R, Crea F, Cuisset

T, Di Mario C, Ferreira JR, Gersh BJ, Gitt AK, Hulot JS, Marx N, Opie LH, Pfisterer M, Prescott E, Ruschitzka F, Sabate M, Senior R, Taggart DP, van der Wall EE, Vrints CJ, Guidelines ESCCfP, Zamorano JL, Achenbach S, Baumgartner H, Bax JJ, Bueno H, Dean V, Deaton C, Erol C, Fagard R, Ferrari R, Hasdai D, Hoes AW, Kirchhof P, Knuuti J, Kolh P, Lancellotti P, Linhart A, Nihoyannopoulos P, Piepoli MF, Ponikowski P, Sirnes PA, Tamargo JL, Tendera M, Torbicki A, Wijns W, Windecker S, Document R, Knuuti J, Valgimigli M, Bueno H, Claeys MJ, Donner-Banzhoff N, Erol C, Frank H, Funck-Brentano C, Gaemperli O, Gonzalez-Juanatey JR, Hamilos M, Hasdai D, Husted S, James SK, Kervinen K, Kolh P, Kristensen SD, Lancellotti P, Maggioni AP, Piepoli MF, Pries AR, Romeo F, Ryden L, Simoons ML, Sirnes PA, Steg PG, Timmis A, Wijns W, Windecker S, Yildirir A, Zamorano JL (2013) 2013 ESC guidelines on the management of stable coronary artery disease: the Task Force on the management of stable coronary artery disease of the European Society of Cardiology. Eur Heart J 34(38):2949–3003. https://doi.org/10.1093/eurheartj/eht296

3. Reisner SA, Lysyansky P, Agmon Y, Mutlak D, Lessick J, Friedman Z (2004) Global longitudinal strain: a novel index of left ventricular systolic function. J Am Soc Echocardiogr 17(6):630–633. https://doi.org/10.1016/j.echo.2004.02.011
4. Leitman M, Lysyansky P, Sidenko S, Shir V, Peleg E, Binenbaum M, Kaluski E, Krakover R, Vered Z (2004) Two-dimensional strain-a novel software for real-time quantitative echocardiographic assessment of myocardial function. J Am Soc Echocardiogr 17(10):1021–1029. https://doi.org/10.1016/j.echo.2004.06.019
5. Choi JO, Cho SW, Song YB, Cho SJ, Song BG, Lee SC, Park SW (2009) Longitudinal 2D strain at rest predicts the presence of left main and three vessel coronary artery disease in patients without regional wall motion abnormality. Eur J Echocardiogr 10(5):695–701. https://doi.org/10.1093/ejechocard/jep041
6. Biering-Sorensen T, Hoffmann S, Mogelvang R, Zeeberg Iversen A, Galatius S, Fritz-Hansen T, Bech J, Jensen JS (2014) Myocardial strain analysis by 2-dimensional speckle tracking echocardiography improves diagnostics of coronary artery stenosis in stable angina pectoris. Circ Cardiovasc Imaging 7(1):58–65. https://doi.org/10.1161/CIRCIMAGING.113.000989
7. Montgomery DE, Puthumana JJ, Fox JM, Ogunyankin KO (2012) Global longitudinal strain aids the detection of non-obstructive coronary artery disease in the resting echocardiogram. Eur Heart J Cardiovasc Imaging 13(7):579–587. https://doi.org/10.1093/ejechocard/jer282
8. Eek C, Grenne B, Brunvand H, Aakhus S, Endresen K, Smiseth OA, Edvardsen T, Skulstad H (2010) Strain echocardiography predicts acute coronary occlusion in patients with non-ST-segment elevation acute coronary syndrome. Eur J Echocardiogr 11(6):501–508. https://doi.org/10.1093/ejechocard/jeq008
9. Adamu U, Schmitz F, Becker M, Kelm M, Hoffmann R (2009) Advanced speckle tracking echocardiography allowing a three-myocardial layer-specific analysis of deformation parameters. European J Echocardiogr 10(2):303–308. https://doi.org/10.1093/ejechocard/jen238
10. Kim SA, Park SM, Kim MN, Shim WJ (2016) Assessment of left ventricular function by layer-specific strain and its relationship to structural remodelling in patients with hypertension. Can J Cardiol 32(2):211–216. https://doi.org/10.1016/j.cjca.2015.04.025
11. Sarvari SI, Haugaa KH, Zahid W, Bendz B, Aakhus S, Aaberge L, Edvardsen T (2013) Layer-specific quantification of myocardial deformation by strain echocardiography may reveal significant CAD in patients with non-ST-segment elevation acute coronary syndrome. JACC Cardiovasc Imaging 6(5):535–544. https://doi.org/10.1016/j.jcmg.2013.01.009
