PREDICTION OF SUBCLINICAL LEFT VENTRICULAR SYSTOLIC DYSFUNCTION BY MITRAL ANNULAR PLANE SYSTOLIC EXCURSION IN M-MODE ECHOCARDIOGRAPHY IN CORRELATION TO SPECKLE TRACKING BY 2-DIMENSIONAL ECHOCARDIOGRAPHY AND TISSUE DOPPLER IMAGING IN PATIENTS WITH TYPE 2 DIABETES MELLITUS

By

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ABSTRACT

Background: Diabetes mellitus (DM) is considered as a major risk factor for heart failure with preserved Ejection fraction (EF). Using M-Mode echocardiographic method may be useful in prediction of early LV systolic dysfunction in patient with type II diabetes mellitus by measurement of mitral annular plane systolic excursion (MAPSE) in correlation to global longitudinal strain (GLS) and to tissue Doppler imaging (TDI).

Objective: To emphasize on the usefulness of M-mode echocardiographic methods like the mitral annular plane systolic excursion (MAPSE) correlated to speckle tracking by 2 dimensional (STE) and tissue Doppler imaging in prediction of subclinical LV systolic dysfunction in patients with type 2 diabetes.

Patients and methods: The study included 100 asymptomatic patients with type II DM and 50 healthy subjects as a control group. Echocardiography was performed to all of them All cases were recruited from the outpatient clinic in Maadi military Hospital in Cairo.

Results: The study included 100 asymptomatic patients with type II DM, 65 of them were females, and 50 healthy subjects as a control group, 27 of them were females. MAPSE were significantly lower in DM group [1.2 (1.1 – 1.25) cm] compared to control group [1.4 (1.3 – 1.5) cm] (p<0.001). MAPSE linearly had a significant positive correlation with EF, GLS and Sa wave [r= 0.565, 0.723, 0.595 respectively], (p < 0.01). MAPSE linearly had a significant negative correlation with DM duration and HbA1C Level [r= -0.495, -0.776 respectively], (p < 0.01). Average MAPSE at a cut-off point (≤1.2cm) with good (82%) accuracy, sensitivity= 68% and specificity= 82% (p < 0.01).

Conclusions: MAPSE is a useful M-mode method in prediction of subclinical LV systolic dysfunction in asymptomatic type II diabetic patients, and it has a positive correlation to GLS and Sa wave of TDI. It has a negative correlation with duration of DM and HbA1C level.

Keywords: Subclinical Dysfunction, Diabetes Mellitus, Mitral Annular Plane Systolic Excursion, Speckle Tracking Echocardiography, Tissue Doppler imaging.

INTRODUCTION

Diabetes mellitus (DM) is a major risk factor for heart failure with preserved Ejection fraction (HFPF) this is known as diabetic cardiomyopathy (DCM) *(Cosentino et al., 2019).*

Although diastolic dysfunction considered as the first hallmark of DCM, Studies highlighted that even when diastolic function is normal and LVEF is preserved, there still exists a systolic LV strain alteration assessed through longitudinal systolic LV strain and mitral annular plane Systolic Excursion (MAPSE), leading to the idea that diastolic dysfunction should not be considered the first sign of subclinical diabetic cardiomyopathy *(Ernande et al., 2011).*

Mitral annular plane systolic excursion (MAPSE) is a useful surrogate of left ventricular (LV) longitudinal function and has long been used as a complimentary measure for ejection fraction (EF) *(Hu et al., 2013).*

Two-dimensional speckle tracking echocardiography (2D STE) is a novel technique of cardiac imaging for assessment of cardiac movement based on frame-to-frame tracking of ultrasonic speckles in gray level 2D images it is known as a relatively angle independent technology that can determine global and regional strain *(Mondillo et al., 2011).*

Unlike 2D STE which is vendor, image quality and hemodynamics dependent that need more standardization and experience, MAPSE is, available in every echo machine and, feasible even in a poor image quality. So, MAPSE could be proposed as well-established clinically

useful echocardiographic parameter for the assessment of LV longitudinal function and correlates with global systolic function of the LV compared to gold standard method (GLS) in patients with type II diabetes mellitus and with preserved LVEF *(Hu et al., 2012 and Matos et al., 2012).*

In this study, we aimed to emphasizes on the usefulness of M-mode echocardiographic methods like the mitral annular plane systolic excursion (MAPSE) correlated to speckle tracking by 2 dimensional (STE) and tissue Doppler imaging in prediction of subclinical LV systolic dysfunction in patients with type 2 diabetes.

