



# Investigation of the Possible Risk Factors Associated with Subclinical Left Ventricular Dysfunction Assessed by 2D Speckle Tracking Echocardiography in Patients with Type 2 Diabetes Mellitus and Normal Ejection Fraction

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## ABSTRACT

**Background:** Subclinical left ventricular dysfunction is an important predictor of cardiovascular death, detected via the global longitudinal strain (GLS) echocardiographic parameter. While T2DM is associated with a worse GLS, it is not cost-effective to perform 2D speckle tracking echocardiography for all asymptomatic T2DM patients to screen for early signs of subclinical left ventricular dysfunction.

**Objectives:** We aimed to investigate the association of possible risk factors with subclinical left ventricular dysfunction assessed by 2D speckle tracking echocardiography in T2DM patients with normal ejection fraction, excluding patients with comorbidities associated with left ventricular dysfunction such as hypertension or any cardiovascular disease.

**Methods:** This cross-sectional study involved patients aged >18 with type 2 diabetes mellitus (T2DM) and EF ≥ 50% referred to Shohada-e-Tajrish Hospital, Tehran, Iran, from May 2, 2023, to June 21, 2023. Patients with any history or signs and symptoms of heart disease and hypertension were excluded. The correlation of GLS with age, BMI, duration of diabetes, systolic (SBP) and diastolic blood pressure (DBP), and laboratory parameters was assessed using Pearson's correlation test, with P < 0.05 denoting significance.

**Results:** In this cross-sectional study, 118 patients consisting of 70 women (59.32%) and 48 men (40.68%) with T2DM were enrolled. The mean age of the participants was 49.61 ± 5.10 years. The mean GLS was -16.71 ± 2.14%. The GLS correlated positively with BMI (P = 0.038 and r = 0.197), SBP (P = 0.003 and r = 0.268), DBP (P = 0.023 and r = 0.209), homocysteine (P = 0.001 and r = 0.310), HbA1C (P = 0.046 and r = 0.184), LDL (P = 0.034 and r = 0.203), and TG (P < 0.001 and r = 0.375), and negatively with GFR (P < 0.001 and r = -0.363).

**Conclusions:** BMI, SBP, DBP, homocysteine, HbA1C, LDL, TG, and GFR correlated with subclinical left ventricular dysfunction assessed by GLS in patients with T2DM; these markers may offer value in selecting T2DM patients for cost-effective 2D speckle tracking echocardiography screening of subclinical left ventricular dysfunction.

## 1. Background

Type 2 diabetes mellitus (T2DM) constitutes one of the most prominent public health concerns due to its rising trend

of prevalence and mortality (1). Along with the production of more effective diabetes medications, several studies have been conducted on other medications and even supplements to help control blood glucose levels (2, 3). T2DM has numerous well-known complications, with cardiovascular diseases representing one of the leading causes of death globally (4). Among the wide range of cardiovascular

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diseases, heart failure is one of the major causes of morbidity and mortality (4). T2DM is an independent risk factor for heart failure (5); “diabetic cardiomyopathy” is defined as a condition in which heart failure occurs in patients with diabetes without the presence of any coronary artery disease, valvular disease, or hypertension (5). This medical condition is explained by glucose and lipid toxicity in the setting of insulin resistance and metabolic disorders (5).

Heart failure with preserved or reduced ejection fraction is associated with diabetes (6), beginning with left ventricular diastolic dysfunction and progressing to systolic dysfunction with reduced ejection fraction later in the course of diabetes (5). Therefore, the best strategy for preventing overt heart failure or delaying its progression to an advanced stage is screening for heart failure in stage A (people at risk) or stage B (people without any symptoms having evidence on echocardiography or laboratory biomarkers), which may lead to the early prescription of approved medications to prevent heart failure progression including sodium glucose co-transporter 2 (SGLT2) inhibitors,  $\beta$ -blockers, and renin angiotensin aldosterone system (RAAS) inhibitors (7).

