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General Cardiology

Epicardial Adipose Tissue Was Highly Associated with Reduction in Left Ventricular Diastolic Function as Early as in Adolescence

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Background: Epicardial adipose tissue (EAT) is increased in adolescents with obesity and may play a role in early cardiovascular pathophysiological changes. There is a lack of evidence focusing on the association between EAT and cardiac function in adolescents. This study explored associations between EAT, left ventricle (LV) geometric, and LV functional changes in adolescents.

Methods: Adolescent volunteers between 10 and 20 years of age were included. Body mass index (BMI) was presented as age- and sex-specific BMI z-scores. Blood samples for glucose metabolism, lipid profiles, and high-sensitivity C-reactive protein (hs-CRP) were obtained. EAT thickness, LV hypertrophy, and LV diastolic function were measured by echocardiography.

Results: The mean age of the 276 adolescents was 13.51 ± 2.44 years. BMI z-score was strongly associated with EAT thickness ($r = 0.77$; $p < 0.001$). Multivariable analysis revealed that age, insulin resistance, total cholesterol to high-density lipoprotein cholesterol ratio, and hs-CRP were independent predictors of increased EAT thickness. After adjusting for sex, age, and BMI z-score by multivariable analysis, EAT thickness was a strong predictor of higher LV mass indexed to height$^{2.7}$, higher relative wall thickness, lower mitral annulus e′/a′, and higher E/e′ of the mitral annulus. There was no association between EAT and LV ejection fraction.

Conclusions: EAT was highly associated with LV hypertrophy and reduction in LV diastolic function, independent of BMI z-score in the enrolled adolescents. Of note, the negative impacts of EAT on LV geometry and diastolic function occurred as early as in adolescence. This highlights the importance of preventing obesity and EAT deposition early in life.

Key Words: Adolescent • Diastolic function • Epicardial adipose tissue • LV hypertrophy • Obesity

INTRODUCTION

Epicardial adipose tissue (EAT) is a fat deposit between the myocardium and visceral pericardium. It is located around the epicardial coronary vessels, and it can extend and be interspersed with myocardial muscle fibers. EAT is supplied by branches of the coronary arteries, which share the same microcirculation with the myocardium.[1] EAT is different from pericardial adipose tissue, which is the fat depot outside the visceral pericardium. Pericardial fat is supplied by noncoronary sources, such as the internal mammary artery.[2] The amount of EAT on the right ventricle surface increases with age in people younger than 20 years old.[3] Obesity is also associated with an increased amount of EAT in adults.[4]

EAT thickness, but not pericardial or total cardiac fat, is associated with the incidence of cardiovascular disease (myocardial infarction, need for percutaneous coronary intervention, need for coronary artery bypass

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grafting, congestive heart failure, and atrial fibrillation) and all-cause mortality in patients with type 2 diabetes.[5] The association between EAT and cardiovascular events is well established in community-based adult populations and in patients with coronary artery disease[6,7] independently of traditional cardiovascular risk factors.

A meta-analysis of adults showed that EAT was associated with left atrial dilation, left ventricular (LV) hypertrophy, and diastolic dysfunction (high E/e' ratio, lower e' velocity) independently of body mass index (BMI).[8] Increased EAT volume has been independently associated with increased LV mass in adult patients with untreated hypertension, and an increase in EAT thickness has also been associated with microalbuminuria in hypertensive adults with LV hypertrophy.[9] In patients at risk of heart failure with preserved ejection fraction, medications that can reduce proinflammation of epicardial fat may lower the risk of heart failure.[10] Taken together, these studies demonstrate the negative impact of epicardial fat on the heart. However, these studies have mainly focused on adults, and evidence from children and adolescence is still sparse. Therefore, the aim of this study was to investigate whether reductions in LV diastolic function and LV hypertrophy occurred with increased EAT thickness as early as in adolescence.

MATERIALS AND METHODS

This was a cross-sectional study of adolescents aged were between 10 and 20 years. The study project was approved by the Institutional Review Board of E-Da Hospital (approval number EMRP64106N). Informed consent was well explained, and consent forms were signed by both legal guardians and the participants themselves.