12. Zhang L, Wu WC, Ma H, Wang H (2016) Usefulness of layer-specific strain for identifying complex CAD and predicting the severity of coronary lesions in patients with non-ST-segment elevation acute coronary syndrome: Compared with Syntax score. Int J Cardiol 223:1045–1052. https://doi.org/10.1016/j.ijcard.2016.08.277
13. Qaseem A, Fihn SD, Williams S, Dallas P, Owens DK, Shekelle P, Clinical Guidelines Committee of the American College of P (2012) Diagnosis of stable ischemic heart disease: summary of a clinical practice guideline from the American College of Physicians/American College of Cardiology Foundation/American Heart Association/American Association for Thoracic Surgery/Preventive Cardiovascular Nurses Association/Society of Thoracic Surgeons. Ann Intern Med 157(10):729–734. https://doi.org/10.7326/0003-4819-157-10-201211200-00010
14. Norum IB, Ruddox V, Edvardsen T, Otterstad JE (2015) Diagnostic accuracy of left ventricular longitudinal function by speckle tracking echocardiography to predict significant coronary artery stenosis. A systematic review. BMC Med Imaging 15:25. https://doi.org/10.1186/s12880-015-0067-y
15. Fox K, Garcia MA, Ardissino D, Buszman P, Camici PG, Crea F, Daly C, De Backer G, Hjemdahl P, Lopez-Sendon J, Marco J, Morais J, Pepper J, Sechtem U, Simoons M, Thygesen K, Priori SG, Blanc JJ, Budaj A, Camm J, Dean V, Deckers J, Dickstein K, Lekakis J, McGregor K, Metra M, Morais J, Osterspey A, Tamargo J, Zamorano JL, Task Force on the Management of Stable Angina Pectoris of the European Society of C, Guidelines ESCCfP (2006) Guidelines on the management of stable angina pectoris: executive summary: the Task Force on the Management of Stable Angina Pectoris of the European Society of Cardiology. Eur Heart J 27(11):1341–1381. https://doi.org/10.1093/eurheartj/ehl001
16. Patel MR, Peterson ED, Dai D, Brennan JM, Redberg RF, Anderson HV, Brindis RG, Douglas PS (2010) Low diagnostic yield of elective coronary angiography. N Engl J Med 362(10):886–895. https://doi.org/10.1056/NEJMoa0907272
17. Hoffmann S, Jensen JS, Iversen AZ, Sogaard P, Galatius S, Olsen NT, Bech J, Fritz-Hansen T, Biering-Sorensen T, Badskjaer J, Pietersen A, Mogelvang R (2012) Tissue Doppler echocardiography improves the diagnosis of coronary artery stenosis in stable angina pectoris. Eur Heart J Cardiovasc Imaging 13(9):724–729. https://doi.org/10.1093/ehjci/jes001
18. Lang RM, Bierig M, Devereux RB, Flachskampf FA, Foster E, Pellikka PA, Picard MH, Roman MJ, Seward J, Shanewise JS, Solomon SD, Spencer KT, Sutton MS, Stewart WJ, Chamber Quantification Writing G, American Society of Echocardiography's G, Standards C, European Association of E (2005) Recommendations for chamber quantification: a report from the American Society of Echocardiography's Guidelines and Standards Committee and the Chamber Quantification Writing Group, developed in conjunction with the European Association of Echocardiography, a branch of the European Society of Cardiology. J Am Soc Echocardiogr 18(12):1440–1463. https://doi.org/10.1016/j.echo.2005.10.005
19. Negishi K, Negishi T, Kurosawa K, Hristova K, Popescu BA, Vinereanu D, Yuda S, Marwick TH (2015) Practical guidance in echocardiographic assessment of global longitudinal strain. JACC Cardiovasc Imaging 8(4):489–492. https://doi.org/10.1016/j.jcmg.2014.06.013
20. Saunamäki KEK, Krusell L, Mickley H, Rokkedal J, Schnohr P (2002) Guidelines for clinical exercise testing in relation to ischemic heart disease (composed by a committee appointed by the Danish Society of Cardiology). https://wwwcardiodk/component/docman/doc_download/124-klinisk-arbejdstest?Itemid=118. Accessed 19 Sept 2016
21. Mark DB, Hlatky MA, Harrell FE Jr, Lee KL, Califf RM, Pryor DB (1987) Exercise treadmill score for predicting prognosis in coronary artery disease. Ann Intern Med 106(6):793–800

22. Ma C (2002) From the help desk: comparing areas under receiver operating characteristic curves from two or more probit or logit models. Stata J 2:301–313