The more available strategies, including traditional transthoracic echocardiography or laboratory biomarkers (including troponin or natriuretic peptides), are suboptimal in detecting asymptomatic heart failure patients (7). However, in comparison to the tissue Doppler method, speckle tracking echocardiography assesses the myocardial deformation by global longitudinal strain (GLS) measurement and detects left ventricular systolic dysfunction in patients with normal left ventricular ejection fraction (LVEF); it is easy to calculate and more accurate as it is independent of the angle—unlike the tissue Doppler method (8, 9). Therefore, GLS assessment has become a clinically feasible alternative to LVEF; it is more sensitive to left ventricular dysfunction than LVEF and provides additional prognostic information (10). As a result, it has been used in different clinical settings (11, 12), including the assessment of T2DM patients (13). T2DM is associated with a worse GLS value (14). Early detection of subclinical LV dysfunction by 2D speckle tracking has important clinical and prognostic significance in patients with T2DM (15). However, performing 2D speckle-tracking echocardiography for all asymptomatic patients with T2DM is not cost-effective (7).

## 2. Objectives

We aimed to investigate the association of the possible risk factors with subclinical LV dysfunction assessed by 2D speckle tracking echocardiography in T2DM patients with normal EF, which may help to find the most eligible T2DM patients for subclinical LV dysfunction assessment using this modality.

## 3. Methods

### 3.1. Study Design and Population

This cross-sectional study involved patients aged > 18 with T2DM and systolic LVEF  $\geq$  50% referred to Shohadae-Tajrish Hospital, Tehran, Iran, from May 2, 2023, to June 21, 2023. Patients with any history or signs and symptoms of

heart diseases, including systolic or diastolic heart failure, diastolic dysfunction grade  $\geq$  2, LV hypertrophy (more than mild), coronary artery diseases, arrhythmia, congenital or structural heart disease, considerable valvular heart disease (moderate or severe), regional wall motion abnormality in echocardiography, and hypertension were excluded.

The study was approved by the Shahid Beheshti University of Medical Sciences Ethics Committee (IR.SBMU.MSP.REC.1402.38), and written informed consent was obtained from all patients before enrollment.

### 3.2. Clinical and Laboratory Measurements

The patient’s demographic data, including age, sex, and duration of diabetes (years) since diagnosis, were documented. Body mass index (BMI) was calculated as the ratio of weight (kg) to height squared ( $m^2$ ). Systolic and diastolic blood pressures (SBP and DBP, respectively) were measured by a manual sphygmomanometer in a seated position following 15 minutes of rest and smoke/caffeine avoidance.

All participants underwent transthoracic echocardiography using 2D speckle tracking by an expert cardiologist, and global longitudinal strain (GLS) was calculated automatically from the mean of the global peak systolic strain obtained from three apical long-axis views (16). After 12 hours of fasting, a venous blood sample was collected from the antecubital vein to measure the serum level of vitamin D (vit D), homocysteine, low-density lipoprotein (LDL), high-density lipoprotein (HDL), triglyceride (TG), C-reactive protein (CRP), calcium (Ca), phosphorus (P), glycated hemoglobin (HbA1C), thyroid stimulating hormone (TSH), creatinine (Cr), and estimation of glomerular filtration rate (GFR).

### 3.3. Definition of Terms

In this study, T2DM was defined as fasting plasma glucose (FPG)  $\geq$  126 mg/dL or 2 hours post 75 g glucose intake (2-h PG)  $\geq$  200 mg/dL during oral glucose tolerance test (OGTT) or HbA1C  $\geq$  6.5% or random plasma glucose  $\geq$  200 mg/dL in a patient with classic symptoms of hyperglycemia or taking any glucose-lowering medications (17). Hypertension was defined as SBP  $\geq$  140 mmHg, DBP  $\geq$  90 mmHg, or taking blood pressure-lowering medications (18).

Subclinical LV dysfunction was considered abnormal GLS in the presence of LVEF  $\geq$  50% and no clinical symptoms of heart failure. GLS is a myocardial deformation analysis that mainly reflects the performance of sub-endocardial longitudinal fibers, which are more susceptible to ischemic injury and wall stress. The mean normal value of GLS is considered -20% (16), previously categorized as follows: -15% to -25% normal, -15% to -12.5% mildly reduced, -12.5% to -8.1% moderately reduced, and < -8 % severely reduced (19). As a GLS amount worse than -12.5% is mostly associated with considerable decreases in EF and the population of our study consisted of patients with EF  $\geq$  50%, we considered -20% as the normal value as reported previously (16) and categorized the GLS into three groups of worse than -15 %, -15% to -20%, and better than -20%.