Study population and enrollment criteria

The study recruitment information was posted on the hospital's internet homepage. All of the enrolled adolescents actively contacted the study team, and they voluntarily participated in this study. The inclusion criteria were an age between 10 and 20 years, which fulfills the definition of adolescence according to the World Health Organization statement.[11] Participants were excluded if they had known chronic medical illnesses or acute inflammatory illnesses, including autoimmune diseases, previously known heart disease, diabetes mellitus, kidney disease, and acute or chronic infections. Body height, body weight, BMI, waist circumference, hip circumference, and blood pressure (BP) were measured in all of the participants. Obesity was defined as age- and sex-specific BMI $\geq 95^{th}$ percentile based on the BMI reference for Taiwanese adolescents.[12] BMI was corrected and presented as age- and sex-specific BMI z-scores. BP was acquired in an automated office BP measurement setting, and the mean of five BP measurements was recorded. Hypertension was defined as anaverage systolic BP and/or diastolic BP $\geq$ 95th percentile (on the basis of age, sex, and height percentiles) in those who were 10-12 years old. For those $\geq$ 13 years of age, hypertension was defined as systolic BP $\geq$ 130 mmHg and/or diastolic BP $\geq$ 80 mmHg.[13]

Laboratory biomarkers

Blood samples were obtained after fasting for 8 hours, including fasting glucose, glycated hemoglobin (HbA1c), insulin, total cholesterol, high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), triglycerides, and high-sensitivity C-reactive protein (hs-CRP). Insulin resistance was calculated using the modified homeostasis model assessment of insulin resistance: fasting plasma glucose (mmol/l) times fasting serum insulin (mU/l) divided by 22.5.[14]

Echocardiographic protocol

All of the participants underwent echocardiography, and all echocardiographic indices were measured according to the protocols of the American Society of Echocardiography and the European Association of Cardiovascular Imaging.[15]

EAT thickness was measured between the free wall of the right ventricle myocardium and visceral pericardium on long- and short-axis views of echocardiography at end-systole, perpendicular to the aortic annulus.[16] The thickness of EAT was obtained from the average of 3 cardiac cycles at end-expiratory phase, and expressed in mm (Figure 1). Under the standard measurement technique, the intra-observer and inter-observer agreement and reproducibility for EAT were excellent.[17,18] LV geometry was classified into 4 categories based on LV mass indexed to the height$^{2.7}$ (LVMI$^{height2.7}$) and relative wall thickness.[19] Relative wall thickness was calculated as:

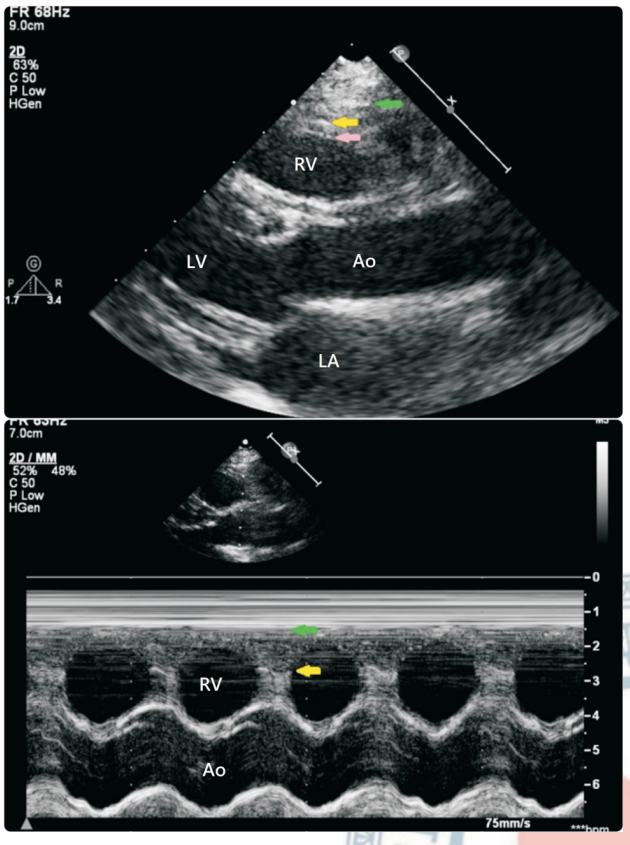

Figure 1. *EAT was measured by 2D echocardiography and M-mode. The pink arrow indicates the endocardium, the yellow arrow indicates the outer margin of the myocardium, and the green arrow indicates the visceral pericardium. EAT is the layer between the outer margin of the myocardium (yellow) and the visceral pericardium (green). Ao, aorta; LA, left atrium; LV, left ventricle; RV, right ventricle.*