23. Shimoni S, Gendelman G, Ayzenberg O, Smirin N, Lysyansky P, Edri O, Deutsch L, Caspi A, Friedman Z (2011) Differential effects of coronary artery stenosis on myocardial function: the value of myocardial strain analysis for the detection of coronary artery disease. J Am Soc Echocardiogr 24(7):748–757. https://doi.org/10.1016/j.echo.2011.03.007
24. Nucifora G, Schuijf JD, Delgado V, Bertini M, Scholte AJ, Ng AC, van Werkhoven JM, Jukema JW, Holman ER, van der Wall EE, Bax JJ (2010) Incremental value of subclinical left ventricular systolic dysfunction for the identification of patients with obstructive coronary artery disease. Am Heart J 159(1):148–157. https://doi.org/10.1016/j.ahj.2009.10.030
25. Segall GM, Atwood JE, Botvinick EH, Dae MW, Lucas JR (1995) Variability of normal coronary anatomy: implications for the interpretation of thallium-SPECT myocardial perfusion images in single-vessel disease. J Nucl 36(6):944–951
26. Gjesdal O, Hopp E, Vartdal T, Lunde K, Helle-Valle T, Aakhus S, Smith HJ, Ihlen H, Edvardsen T (2007) Global longitudinal strain measured by two-dimensional speckle tracking echocardiography is closely related to myocardial infarct size in chronic ischaemic heart disease. Clin Sci (London, England: 1979) 113(6):287–296. https://doi.org/10.1042/cs20070066
27. Geyer H, Caracciolo G, Abe H, Wilansky S, Carerj S, Gentile F, Nesser HJ, Khandheria B, Narula J, Sengupta PP (2010) Assessment of myocardial mechanics using speckle tracking echocardiography: fundamentals and clinical applications. J Am Soc Echocardiogr 23(4):351–369; (**quiz 453–355**). https://doi.org/10.1016/j.echo.2010.02.015
28. Biering-Sorensen T, Solomon SD (2015) Assessing contractile function when ejection fraction is normal: a case for strain imaging. Circ Cardiovasc Imaging 8(11):e004181. https://doi.org/10.1161/CIRCIMAGING.115.004181
29. Leitman M, Lysiansky M, Lysyansky P, Friedman Z, Tyomkin V, Fuchs T, Adam D, Krakover R, Vered Z (2010) Circumferential and longitudinal strain in 3 myocardial layers in normal subjects and in patients with regional left ventricular dysfunction. J Am Soc Echocardiogr 23(1):64–70. https://doi.org/10.1016/j.echo.2009.10.004
30. Shi J, Pan C, Kong D, Cheng L, Shu X (2016) Left ventricular longitudinal and circumferential layer-specific myocardial strains and their determinants in healthy subjects. Echocardiography 33(4):510–518. https://doi.org/10.1111/echo.13132
31. Myers JH, Stirling MC, Choy M, Buda AJ, Gallagher KP (1986) Direct measurement of inner and outer wall thickening dynamics with epicardial echocardiography. Circulation 74(1):164–172
32. Sabbah HN, Marzilli M, Stein PD (1981) The relative role of subendocardium and subepicardium in left ventricular mechanics. Am J Physiol 240(6):H920–926
33. Hagemann CE, Hoffmann S, Olsen FJ, Jorgensen PG, Fritz-Hansen T, Jensen JS, Biering-Sorensen T (2018) Layer-specific global longitudinal strain reveals impaired cardiac function in patients with reversible ischemia. Echocardiography. https://doi.org/10.1111/echo.13830
34. Skaarup KG, Iversen A, Jørgensen PG, Olsen FJ, Grove GL, Jensen JS, Biering-Sørensen T (2018) Association between layer-specific global longitudinal strain and adverse outcomes following acute coronary syndrome. Eur Heart J. https://doi.org/10.1093/ehjci/jey004
35. Camici PG, Prasad SK, Rimoldi OE (2008) Stunning, hibernation, and assessment of myocardial viability. Circulation 117(1):103–114. https://doi.org/10.1161/CIRCULATIONAHA.107.702993
36. Gjesdal O, Helle-Valle T, Hopp E, Lunde K, Vartdal T, Aakhus S, Smith HJ, Ihlen H, Edvardsen T (2008) Noninvasive separation of large, medium, and small myocardial infarcts in survivors of reperfused ST-elevation myocardial infarction: a comprehensive tissue Doppler and speckle-tracking echocardiography study. Circ Cardiovasc Imaging 1(3):189–196. https://doi.org/10.1161/circimaging.108.784900