The risk factors considered to evaluate their possible association with subclinical LV dysfunction were age, BMI,

duration of diabetes since diagnosis, SBP and DBP, and laboratory tests including vit D, homocysteine, LDL, HDL, TG, CRP, Ca, P, HbA1C, TSH, Cr, and GFR.

#### 3.4. Statistical Analysis

Data analysis was done using SPSS statistical software version 22, and the mean and standard deviation were used to report quantitative variables. Frequency and percentage were used to report qualitative variables. The association of subclinical left ventricular dysfunction (measured by GLS) with possible risk factors, including age, BMI, duration of diabetes, SBP, DBP, Cr, vitamin D, homocysteine, LDL, HDL, TG, CRP, Ca, P, HbA1C, GFR, and TSH, was assessed using Pearson's correlation test. P-values < 0.05 were considered statistically significant.

#### 4. Results

This cross-sectional study enrolled 118 patients with T2DM, consisting of 70 women (59.32%) and 48 men (40.68%). The mean age of the participants was  $49.61 \pm 5.10$  years, and the age range was 36–89 years. The demographic, clinical, and laboratory findings are described in Table 1.

The mean GLS of the patients was  $-16.71 \pm 2.14\%$ . According to Table 2, GLS was worse than  $-15\%$  in 48 (40.67%) patients.

As mentioned in Tables 3 and 4, the results of Pearson's correlation test showed a significant positive correlation of GLS's numerical value with BMI ( $P = 0.038$  and  $r = 0.197$ ), SBP ( $P = 0.003$  and  $r = 0.268$ ), DBP ( $P = 0.023$  and  $r = 0.209$ ), homocysteine ( $P = 0.001$  and  $r = 0.310$ ), HbA1C ( $P = 0.046$  and  $r = 0.184$ ), LDL ( $P = 0.034$  and  $r = 0.203$ ), and TG ( $P < 0.001$  and  $r = 0.375$ ). That is, higher levels of the mentioned parameters were associated with worse GLS amount and subclinical LV dysfunction.

**Table 1.** Description of Demographic, Clinical, and Laboratory Findings of the Participants

	Mean $\pm$ SD
Age (Years)	49.61 $\pm$ 5.10
BMI (kg/m <sup>2</sup> )	26.07 $\pm$ 5.24
Duration of diabetes (years)	8.69 $\pm$ 4.24
SBP (mmHg)	121.75 $\pm$ 12.29
DBP (mmHg)	74.09 $\pm$ 6.61
Cr (mg/dL)	1.39 $\pm$ 0.29
Vit D (ng/mL)	20.17 $\pm$ 4.81
Homocysteine (mcmol/L)	14.56 $\pm$ 7.35
LDL (mg/dL)	137.49 $\pm$ 27.78
HDL (mg/dL)	42.58 $\pm$ 10.56
TG (mg/dL)	204.31 $\pm$ 83.74
CRP (mg/L)	5.33 $\pm$ 2.88
Ca (mg/dL)	8.91 $\pm$ 1.44
P (mg/dL)	2.79 $\pm$ 0.90
HbA1C (%)	7.53 $\pm$ 0.73
GFR (mL/min/1.73m <sup>2</sup> )	59.98 $\pm$ 25.60
TSH (mIU/L)	4.68 $\pm$ 2.04

Abbreviations: BMI; body mass index, SBP; systolic blood pressure, DBP; diastolic blood pressure, Cr; creatinine, Vit D; vitamin D, LDL; low-density lipoprotein, HDL; high-density lipoprotein, TG; triglyceride, CRP; C-reactive protein, Ca; calcium, P; phosphorus, HbA1C; glycated hemoglobin, GFR; glomerular filtration rate, TSH; thyroid stimulating hormone.