(septal wall thickness + posterior wall thickness)/LV internal dimension at end-diastole. The 4 geometric patterns of LV mass were: (1) $LVMI^{height2.7}$ < 34.02 $g/m^{2.7}$ for girls and < 37.08 $g/m^{2.7}$ for boys, and a relative wall thickness ≤ 0.36 was classified as normal LV geometry; (2) concentric LV hypertrophy was defined as normal $LVMI^{height2.7}$ with increased relative wall thickness > 0.36; (3) eccentric LV hypertrophy was defined as increased $LVMI^{height2.7}$ (girls ≥ 34.02 $g/m^{2.7}$, boys ≥ 37.08 $g/m^{2.7}$) with normal relative wall thickness; and (4) concentric and increased LV hypertrophy was defined as increased $LVMI^{height2.7}$ and relative wall thickness > 0.36.[20]

LV diastolic function was evaluated by E/A ratio of trans-mitral flow, average peak diastolic mitral annular velocity ratio (mitral lateral annulus e′/a′, mitral septal annulus e′/a′), E/e′ (early inflow to peak early annular velocity ratio) of the mitral lateral annulus, and E/e′ of the mitral septal annulus. Three measurements of each diastolic indices were performed to acquire the mean value. LV systolic function was evaluated according to LV ejection fraction. To avoid technique and measurement bias, all echocardiography was performed by the first experienced cardiologist (MC Yang) using a Philips iE33 system with a 1- to 5-mHz cardiac transducer. All parameters on echocardiographic images were reviewed and re-confirmed by the second cardiologist (JR Wu).

Statistical analysis

Statistical analyses were conducted using SPSS version 22. Numerical data were presented as mean ± standard deviation (mean ± 1 standard deviation). The two-tailed Student's *t*-test was used to compare the means of independent data between non-obese and obese groups, including age, waist circumference, hip circumference, waist-to-hip ratio, BP, glucose metabolic biomarkers, insulin resistance, lipid profiles, hs-CRP, and echocardiographic parameters. If avariable was not normally distributed, the Mann-Whitney U test was used to compare the variable between the two groups. Bonferroni correction was applied to adjust for multiple comparisons. The chi-square test was used to determine statistical differences between categorical data. Pearson correlation coefficients were used to measure linear correlations between BMI z-score and EAT thickness. Univariable analysis for the effect of each covariate on EAT thickness was performed using a linear regression model. The covariates included sex, age, and serum biomarkers (HbA1c, insulin, insulin resistance, total cholesterol, HDL-C, LDL-C, triglycerides, total cholesterol/HDL-C ratio, hs-CRP). The candidate covariates for the multivariable model were the those with a p value ≤ 0.2 in univariable analysis. Multivariable linear regression was further used to determine the association between a covariate and EAT thickness after adjusting for other factors. The forward selection method was used to determine the final multivariable model.

Sex, age, EAT thickness, hypertension status, and BMI z-score were examined in multivariable linear regression models to determine the predictors of LV diastolic function and LV hypertrophy. Multicollinearity was examined using the variance inflation factor in each of the multivariable linear regression models. A variance inflation factor < 5 indicated that the associated inde-

pendent variable was not collinear with the other variables in this model. All p values were 2-sided, and a p value < 0.05 was regarded as statistically significant.

RESULTS

Among the 300 adolescents who participated in the study protocol, 10 were excluded from the study because they were unable to follow the examination schedules, 10 were excluded because of incomplete data, and 4 were excluded because of acute or chronic illnesses. The remaining 276 adolescents were enrolled in this study, of whom 142 participants fulfilled the criteria of obesity. All 276 participants received echocardiography and provided blood samples. The mean age was 13.51 ± 2.44 years. The baseline characteristics of the non-obese and obese groups are detailed in Table 1.