PATIENTS AND METHODS

The study included 100 asymptomatic patients with type II DM and 50 healthy control subjects. The included cases and healthy individuals were recruited from the outpatient clinic in Maadi Military Hospital in Cairo between October 2019 and July 2020.

All patients and control group were subjected to full history taking laboratory investigations including HbA1c, lipid profile and 2D Transthoracic Echocardiography & speckle tracking echocardiography.

Ejection fraction (EF) was calculated from estimation EDV and ESV using the following formula: EF = (EDV – ESV) / EDV (Modified Simpson's method). It was obtained from apical 4 (single plane) combined with apical 2 (biplane method) *(Lang et al., 2015).*

Pulsed wave Doppler of mitral inflow velocities were obtained to measure

diastolic early filling velocity (E) wave and late diastolic velocity (A) wave, E/A ratio, and E wave. Pulsed wave TDI velocity measurements were obtained by placing the sample volume at the mitral annular level either from the septal or lateral annulus. From these recordings, myocardial systolic (Sa), early diastolic (Ea), and E/Ea ratio were measured and averaged *(Kadappu and Thomas, 2015)*.

MAPSE was measured using M-mode imaging in the apical four chamber view. The M-mode cursor was placed on the septal and lateral mitral annulus parallel to the LV walls, and then both values were averaged. Measurements took place from the end of diastole, until maximal expansion in systole *(Zaky et al., 1967)*.

GLS was measured by using 2DSTE by apical 4 chamber view, apical 2 chamber view and apical long axis view. Tracking quality was visually checked, and accepted by the operator. Strain values of LV were assessed automatically in 17 segments, and then averaged manually by the operator as mean value of each strain and presented as a bull- eye map and GLS was automatically calculated *(Lang et al., 2015)*.

Statistical Analysis:

Data entry, processing and statistical analysis was carried out using MedCalc ver. 18.11.3 (MedCalc, Ostend, Belgium). Mean, Standard deviation (± SD) and range for parametric numerical data, while Median and Inter-quartile range (IQR) for non-parametric numerical data, Frequency and percentage of non-numerical data. Tests of significance (Mann-Whitney's, Chi square tests, linear regression analysis and ROC Curve) analysis were used. Data were presented and suitable analysis was done according to the type of data (parametric and non-parametric) obtained for each variable. P-values less than 0.05 were considered statistically significant. Mann-Whitney's Test (U test) was used to assess the statistical significance of the difference of a non-parametric variable between two study groups. Chi-Square test was used to examine the relationship between two qualitative variables. Linear regressions used in Correlations between continuous variables were presented as Pearson correlation coefficient. The ROC Curve (receiver operating characteristic) provided a useful way to evaluate the Sensitivity and specificity for quantitative Diagnostic measures that categorize cases into one of two groups.

RESULTS

The study included 100 asymptomatic patients with type II DM 65 of them were females, with age [55 (53 – 58) years]. And 50 healthy subjects as a control group 27 of them were females, with age [55 (50 – 60) years]. Comparative study between the 2 groups revealed non-significant difference as regards age and sex of the patients (p > 0.05).

There were significant difference in serum creatinine, urea, RBS, HbA1C, albuminuria, TGs and LDL&HDL in DM in DM cases group; compared to control group (p < 0.01 respectively) **(Table 1)**.

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Table (1): Comparison between the two groups as regarding demographic, clinical and laboratory data