**Table 2.** Distribution of Global Longitudinal Strain Categories among Patients

	Frequency (Percentage)
GLS worse than $-15\%$	48 (40.67%)
GLS $-15\%$ to $-20\%$	29 (24.57%)
GLS better than $-20\%$	41 (34.76%)

Abbreviations: GLS: global longitudinal strain

**Table 3.** Evaluation of the Correlation of Global Longitudinal Strain with Demographic Features and Blood Pressure Values

Parameter	r	P value
Age (years)	0.134	0.147
BMI (kg/m <sup>2</sup> )	0.197	0.038
Duration of diabetes (years)	0.031	0.736
SBP (mmHg)	0.268	0.003
DBP (mmHg)	0.209	0.023

Abbreviations: BMI; body mass index, SBP; systolic blood pressure, DBP; diastolic blood pressure

**Table 4.** Evaluation of the Correlation of Global Longitudinal Strain with Laboratory Findings

Laboratory Parameter	r	P value
Cr (mg/dL)	0.140	0.130
Vitamin D (ng/mL)	-0.105	0.257
Homocysteine (mcmol/L)	0.310	0.001
LDL (mg/dL)	0.203	0.034
HDL (mg/dL)	-0.136	0.143
TG (mg/dL)	0.375	< 0.001
CRP (mg/L)	0.023	0.877
Ca (mg/dL)	0.101	0.067
P (mg/dL)	-0.113	0.223
HbA1C (%)	0.184	0.046
GFR (mL/min/1.73m <sup>2</sup> )	-0.363	< 0.001
TSH (mIU/L)	0.056	0.602

Abbreviations: Cr; creatinine, Vit D; vitamin D, LDL; low-density lipoprotein, HDL; high-density lipoprotein, TG; triglyceride, CRP; C-reactive protein, Ca; calcium, P; phosphorus, HbA1C; glycated hemoglobin, GFR; glomerular filtration rate, TSH; thyroid stimulating hormone.

However, a significant negative correlation was seen between GLS and GFR ( $P < 0.001$  and  $r = -0.363$ ), meaning that decreased GFR was associated with a worse GLS amount and subclinical LV dysfunction.

## 5. Discussion

Our study shows that in patients with T2DM and normal EF, clinical findings including higher BMI, SBP, and DPB, are associated with a higher GLS, indicating worse subclinical LV systolic function. These readily available parameters are valuable as they can be measured during each medical consult. In terms of diabetes-related parameters, the HbA1C level showed a positive correlation with the numerical amount of GLS, also meaning worse subclinical LV systolic function with poorly controlled diabetes. However, the duration of diabetes did not show a significant correlation, which means it may not be a reliable indicator of diabetes onset. Among laboratory parameters, the serum levels of homocysteine, LDL, and TG were positively correlated, and GFR was inversely correlated with GLS, indicating the association of these factors with subclinical LV systolic dysfunction in patients with T2DM. The mentioned parameters are also easily accessible, and our results show that they may be considered to help find the most eligible T2DM patients with normal ejection fraction being cost-effective for subclinical LV dysfunction assessment using 2D speckle tracking echocardiography.

Several studies have been conducted on subclinical LV dysfunction. A significantly lower LV systolic function (as measured by tissue Doppler echocardiography) has been reported in patients with T2DM, which correlated inversely with DBP, LDL, age, and HbA1C (20). GLS, more sensitive for detecting subclinical systolic dysfunction, is significantly impaired in asymptomatic T2DM patients with normal EF (21). Moreover, subclinical LV dysfunction assessed by GLS was independently associated with adverse outcomes including all-cause mortality and hospitalization during ten years of follow-up in asymptomatic patients with T2DM and normal EF (22). Although the study of Nakai et al. showed a correlation between GLS and duration of diabetes (23), this correlation was not significant in our study.

In previous studies, a positive linear correlation was reported between BMI and subclinical LV dysfunction assessed by 3D speckle-tracking echocardiography, not only in obese patients but also in overweight people (24). Our study replicated this correlation in patients with T2DM.