Table 1. Demographic characteristics of age, waist circumference, hip circumference, blood pressure, glucose metabolism, lipid profiles, epicardial adipose tissue, LV mass, LV functions for all and by obesity status

	All	Non-obesity	Obesity	p value
No. of participants	276	134	142	
Age (year)	13.51 ± 2.44	13.50 ± 2.46	13.52 ± 2.43	0.93*
Sex (male)	217	103 (76%)	114 (80%)	0.56#
BMI z-score	2.24 ± 2.17	0.42 ± 1.07	3.96 ± 1.42	< 0.001*
Waist circumference (cm)	83 ± 15	71 ± 9	94 ± 10	< 0.001*
Hipcircumference (cm)	95 ± 13	87 ± 9	103 ± 10	< 0.001†
Waist/hip ratio	0.87 ± 0.08	0.82 ± 0.06	0.91 ± 0.07	< 0.001*
Systolic BP (mmHg)	115 ± 15	110 ± 14	119 ± 14	< 0.001†
Diastolic BP (mmHg)	70 ± 11	67 ± 10	73 ± 10	< 0.001*
Glucose metabolism				
Fasting glucose (mg/dL)	93 ± 11	92 ± 11	93 ± 11	0.52*
HbA1c (%)	5.47 ± 0.34	5.42 ± 0.29	5.50 ± 0.37	0.07*
Insulin (uU/mL)	11.97 ± 10.35	6.21 ± 4.21	17.83 ± 11.43	< 0.001*
Insulin resistance	2.83 ± 2.94	1.40 ± 1.02	4.30 ± 3.50	< 0.001*
Lipid profiles				
Total cholesterol (mg/dL)	168 ± 30	164 ± 30	171 ± 29	0.06*
HDL-C (mg/dL)	50 ± 12	56 ± 11	46 ± 11	< 0.001*
LDL-C (mg/dL)	99 ± 38	93 ± 48	104 ± 27	0.02*
Triglyceride (mg/dL)	89 ± 56	72 ± 62	103 ± 47	< 0.001*
Total cholesterol-to-HDL-C ratio	3.48 ± 0.94	3.04 ± 0.81	3.83 ± 0.89	< 0.001*
Hs-CRP (mg/L)	2.04 ± 2.89	0.72 ± 1.13	3.06 ± 3.38	< 0.001*
Epicardial adipose tissue (mm)	5.58 ± 2.72	3.83 ± 1.42	7.73 ± 2.30	< 0.001*
LV mass				
LV mass index (g/m$^{2.7}$)	41.25 ± 11.84	34.68 ± 8.75	47.54 ± 10.98	< 0.001*
Relative wall thickness	0.42 ± 0.10	0.37 ± 0.07	0.45 ± 0.10	< 0.001*
LV function				
LV ejection fraction (%)	69 ± 6	69 ± 6	69 ± 6	0.79†
E/A ratio	2.12 ± 0.52	2.23 ± 0.53	2.02 ± 0.49	0.001*
Mitral e′ (lateral annulus)	16.33 ± 2.68	17.24 ± 2.47	15.46 ± 2.59	< 0.001†
Mitral a′ (lateral annulus)	6.73 ± 1.63	6.34 ± 1.42	7.11 ± 1.72	< 0.001*
Mitral e′/a′ (lateral annulus)	2.56 ± 0.76	2.84 ± 0.73	2.30 ± 0.69	< 0.001*
Mitral e′ (septal annulus)	11.38 ± 1.67	11.89 ± 1.65	10.90 ± 1.56	< 0.001*
Mitral a′ (septal annulus)	6.43 ± 1.49	6.24 ± 1.43	6.62 ± 1.54	0.04*
Mitral e′/a′ (septal annulus)	1.86 ± 0.51	2.00 ± 0.53	1.73 ± 0.44	< 0.001*
Mitral E/e′ (lateral annulus)	6.50 ± 1.44	6.02 ± 1.20	6.97 ± 1.50	< 0.001*
Mitral E/e′ (septal annulus)	9.26 ± 1.86	8.70 ± 1.64	9.80 ± 1.91	< 0.001*

Data presented as mean ± 1 standard deviation.

BMI, body mass index; BP, blood pressure; E/A ratio, trans-mitral early and late wave peak velocity ratio; E/e′, early inflow to peak early annular velocity ratio; e′/a′, average peak diastolic mitral annular velocity ratio; HbA1c, glycated hemoglobin; HDL-C, high-density lipoprotein cholesterol; Hs-CRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein cholesterol; LV, left ventricle.