Parameters \ Groups		Control group (50)	DM cases group (100)	Chi square test P value
Gender Female male		27 (54%) 23 (46%)	65 (65%) 35 (35%)	= 0.1937
Variables		Control group (50)	DM cases group (100)	Mann-Whitney's U test
		Median (IQR)	Median (IQR)	P value
Age (years)		55 (50 – 60)	55 (53 – 58)	= 0.8182
BMI		24.5 (23.1 – 25.1)	26.6 (25.7 – 28.3)	< 0.0001
SBP (mmHg)		115 (110 – 120)	130 (120 – 135)	< 0.0001
DBP (mmHg)		70 (70 – 75)	75 (70 – 85)	< 0.0001
Routine lab	**Creat.** (mg/dL)	0.7 (0.7 – 0.8)	1 (0.8 – 1.1)	< 0.0001
	Urea (mg/dL)	21 (18 – 24)	30.5 (25 – 37)	< 0.0001
	RBS (mg/dL)	92 (87 – 102)	194 (184 – 210)	< 0.0001
	HbA1C (%)	5.3 (5.2 – 5.7)	7.8 (7.4 – 8.2)	< 0.0001
	Albuminuria (mg)	19 (17 – 24)	25 (21 – 27)	< 0.0001
Lipid profile	**T.Cholesterol**(mg/dL)	132 (126 – 145)	195 (185 – 197)	< 0.0001**
	TGs (mg/dL)	114.5 (106 – 124)	187 (171 – 196)	< 0.0001**
	HDL (mg/dL)	53 (50 – 54)	42 (40 – 46)	< 0.0001**
	LDL (mg/dL)	85 (75 – 102)	135 (128 – 140.5)	< 0.0001**

BP: blood pressure; DM: diabetes mellitus; RBS: random blood sugar FBS: fasting blood sugar; Creat: creatinine, HDL: high density lipoprotein; LDL: low density lipoprotein; TGs: triglycerides. T. Cholesterol: total cholesterol

Regarding correlation with 2D Echo-Doppler parameters; the study revealed highly significant difference in A wave [0.9 (0.84 – 0.92)] in DM cases group; compared to control group [0.65 (0.65 – 0.8)] ($p < 0.01$). It also revealed; highly significant difference in EF [60 (57.5 – 62)], E wave [0.69 (0.62 – 0.71)] and E/A ratio [0.79 (0.73 – 0.85)], in DM cases group; compared to control group EF [64 (63 – 68)], E [0.9 (0.9 – 0.9)] E/A ratio[1.29 (1.12 – 1.4)] .($p < 0.01$ respectively).

Regarding 2DSTE, study revealed; highly significant difference in GLS in DM cases group [-16.8 (-17.8 to -16.3)]; compared to control group [-22.8 (-23.1 to -21.4)] ($p < 0.01$).

Regarding MAPSE; Comparative study between the 2 groups revealed; highly significant decrease in septal, lateral and average MAPSE, in DM cases group, average [MAPSE 1.2 (1.1 – 1.25)] compared to control group [average MAPSE 1.4 (1.3 – 1.5)] ($p < 0.05$ respectively).

Regarding TDI; study between the 2 groups revealed; highly significant difference in Aa [0.09 (0.08 – 0.1)] and E/Ea ratio [8.6 (7.7 – 9.5)] in DM cases group; compared to control group Aa (m/s) [0.07 (0.07 – 0.07)] E/Ea ratio [6 (6 – 6.4)] ($p < 0.01$ respectively). And it also revealed; highly significant difference in Sa [0.08 (0.07 – 0.09)] and Ea [0.08 (0.07 0.08)] in DM cases group; compared to control group Sa [0.1 (0.09 – 0.11)] and Ea [0.15 (0.14 – 0.15)] ($p < 0.01$ respectively) **(Table 2)**.

Table (2): Comparison between the 2 groups as regards Echocardiographic data using Mann-Whitney's U test

Variables \ Groups		Control group (50) Median (IQR)	DM cases group (100) Median (IQR)	Mann-Whitney's U test P value
2D Echo-Doppler	**EF** (%)	64 (63 – 68)	60 (57.5 – 62)	< 0.0001
	E (m/s)	0.9 (0.9 – 0.9)	0.69 (0.62 – 0.71)	< 0.0001
	A (m/s)	0.65 (0.65 – 0.8)	0.9 (0.84 – 0.92)	< 0.0001
	E/A ratio	1.29 (1.12 – 1.4)	0.79 (0.73 – 0.85)	< 0.0001
TDI	**Sa** (m/s)	0.1 (0.09 – 0.11)	0.08 (0.07 – 0.09)	< 0.0001
	Ea (m/s)	0.15 (0.14 – 0.15)	0.08 (0.07 – 0.08)	< 0.0001
	Aa (m/s)	0.07 (0.07 – 0.07)	0.09 (0.08 – 0.1)	< 0.0001
	E/Ea ratio	6 (6 – 6.4)	8.6 (7.7 – 9.5)	< 0.0001
2D STE	**GLS** (%)	-22.8 (-23.1 to -21.4)	-16.8 (-17.8 to -16.3)	< 0.0001
MAPSE	**Septal MAPSE** (cm)	1.3 (1.3 – 1.4)	1.2 (1.1 – 1.2)	< 0.0001
	Lateral MAPSE (cm)	1.5 (1.4 – 1.6)	1.2 (1.1 – 1.3)	< 0.0001
	Average MAPSE (cm)	1.4 (1.3 – 1.5)	1.2 (1.1 – 1.25)	< 0.0001