Moreover, a significant correlation between systolic and diastolic blood pressure with GLS was shown in our study, which may propose that higher blood pressure and diabetes are both involved in accelerating subclinical LV dysfunction. Similarly, Holland et al. showed a significant correlation of GLS with SBP (22). Ballo et al. reported that the co-existence of diabetes and hypertension led to greater impairment of LV longitudinal systolic function (25). However, patients with hypertension were excluded from our study, and therefore, our results show that even in normal systolic and diastolic blood pressure, the correlation between SBP or DBP and GLS remains significant. Apart from patients with T2DM, the results of a study on antihypertensive medication-naïve individuals showed that

systolic hypertension and isolated diastolic hypertension were associated with a risk of abnormal left ventricular GLS in both sexes, while this association between blood pressure and GLS remained significant only among women with elevated blood pressure (26).

Homocysteine is another factor that showed a significant correlation with GLS in our study, proposing that an elevated homocysteine level may confer an additive risk of developing LV dysfunction in patients with T2DM. Although higher homocysteine levels have been proposed for the prediction of congestive heart failure development in adults without a history of previous myocardial infarction (27), there is a lack of studies on the association between serum homocysteine level and subclinical LV dysfunction, especially assessed by 2D speckle tracking echocardiography. In a study on patients with diabetes, the serum homocysteine level was associated with a higher risk of cardiovascular and all-cause mortality (28). Moreover, homocysteine has been proposed as a promising biomarker for vascular complications in the setting of diabetes (29). While an inverse correlation between serum homocysteine level and LVEF was reported in patients with diabetes (30), our results propose that homocysteine can be useful in subclinical reduced LV systolic function detection. Some mechanisms have been proposed for the role of homocysteine in diabetes and its cardiovascular complications, including exacerbation of endothelial dysfunction and insulin resistance, possibly associated with its oxidative stress property (31-33).

Another finding of the current study was the significant correlation between GLS and GFR in T2DM patients, previously reported in patients with chronic kidney disease (CKD)(34). This may be due to the progressive myocardial structure change mediated by uremia, biochemical disorders secondary to CKD, and pressure or volume overload (35). The mentioned study also reported an association between phosphate level and GLS (34), which was not replicated in our study.

In terms of lipid profile, a significant correlation between GLS and the levels of LDL and TG was reported in patients with asymptomatic carotid stenosis (36). In our study on patients with T2DM, LDL and TG showed a significant correlation with GLS and subclinical LV systolic dysfunction. This may be explained by their lipotoxic effects on the myocardium (5). The fatty acids of the cardiomyocytes increase in response to free fatty acids of the serum; this excess fatty acids accumulation in cardiomyocytes leads to an increase of toxic lipid intermediates, including diacylglycerol. Diacylglycerol aggravates insulin resistance and mediates oxidative stress, resulting in cardiac dysfunction (5). Moreover, insulin resistance has been proposed as a cause of impaired metabolic flexibility of the heart. A decrease in glucose uptake from cardiomyocytes may lead to a change in the substrate towards an increase in fatty acid oxidation, which causes lipotoxicity and, as a result, a decrease in cardiac efficiency, as well as an increase in myocardial oxygen consumption and reactive oxygen species production. In addition, hyperglycemia itself may induce advanced glycosylation end products that stimulate collagen expression and accumulation, leading to myocardial fibrosis with increased myocardial

stiffness and decreased cardiac compliance (37-39). High glucose levels also cause calcium misuse and mitochondrial dysfunction, which may play important roles in the early stages of diabetic cardiomyopathy (40).

One of the present study's strengths is being the first to investigate possible risk factors affecting GLS in T2DM patients without hypertension or any cardiovascular disease. Also, to our knowledge, we are the first to assess the correlation of SBP and DPB with GLS in normotensive patients with T2DM. This can itself be evidence of the possible effect of higher systolic and diastolic blood pressure (even within the normal range) in causing diabetic cardiomyopathy. As a result, more studies to investigate this association and the possible need to reduce the threshold of high blood pressure definition in patients with T2DM having risk factors of developing diabetic cardiomyopathy are proposed. One limitation of this study is that patients were from a single center; conducting multicenter studies with larger sample sizes will increase the accuracy of the findings. Moreover, this was a cross-sectional study, and performing a cohort study to confirm our findings and the factors affecting the development of subclinical left ventricular dysfunction over time is recommended.