* Mann-Whitney U test; # Chi-square; † Student's t test.

BMI z-score was strongly associated with EAT thickness ($r = 0.77$; $p < 0.001$). Of the 276 participants, 28 had hypertension, and obesity was a significant predictor of hypertension (22/142 vs. 6/134, risk ratio: 3.88, 95% confidence interval: 1.52-9.92). The adolescents with hypertension had higher $LVMI^{height2.7}$ (48.1 ± 15.2 vs. 40.7 ± 11.1, $p = 0.002$) and RWT (0.46 ± 0.08 vs. 0.41 ± 0.10, $p = 0.005$) compared to those without hypertension.

LV geometric changes and LV function

LV hypertrophy was confirmed in 130 (91%) of the obese participants. Among these obese adolescents, the most common geometric pattern of LV hypertrophy was concentric and increased LV hypertrophy (101/142, 71%), followed by eccentric hypertrophy (19/142, 13%), and concentric hypertrophy (10/142, 7%). Table 1 also summarizes the LV diastolic indices and LV ejection fraction in the obese and non-obese groups. All diastolic function variables were worse in the obese group compared to the non-obese group. There was no significant difference in LV systolic function between the two groups.

Independent predictors of EAT thickness

EAT thickness significantly increased with age in the adolescents (Table 2 and Table 3). In subgroup analysis, EAT thickness was significantly associated with increased age ($p < 0.001$) in the obese group, but not in the non-obese group. Univariable analysis revealed that BMI z-score, insulin, insulin resistance, HDL-C, triglycerides, total cholesterol/HDL-C ratio, and hs-CRP were significant predictors of increased EAT thickness. After adjustments in multivariable analysis, age, BMI z-score, insulin resistance, total cholesterol/HDL-C ratio, and hs-CRP were independent predictors of increased EAT thickness.

Table 2. Univariable analysis of predictors of epicardial adipose tissue

	Unstandardized β-coefficients	Standardized β-coefficients	Unstandardized 95% CI	p value
Sex (male)	0.538	0.089	-0.505 – 1.581	0.31
Age (year)	0.204	0.176	0.005 – 0.402	0.04
BMI z-score	1.018	0.770	0.871 – 1.165	< 0.001
HbA1c (%)	0.724	0.098	-0.623 – 2.070	0.29
Insulin (uU/mL)	0.127	0.481	0.085 – 0.169	< 0.001
Insulin resistance	0.386	0.414	0.232 – 0.540	< 0.001
Total cholesterol (mg/dL)	0.013	0.151	-0.002 – 0.029	0.10
HDL-C (mg/dL)	-0.067	-0.317	-0.103 – -0.031	< 0.001
LDL-C (mg/dL)	0.005	0.082	-0.006 – 0.015	0.37
Triglyceride (mg/dL)	0.013	0.311	0.006 – 0.021	< 0.001
Total cholesterol-to-HDL-C ratio	1.217	0.421	0.743 – 1.690	< 0.001
Hs-CRP (mg/L)	0.316	0.336	0.155 – 0.476	< 0.001

BMI, body mass index; CI, confidence interval; HbA1c, glycated hemoglobin; HDL-C, high-density lipoprotein cholesterol; Hs-CRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein cholesterol.

Table 3. Multivariable analysis of independent predictors of epicardial adipose tissue

	Unstandardized β-coefficients	Standardized β-coefficients	Unstandardized 95% CI	p value	Collinearity statistics (VIF)
Age (year)	0.356	0.302	0.173 – 0.540	< 0.001	1.12
BMI z-score	0.973	0.713	0.762 – 1.185	< 0.001	1.92
Insulin resistance	0.301	0.322	0.148 – 0.453	< 0.001	1.24
Total cholesterol-to-HDL-C ratio	1.095	0.378	0.253 – 1.938	0.01	3.91
Hs-CRP (mg/L)	0.169	0.180	0.015 – 0.323	0.03	1.25

BMI, body mass index; CI, confidence interval; HDL-C, high-density lipoprotein cholesterol; Hs-CRP, high-sensitivity C-reactive protein; VIF, The Variance Inflation Factor.
Candidate covariates used in the multivariable model included age, BMI z-score, insulin resistance, HDL-C, triglyceride, Total cholesterol-to-HDL-C ratio, and hs-CRP.