A: late diastolic velocity, Aa: late diastolic annular velocity, E: early diastolic velocity, Ea: early diastolic annular velocity, EF: ejection fraction, GLS: global longitudinal strain, MAPSE: mitral annular plane systolic excursion, Sa: systolic annular velocity.

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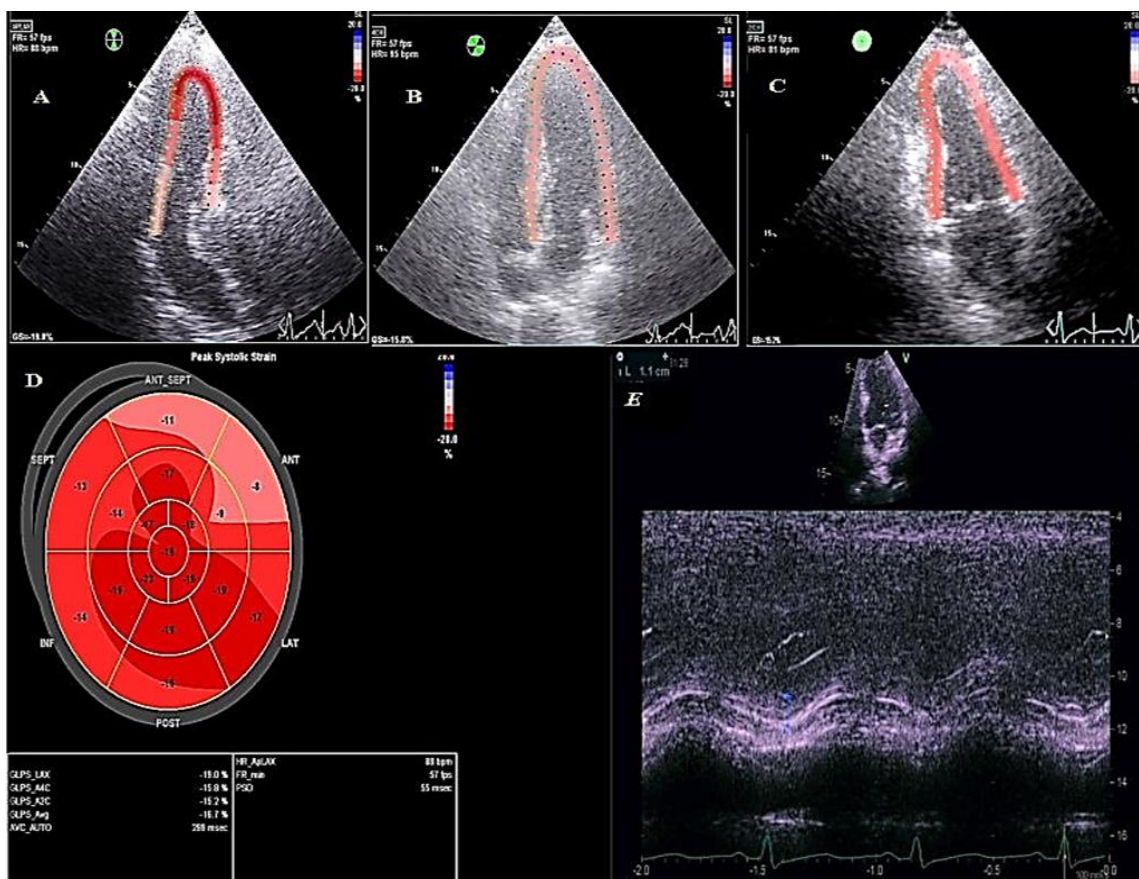
Figure (1): 2D-speckle tracking echocardiography images for a diabetic Case, showing longitudinal strain in different segments of LV in apical 4,2,3 chamber views (image A, B, C respectively) and the Bull`s eye showing the reduced global longitudinal strain (image D), the last image E shows MAPSE of the same patient at the lateral mitral annulus