### 5.1. Conclusion

The results of the present study show a significant correlation between GLS and BMI, SBP, DBP, homocysteine, HbA1C, LDL, TG, and GFR in patients with T2DM. These clinical and laboratory parameters are readily available and might help identify patients who are at a greater risk of developing subclinical LV dysfunction among those with T2DM and normal EF; therefore, to achieve greater cost-efficiency, it is worth considering prioritizing subclinical LV dysfunction assessment using 2D speckle tracking echocardiography for people in this subgroup rather than all patients with T2DM.

### 5.2. Ethical Approval

This study was approved by the Shahid Beheshti University of Medical Sciences Ethics Committee (IR.SBMU.MSP.REC.1402.38).

### 5.3. Informed Consent

Written informed consent was obtained from all participants.

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### Authors' Contribution

M.P. conceived and designed the evaluation. F.B., R.B., and K.K.T. participated in designing the evaluation, S.G.F., H.R.S., and S.M.M. collected the clinical data and performed parts of the statistical analysis. K.K.T. drafted and revised the manuscript. All authors read and approved the final manuscript.

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### References

1. Sang S, Chu C, Zhang T, Chen H, Yang X. The global burden of disease attributable to ambient fine particulate matter in 204 countries and territories, 1990–2019: A systematic analysis of the Global Burden of Disease Study 2019. *Ecotoxicology and environmental safety*. 2022;**238**:113588.
2. Mosalanejad S, Shahrzad MK, Pishgahi M, Toudeshki KK, Ghanefard S, Ebadi SA. Effect of Vitamin D Administration on Glycemic Control in Patients with Type II Diabetes Mellitus and Vitamin D Deficiency. *Men's Health Journal*. 2021;**5**(1):e29-e.
3. Ebadi SA, Pishgahi M, Toudeshki KK, Forootan M, Ghanefard S, Shahrzad MK. Effect of Proton Pump Inhibitor Administration on Glycemic Parameters in Patients with Type 2 Diabetes Mellitus. *Men's Health Journal*. 2021;**5**(1):e28-e.
4. Gaidai O, Cao Y, Loginov S. Global cardiovascular diseases death rate prediction. *Current Problems in Cardiology*. 2023:101622.
5. Nakamura K, Miyoshi T, Yoshida M, Akagi S, Saito Y, Ejiri K, et al. Pathophysiology and treatment of diabetic cardiomyopathy and heart failure in patients with diabetes mellitus. *International journal of molecular sciences*. 2022;**23**(7):3587.
6. Redfield MM, Jacobsen SJ, Burnett JC, Mahoney DW, Bailey KR, Rodeheffer RJ. Burden of systolic and diastolic ventricular dysfunction in the community: appreciating the scope of the heart failure epidemic. *Jama*. 2003;**289**(2):194-202.
7. Pop-Busui R, Januzzi JL, Brummer D, Butalia S, Green JB, Horton WB, et al. Heart failure: an underappreciated complication of diabetes. A consensus report of the American Diabetes Association. *Diabetes care*. 2022;**45**(7):1670-90.
8. Bshiebish HAH, Al-Musawi AH, Khudeir SA. Role of global longitudinal strain in assessment of left ventricular systolic function in patients with heart failure with preserved ejection fraction. *J Saudi Heart Assoc*. 2019;**31**(2):100-5.
9. Hill JC, Palma RA. Doppler tissue imaging for the assessment of left ventricular diastolic function: a systematic approach for the sonographer. *Journal of the American Society of Echocardiography*. 2005;**18**(1):80-8.
10. Potter E, Marwick TH. Assessment of left ventricular function by echocardiography: the case for routinely adding global longitudinal strain to ejection fraction. *JACC: Cardiovascular Imaging*. 2018;**11**(2 Part 1):260-74.
11. Pishgahi M, Mahmoudi F, Toudeshki KK. Acute Effects of Smoking on Diastolic and Systolic Cardiac Function of Healthy Smokers according to 2D Speckle Tracking Echocardiography. *International Cardiovascular Research Journal*. 2022(In Press).
12. Abou R, van der Bijl P, Bax JJ, Delgado V. Global longitudinal strain: clinical use and prognostic implications in contemporary practice. *Heart (British Cardiac Society)*. 2020;**106**(18):1438-44.
13. Pishgahi M, Bozorgmehr R, Farahani F, Soltani F, Haghshenas M. The prevalence of Sub-Clinical Left Ventricle Dysfunction and Relevant Intensifying Factors in Type 2 DM Patients using Strain Echocardiography Indices. *Journal of Advanced Pharmacy Education & Research | Oct-Dec*. 2018;**8**(S2):117.
14. Soufi Taleb Bendiab N, Ouabdesselam S, Henaoui L, Lopez-Sublet M, Monsuez J-J, Benkhedda S. Impact of Diabetes on Cardiac Function in Patients with High Blood Pressure. *International Journal of Environmental Research and Public Health*. 2021;**18**(12):6553.
15. Stevanovic A, Dekleva M. The importance of subclinical left ventricular dysfunction and blood pressure pattern in asymptomatic type-2 diabetic patients: The diagnostic and prognostic significance of Tissue Doppler parameters, left ventricular global longitudinal strain, and nighttime blood pressure during sleep. *Journal of diabetes and its complications*. 2018;**32**(1):41-7.
16. Joseph G, Zaremba T, Johansen MB, Ekeloef S, Heiberg E, Engblom H, et al. Echocardiographic global longitudinal strain is associated with infarct size assessed by cardiac magnetic resonance in acute myocardial infarction. *Echo research and practice*. 2019;**6**(4):81-9.