Association between EAT thickness, LV geometric changes and LV function

Most of the obese adolescents (101/142, 71%) had both increased LVMI$^{height2.7}$ and relative wall thickness. Multivariable analysis (using male sex, age, BMI z-score, hypertension status, and EAT as covariates) demonstrated that EAT thickness, male sex, and BMI z-score were independent predictors of both increased LVMI$^{height2.7}$ and increased relative wall thickness (Table 4).

Multivariable regression analysis also showed that EAT, hypertension status, and BMI z-score were independent predictors of both lower mitral lateral annular velocity e′/a′ ratio and lower mitral septal annular velocity e′/a′ ratio. In addition, EAT, age, and BMI z-score were independent predictors of both higher E/e′ ratio of the mitral lateral annulus and higher E/e′ ratio of the mitral septal annulus in multivariable analysis (Table 5). All of the statistical data indicated that EAT thickness was significantly associated with a reduction in diastolic function.

DISCUSSION

Our previous study showed that LV diastolic dysfunction occurred as early as in adolescence in obese subjects.[21] To the best of our knowledge, this is the first study to show a strong correlation between EAT thickness and early LV diastolic dysfunction in adolescents. There are several important findings in this present study. First, BMI z-score was an independent predictor of EAT thickness. Second, insulin resistance, total cholesterol/HDL-C ratio, and hs-CRP were also independent predictors of EAT thickness. Third, EAT thickness was highly associated with a reduction in LV diastolic function, independent of age, sex, hypertension status, and BMI z-score. EAT thickness was also an independent predictor of LVMI$^{height2.7}$ and increased relative wall thickness.

There are several differences between EAT and other types of body adipose tissue. Fatty acid synthesis and incorporation into EAT are higher than in pericardial, perirenal, and popliteal fat.[22] The rate of insulin-induced lipogenesis is also higher in EAT than in pericardial fat, perirenal, and popliteal adipose tissue. Moreover, the expressions of enzymes involved in lipid metabolism, mRNA for lipoprotein lipase, and stearoyl-CoA desaturase are lower in EAT compared to other adipose tissue depots.[23] EAT may also play a pathogenic role in coronary artery disease in patients with diabetes, as there are more obvious changes in fatty acid molecules in EAT compared to subcutaneous fat tissue in patients with coronary artery disease and type 2 diabetes.[24]

Serum free fatty acid level has been related to the accumulation of triglycerides in EAT and myocardium, which has also been related to peripheral vascular resistance and LV hypertrophy.[25] In a study of 74 children with obesity, triglycerides and LDL-C were related to EAT thickness in univariable analysis.[26] In addition, in a study of adults (mean age 46 ± 8 years), EAT thickness was

Table 4. Multivariable analysis of predictors of LV hypertrophy indices

	Unstandardized β-coefficients	Standardized β-coefficients	Unstandardized 95% CI	p value	Collinearity statistics (VIF)
LVMI$^{height2.7}$					
Sex (male)	5.205	0.228	1.940 – 8.471	0.002	1.01
Age (year)	-0.381	-0.087	-1.014 – 0.252	0.24	1.04
BMI z-score	3.670	0.660	3.242 – 4.340	< 0.001	1.09
Hypertension	0.055	0.001	-6.872 – 6.982	0.99	1.03
EAT (mm)	2.068	0.551	1.516 – 2.621	< 0.001	1.08
Relative wall thickness					
Sex (male)	0.031	0.168	0.002 – 0.061	0.03	1.01
Age (year)	0.003	0.079	-0.003 – 0.008	0.33	1.04
BMI z-score	0.020	0.429	0.014 – 0.025	< 0.001	1.06
Hypertension	-0.001	-0.002	-0.063 – 0.062	0.99	1.03
EAT (mm)	0.013	0.411	0.008 – 0.018	< 0.001	1.08

BMI, body mass index; CI, confidence interval; EAT, epicardial adipose tissue; LV, left ventricle; VIF, The Variance Inflation Factor. Candidate covariates used in the multivariable model included sex, age, EAT thickness, hypertension status, and BMI z-score.