On correlation analysis by applying linear regression analysis we found that MAPSE linearly had a significant positive correlation with EF, GLS and Sa wave [r= 0.565, 0.723, 0.595 respectively]. (p < 0.01). But it was linearly had a significant negative correlation with DM duration and HbA1C Level[r= -0.495, -0.776 respectively] (p < 0.01) **(Table 3 and fig. 2).**

Table (3): Linear correlation analysis between MAPSE and GLS, Sa, DM & HbA1C.

Associated MAPSE Factor	r	P
EF	0.565	<0.0001**
GLS	0.723	<0.0001**
Sa wave	0.595	<0.0001**
DM duration	-0.495	<0.0001**
HbA1C	-0.776	<0.0001**
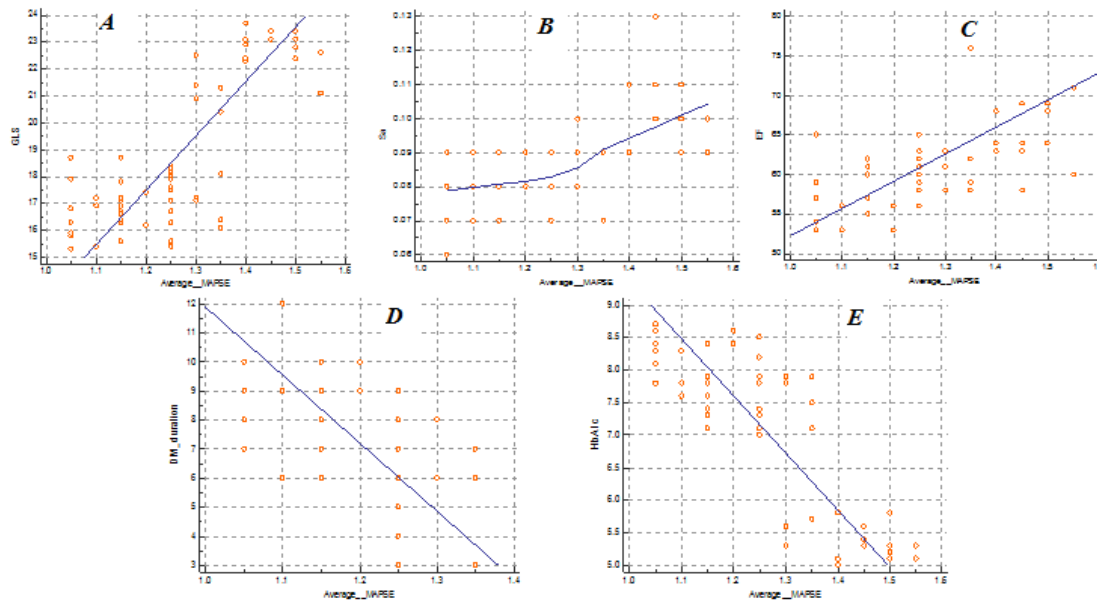
Figure (2):Linear regression curves between MAPSE and GLS, Sa, DM & HbA1C A= MAPSE and GLS., B= MAPSE and Sa, C= MAPSE and EF. D= MAPSE and DM duration, E=MAPSE and. HbA1c

Comparing between sensitivity and specificity of MAPSE and other echocardiographic modality in detection of subclinical LV dysfunction by using ROC Curve **(Table 4)**.

Table (4): Roc-curve of MAPSE, E/Ea& Sa to predict patients with Subclinical LVSD

Variable	AUC	SE	Best Cut off point (Criterion)	Sensitivity (%)	Specificity (%)	P value
E/Ea ratio	0.760	0.0401	>7	92.59	59.38	<0.0001**
Average MAPSE	0.820	0.0341	≤1.2	68.52	82.29	<0.0001**
Sa wave	0.681	0.0433	≤0.09	100	28.12	<0.0001**