17. ElSayed NA, Aleppo G, Aroda VR, Bannuru RR, Brown FM, Bruemmer D, *et al.* 2. Classification and Diagnosis of Diabetes: Standards of Care in Diabetes—2023. *Diabetes care.* 2023;**46**(Supplement\_1):S19-S40.
18. Chakraborty DS, Lahiry S, Choudhury S. Hypertension Clinical Practice Guidelines (ISH, 2020): What Is New? *Medical Principles and Practice.* 2021;**30**(6):579-84.
19. Vijayaraghavan G, Sivasankaran S. Global longitudinal strain: A practical step-by-step approach to longitudinal strain imaging. *Journal of The Indian Academy of Echocardiography & Cardiovascular Imaging.* 2020;**4**(1):22.
20. Vinereanu D, Nicolaides E, Tweddel AC, MÄDLER CK, Holst B, Boden LE, *et al.* Subclinical left ventricular dysfunction in asymptomatic patients with Type II diabetes mellitus, related to serum lipids and glycated haemoglobin. *Clinical Science.* 2003;**105**(5):591-9.
21. Ng AC, Delgado V, Bertini M, van der Meer RW, Rijzewijk LJ, Shanks M, *et al.* Findings from left ventricular strain and strain rate imaging in asymptomatic patients with type 2 diabetes mellitus. *The American journal of cardiology.* 2009;**104**(10):1398-401.
22. Holland DJ, Marwick TH, Haluska BA, Leano R, Hordern MD, Hare JL, *et al.* Subclinical LV dysfunction and 10-year outcomes in type 2 diabetes mellitus. *Heart (British Cardiac Society).* 2015;**101**(13):1061-6.
23. Nakai H, Takeuchi M, Nishikage T, Lang RM, Otsuji Y. Subclinical left ventricular dysfunction in asymptomatic diabetic patients assessed by two-dimensional speckle tracking echocardiography: correlation with diabetic duration. *European Journal of Echocardiography.* 2009;**10**(8):926-32.
24. Dođduş M, Kılıç S, Vuruşkan E. Evaluation of subclinical left ventricular dysfunction in overweight people with 3D speckle-tracking echocardiography. *Anatolian journal of cardiology.* 2019;**21**(4):180-6.
25. Ballo P, Cameli M, Mondillo S, Giacomini E, Lisi M, Padeletti M, *et al.* Impact of diabetes and hypertension on left ventricular longitudinal systolic function. *Diabetes research and clinical practice.* 2010;**90**(2):209-15.
26. Nakanishi K, Daimon M, Yoshida Y, Ishiwata J, Sawada N, Hirokawa M, *et al.* Blood pressure categorization and subclinical left ventricular dysfunction in antihypertensive medication-naïve subjects. *ESC heart failure.* 2022;**9**(3):1766-74.
27. Vasan RS, Beiser A, D'Agostino RB, Levy D, Selhub J, Jacques PF, *et al.* Plasma Homocysteine and Risk for Congestive Heart Failure in Adults Without Prior Myocardial Infarction. *Jama.* 2003;**289**(10):1251-7.
28. Lu J, Chen K, Chen W, Liu C, Jiang X, Ma Z, *et al.* Association of Serum Homocysteine with Cardiovascular and All-Cause Mortality in Adults with Diabetes: A Prospective Cohort Study. *Oxidative Medicine and Cellular Longevity.* 2022;**2022**.