Table 5. Multivariable analysis of predictors of LV function indices

	Unstandardized β-coefficients	Standardized β-coefficients	Unstandardized 95% CI	p value	Collinearity statistics (VIF)
Mitral lateral e′/a′					
Sex (male)	-0.269	-0.156	-0.559 – 0.022	0.07	1.01
Age (year)	-0.020	-0.060	-0.076 – 0.037	0.49	1.04
BMI z-score	-0.120	-0.341	-0.160 – -0.080	< 0.001	1.06
Hypertension	-0.647	-0.178	-1.264 – -0.031	0.04	1.03
EAT (mm)	-0.052	-0.185	-0.102 – -0.003	0.037	1.08
Mitral septal e′/a′					
Sex (male)	-0.209	-0.192	-0.389 – -0.029	0.023	1.01
Age (year)	0.003	0.013	-0.032 – 0.038	0.88	1.03
BMI z-score	-0.077	-0.329	-0.110 – -0.052	0.001	1.06
Hypertension	-0.611	-0.266	-0.994 – -0.229	0.002	1.03
EAT (mm)	-0.032	-0.173	-0.064 – -0.001	0.045	1.06
E/e′ (mitral lateral annulus)					
Sex (male)	0.292	0.104	-0.180 – 0.765	0.22	1.01
Age (year)	-0.108	-0.201	-0.199 – -0.016	0.021	1.04
BMI z-score	0.230	0.346	0.154 – 0.320	0.001	1.06
Hypertension	0.422	0.071	-0.580 – 1.425	0.41	1.03
EAT (mm)	0.131	0.282	0.051 – 0.211	0.002	1.08
E/e′ (mitral septal annulus)					
Sex (male)	0.670	0.171	0.042 – 1.297	0.037	1.01
Age (year)	-0.244	-0.323	-0.366 – -0.122	< 0.001	1.03
BMI z-score	0.266	0.315	0.181 – 0.371	0.02	1.06
Hypertension	0.696	0.084	-0.637 – 2.029	0.30	1.03
EAT (mm)	0.195	0.290	0.085 – 0.305	0.001	1.06
LV ejection fraction					
Sex (male)	-0.181	-0.015	-2.364 – 2.001	0.87	1.01
Age (year)	-0.189	-0.080	-0.626 – 0.247	0.39	1.09
BMI z-score	-0.233	-0.086	-1.002 – 0.535	0.55	2.60
EAT (mm)	0.133	0.065	-0.457 – 0.722	0.66	2.67

BMI, body mass index; CI, confidence interval; EAT, epicardial adipose tissue; E/e′, early inflow to peak early annular velocity ratio; e′/a′, average peak diastolic mitral annular velocity ratio; LV, left ventricle; VIF, The Variance Inflation Factor.
Candidate covariates used in the multivariable model included sex, age, EAT thickness, and BMI z-score.

correlated with serum triglycerides, HDL-C, and total cholesterol/HDL-C ratio. The authors also found that EAT was related to carotid intima-media thickness and the presence of carotid intima plaque.[27] Among the serum lipid profiles, we found that low HDL-C, high triglycerides, and high total cholesterol/HDL-C ratio were associated with increased EAT thickness in univariable analysis, but that only high total cholesterol/HDL-C ratio was an independent risk factor for increased EAT thickness in multivariable analysis.

Increased EAT thickness has been associated with LV hypertrophy, regardless of hypertension.[28] Small irregular aggregates of adipose tissue accumulate fat between myocardial muscle fibers and separate myocardial cells, and intracardiac and extracardiac adiposity can result in increased heart weight and cause LV hypertrophy.[29] Increased plasma fatty acid, which is related to triglyceride accumulation in EAT and myocardium, can cause elevation of catecholamine concentrations and cardiac autonomic nervous system activity, which may also lead to myocardial hypertrophy.[30] Proinflammatory cytokines, including interleukin-6, transforming growth factor-β, and macrophage chemotactic factor-1 can be secreted by EAT,[31,32] and cell culture models have revealed that these proinflammatory cytokines can accelerate myocardial hypertrophy.[33] Taken together, these findings sug-

gest that higher EAT thickness contributes to hypertrophic geometric patterns.