To measure the sensitivity and specificity of MAPSE for predicting subclinical left ventricular systolic dysfunction in type II diabetic patients,the ROC curve was applied.Average MAPSE at a cut-off point (≤1.2cm) with good (82%) accuracy, sensitivity= 68% and specificity= 82% (p < 0.01). when compared to the GLS in prediction of subclinical dysfunction.E/Ea ratio at a cutoff point (>7) predicted patients with Subclinical LVSD, with fair (76%) accuracy, sensitivity= 92% and specificity= 59% (p < 0.01).Sa wave at a cutoff point (≤0.09m/s) predicted patients with Subclinical LVSD, with fair (70%) accuracy, sensitivity= 100% and specificity= 28% (p < 0.01) **(Figure 3)**.
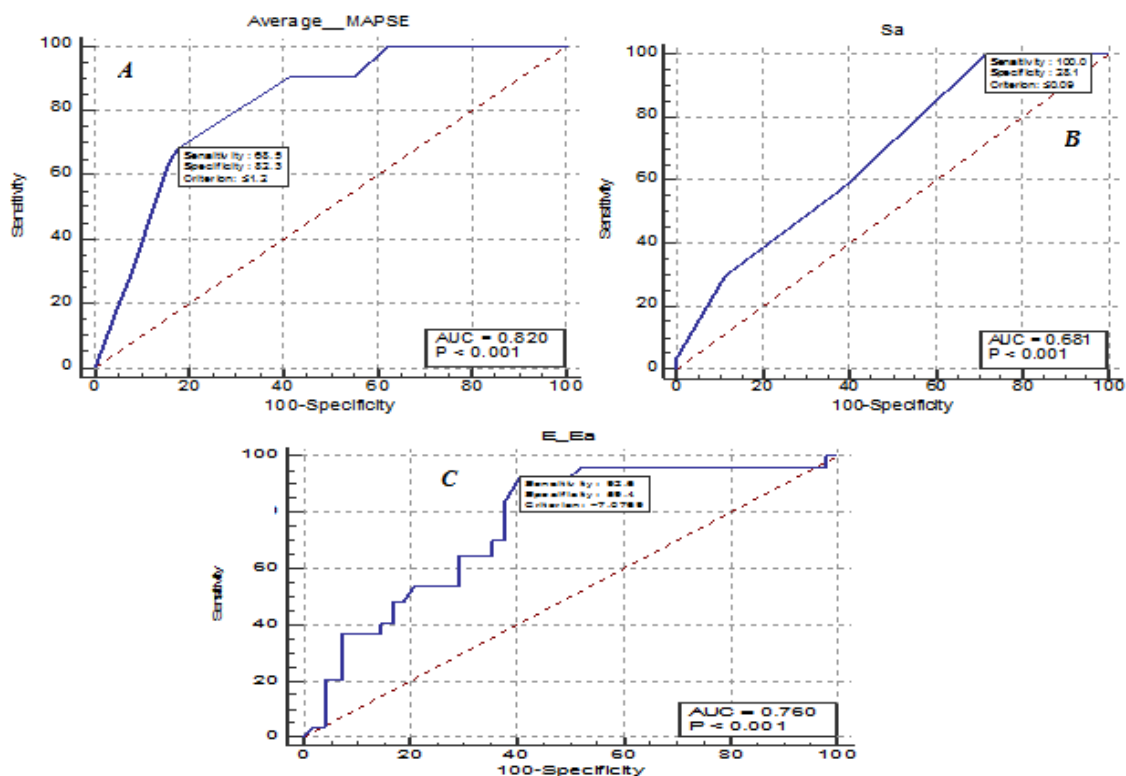
Figure (3): Roc-curve of MAPSE, E/Ea & Sa to predict patients with Subclinical LVSD
A= ROC curve of MAPSE,B= ROC curve of E/Ea ratio, C= ROC curve of S wave

DISCUSSION

Our case-control study revealed a significant decrease in average MAPSE, in DM cases group compared to control group with average MAPSE at a cut-off point (≤1.2) predicted patients with Subclinical LVSD, with good (82%) accuracy, sensitivity= 68% and specificity= 82%. also, Linear correlation analysis showed that, EF had a significant positive correlation with MAPSE. This was in agreement with *Loncarevic et al. (2016)* who found that MAPSE were significantly lower in all patients with DM compared to control.