29. Soedamah-Muthu S, Chaturvedi N, Teerlink T, Idzior-Walus B, Fuller JH, Stehouwer C, *et al.* Plasma homocysteine and microvascular and macrovascular complications in type 1 diabetes: a cross-sectional nested case-control study. *Journal of internal medicine.* 2005;**258**(5):450-9.
30. Badiou S, Dupuy A-M, Jausse I, Sultan A, Mariano-Goulard D, Cristol J-P, *et al.* Homocysteine as a determinant of left ventricular ejection fraction in patients with diabetes. *Clinical Chemistry and Laboratory Medicine.* 2012;**50**(6):1099-106.
31. Hu Y, Xu Y, Wang G. Homocysteine levels are associated with endothelial function in newly diagnosed type 2 diabetes mellitus patients. *Metabolic syndrome and related disorders.* 2019;**17**(6):323-7.
32. Lindschinger M, Tatzber F, Schimetta W, Schmid I, Lindschinger B, Cvirn G, *et al.* A randomized pilot trial to evaluate the bioavailability of natural versus synthetic vitamin B complexes in healthy humans and their effects on homocysteine, oxidative stress, and antioxidant levels. *Oxidative medicine and cellular longevity.* 2019;**2019**.
33. Muzurović E, Kraljević I, Solak M, Dragnić S, Mikhailidis DP. Homocysteine and diabetes: role in macrovascular and microvascular complications. *Journal of diabetes and its complications.* 2021;**35**(3):107834.
34. Krishnasamy R, Isbel NM, Hawley CM, Pascoe EM, Leano R, Haluska BA, *et al.* The association between left ventricular global longitudinal strain, renal impairment and all-cause mortality. *Nephrology Dialysis Transplantation.* 2014;**29**(6):1218-25.
35. Herzog CA, Asinger RW, Berger AK, Charytan DM, Diez J, Hart RG, *et al.* Cardiovascular disease in chronic kidney disease. A clinical update from Kidney Disease: Improving Global Outcomes (KDIGO). *Kidney international.* 2011;**80**(6):572-86.
36. Hong L, Xing L, Li R, Zhang L, Ma C, An J, *et al.* Subclinical left ventricular dysfunction assessed by two-dimensional speckle tracking echocardiography in asymptomatic patients with carotid stenosis. *The International Journal of Cardiovascular Imaging.* 2019;**35**:2205-12.
37. Paolillo S, Marsico F, Prastaro M, Renga F, Esposito L, De Martino F, *et al.* Diabetic cardiomyopathy: definition, diagnosis, and therapeutic implications. *Heart failure clinics.* 2019;**15**(3):341-7.
38. Fang ZY, Prins JB, Marwick TH. Diabetic cardiomyopathy: evidence, mechanisms, and therapeutic implications. *Endocrine reviews.* 2004;**25**(4):543-67.
39. Liu Q, Wang S, Cai L. Diabetic cardiomyopathy and its mechanisms: role of oxidative stress and damage. *Journal of diabetes investigation.* 2014;**5**(6):623-34.
40. Jaquenod De Giusti C, Palomeque J, Mattiazzi A. Ca<sup>2+</sup> mishandling and mitochondrial dysfunction: a converging road to prediabetic and diabetic cardiomyopathy. *Pflügers Archiv-European Journal of Physiology.* 2022;**474**(1):33-61.