Our study disagreed with *Grue et al. (2018)* who studied that mitral annular plane systolic excursion ≤10 mm detected left ventricular dysfunction with 82%

specificity, 76% specificity, 56% positive predictive value and 92% negative predictive value. This discrepancy might be a result of different sample size and also due to study population including patients with systolic and diastolic dysfunction which might lower the results.

In our study, regarding linear correlation of MAPSE with the GLS, there was a strong correlation between MAPSE and longitudinal strain. Linear correlation analysis showed that; GLS had a significant positive correlation with MAPSE. This was in agreement with *Wenzelburger et al. (2011)* who found that, at rest MAPSE correlated with longitudinal strain and on exercise with longitudinal strain. *Huang et al. (2017)* also found that longitudinal strain was

significantly lower in diabetes patients as compared to the control.

Our case-control study revealed significant difference in GLS in DM cases group compared to control group with significant positive correlation with MAPSE. 54 % of DM patients with preserved LVEF had LV longitudinal systolic dysfunction determined as GLS < 17.1%. This was in agreement with *Ernande et al. (2011)* who found that 23% of DM patients with preserved LVEF had LV longitudinal systolic dysfunction determined as GLS < 18%. *Zoroufian et al. (2014)* reported a significant reduction in longitudinal strain of LV in diabetic patients with evidence of diastolic dysfunction and in those with normal LV filling compared to the control.

In our study, regarding linear correlation with the Sa and tissue Doppler systolic annular velocity provided a sensitive tool for detection of the global longitudinal LV function. Our case-control study revealed significant difference in Sa wave in DM cases group comparing to control group with significant positive correlation with MAPSE. By using ROC-curve analysis, Sa wave at a cut-off point (≤0.09m/s) predicted patients with Subclinical LVSD, with fair (70%) accuracy, sensitivity= 100% and specificity= 28%. It also revealed significant difference in E/Ea ratio, in DM cases group compared to control group. E/Ea ratio at a cut-off point (>7m/s) predicted patients with Subclinical LVSD, with fair (76%) accuracy, sensitivity= 92% and specificity= 59%.

Dalen et al. (2010) found that mean±SD systolic mitral annular velocities (Sa) were 8.2±1.3 cm/s in women and 8.6±1.4 cm/s in men. *From et al. (2010)* concluded that pre-clinical diastolic dysfunction is prevalent in patients with DM with increase in the E/e′ ratio in diabetic patients which was associated with subsequent HF and mortality independent of hypertension, coronary disease, and other echocardiographic parameters. *Grue et al. (2018)* found that MAPSE ≤10 mm could detect left ventricular dysfunction. with 82% sensitivity and 76% specificity. Cut of value of Sa wave ≤5.4 cm/s with 83% sensitivity 68 % specificity. The discrepancy might be attributed to gain settings and, misalignment of the ultrasound beam will cause under estimation of the measurement in the Doppler spectrum of the tissue velocity.

In Our study through linear correlation analysis it has been shown that there was a negative correlation between longitudinal strain and plasma level of HBA1C and duration of diabetes. Therefore, the better control of DM may delay the progression of cardiac pathology. Many studies are recommended to strength this issue. *Elgohary et al. (2017)* reported significant statistical difference in GLS in patients with longer diabetic duration and concluded that, diabetic duration was strongly correlated with reduction of global longitudinal strain (LS).

CONCLUSION

MAPSE is considered a useful M-mode method in prediction of subclinical LV systolic dysfunction in asymptomatic type II diabetic patients and it has positive correlation to GLS & Sa wave of TDI. it

has a negative correlation with duration of DM and HbA1C Level.

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توقع الإعتلال في وظيفة البطين الأيسر الانقباضي بواسطة قياس الإنحراف الإنقباضي المسطح الحلقي الميترالي بالموجات الصوتية الخطية وعلاقتة بالتتبع النقطي لعضلة القلب بالموجات الصوتية ثنائية الأبعاد والدوبلر النسيجي في مرضي السكري النوع الثاني

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خلفيــــة البحـــث: يعتبــر داء الســكري مـــن عوامـــل الخطورةالمؤديــة اليفشـل وظـائف القلــب المصــاحببكفاءة طبيعيــة لعضـلة القلب.واكتشــاف ذلـك بمكــن بالموجـــات الصــوتية الخطيــة وقيــاس الاجهــاد الطـولي الكلــي التــي تعتبــر الطريقــة المثاليــة لاكتشــاف الاعتلال في وظائف البطين الايسر الانقباضي بين مرضي السكري.

الهـدف مـن البحـث: دراسةإسـتخدام طريقــة مــن طـرق الموجـات الصـوتية الخطيـة مثـل قيــاس الانحـــراف الانقباضـــي المسـطح الحلقـي الميترالــي فــي توقـع الاعــتلال لوظيفــة البطـين الايسـر الانقباضـي بــين مرضـي السـكري مـن النـوع وعلاقتهـا بـالتتبع النقطـي لعضلة القلب بالموجات الصوتية ثنائية الابعاد والدوبلر النسيجي الثاني.

المرضـي وطـرق البحـث: إشـتملت الدراسـة 100 مريضـا مصـابين بمـرض السـكري مــن النـــوع الثـــاني ونسـبة كفـاءة عضـلة القلب لـديهم أكثـر مـن 50 بالمئـة وخمسـين شـخص مـن الاصـحاء لا اعـراض لـديهم كمجموعـة للـتحكم. وتـم عمـل تخطـيط صـدي القلــب ثنـــائي الابعـــاد للمجمـــوعتين لجميـــع المرضـــى ومجموعـــة الـــتحكم وقيـاس الانحــراف الانقباضـــي المســطح الحلقـي الميترالــي بالموجـــات الصــوتية الخطيـة، وقيـاس الاجهــاد الطـولي الكلــي بـالتتبع النقطـي لعضـلة القلــب، وموجـات الـدوبلر النسيجي.

نتــائج البحـــث: شـملت الدراسـة 150 مريضـا قسـموا الـي 100 مـريض مـنهم بالسـكري النـوع الثـاني (65 مـنهم مـن الانـاث) و50 شـخص مـن الاصـحاء (27 مـنهم

مـــن الانـــاث) وقـــد اظهـــرت الدراســـة المقارنـــة بـــين المجمـــوعتين انخفاضـــاً كبيـــراً فـــي قيـــاس الانحـــراف الانقباضـــي المســـطح الحلقـــي الميترالـــي بـــين مجموعـــة مرضـــي الســـكري[1.2 (1.25-1.1) ســـم [مقارنـــة بمجموعـــة الـــتحكم مـــن الاصـــحاء] 1.4 (1.5-1.3) ســـم [كمـــا أظهـــرت قياســـات الارتبـــاط الخطـــي وجـــود إرتباطـــا إيجابيـــا بـــين الانحـــراف الانقباضـــي المســـطح الحلقـــي الميترالـــي وكـــلا منالاجهـــاد الطـــولي الكلـــي والـــدوبلر النســـيجي، وأيضـــا قياســـات كفـــاءة عضـــلة القلـــب. وأظهـــرت النتـــائج وجـــود ارتباطـــا ســـلبيا بينـــه وبـــين فتـــرة الإصـــابة بالســـكري ومســـتوي الهيموجلـــوبين الســـكري فـــي الـــدم. الحـــد الأدنـــى الفاصـــل فـــي متوســـط الانحـــراف الانقباضـــي المســـطح الحلقـــي الميترالـــي لاكتشـــاف الاعـــتلال فـــي وظيفـــة البطـــين الايســـر الانقباضـــي هـــو اقـــل مـــن 1.2 سمبدقة جيدة (82%) وحساسية (68%) وخصوصية (82%).

الاســـتنتاج: يعتبـــر قيـــاس الإنحـــراف الانقباضـــي المســـطح الحلقـــي الميترالـــي طريقـــة مفيـــدةفي توقـــع الإعـــتلال لوظيفـــة البطـــين الايســـر الانقباضـــي بـــين مرضـــي الســـكري مـــن النـــوع الثـــاني مـــع ظهـــور تـــرابط قـــوي بينهـــا وبـــين الطـــرق الرئيســـية المســـتخدمة فـــي التشخيص مثل الإجهاد الطولي الكلي والدوبلر النسيجي.

الكلمـــات الدالـــة: الإعـــتلال فـــي وظيفـــة البطـــين الأيســـر الانقباضـــي، مـــرض الســـكري، الإنحـــراف الإنقباضـــي المســـطح الحلقـــي الميترالـــي، التتبـــع النقطـــي لعضـــلة القلـــب، والدوبلر النسيجي.