One previous study has evaluated the relationship between EAT thickness with atherosclerosis indicators and cardiac functional changes in obese adolescents.[34] The authors concluded that EAT thickness was positively correlated with BMI z-score, homeostasis model assessment-insulin resistance, triglyceride levels, HDL-C, LV hypertrophy, and myocardial performance index in adolescents with both obesity and metabolic syndrome, but that LV diastolic functional changes were not related to EAT in the adolescents with obesity. However, they only evaluated diastolic function by transmitral early and late wave peak velocities (E, A, E/A ratio). In our study, we also found that EAT was not associated with early and late wave peak velocities. Nonetheless, when using E/e′ and e′/a′, EAT thickness was significantly correlated with LV diastolic functional changes.

Several adult studies have reported a correlation between EAT and LV diastolic dysfunction. Several LV diastolic dysfunction indices have been used, including peak early mitral annular velocities (e′ septal, or e′ lateral), transmitral flow [early (E) and late (A) diastolic peak flow velocities ratio (E/A ratio)], and E/e′ ratio. Of these parameters, E/e′ and peak early mitral annular velocity e′ are most commonly evaluated. E/e′ has been positively correlated with EAT thickness, and e′ has been inversely correlated with EAT thickness. However, there is no consistent association between E/A ratio and EAT thickness.[35] In our study, we found a significantly positive correlation between EAT thickness and E/e′, and an inverse correlation between EAT thickness and e′/a′. This demonstrates that the negative impact of EAT on diastolic function also occurs in adolescents.

In adults, EAT thickness has also been reported to be significantly higher in patients with diastolic dysfunction compared to patients with normal diastolic function. In a previous study, most patients had grade 1 diastolic dysfunction, and fewer had grade 2 diastolic dysfunction.[35] Of the studies reporting LV diastolic dysfunction, all populations were adults, including those who had suspected or established coronary artery disease,[36,37] atrial fibrillation,[38] cardiomyopathy,[39] congestive heart failure,[40] post-myocardial infarction,[41] or those who were referred for screening.[42] In these adult studies, comorbidities were common. Therefore, diastolic dysfunction may be the result or the cause of the associated cardiac diseases. EAT may play a certain pathogenic role leading to diastolic dysfunction in such situations, however it may be also augmented by coexisting cardiac comorbidities. In our study, although the adolescents were obese, they were otherwise healthy. The impact of other cardiac comorbidities or chronic illnesses was eliminated. Our results showed that EAT was not only highly correlated with diastolic dysfunction, but also that the process occurred as early as in adolescence.

The causes of diastolic dysfunction are multifactorial. EAT has been shown to play a pathophysiological role in diastolic dysfunction, and it has anatomical contact with the myocardium without fascial interruption.[43] This may therefore result in a direct compressive force on the myocardium.[44] EAT is located between the myocardium and visceral layer of the pericardium, and mainly at the interventricular and atrioventricular sulcus along with the main coronary arteries, and therefore the coronary arteries are embedded in EAT. Increased EAT thickness has been highly associated with impaired coronary blood flow reserve, even in those with angiographically normal coronary arteries, because of a passive perivascular compressive effect.[45,46] Impaired endothelial coronary flow reserve is strongly related to LV diastolic dysfunction. In addition, the myocardium and surrounding EAT share the same coronary blood supply, so paracrine effects on the neighboring myocardium are also present. Adipokines within EAT can cause increases in collagen turnover, local inflammation, and impaired microvascular relaxation, which have a negative impact on diastolic dysfunction.[47] Cytokines secreted from EAT, including interleukin-6 and transforming growth factor-B, mediate myocardial fibrosis and may contribute to diastolic dysfunction.[48,49]

Limitations

Our results demonstrated a positive association between EAT thickness and LV hypertrophy in the enrolled adolescents, and also an inverse correlation between EAT thickness and LV diastolic dysfunction parameters. Although possible interference effects from comorbidities were eliminated in these young otherwise healthy participants, there are still some limitations. First, the pathophysiological interactions between LV hypertrophy and other biophysiological processes are complex. Sev-

eral factors can influence LV remodeling and hypertrophy, such as glucose metabolism, lipid dysregulation, inflammation reactions, and microcirculation of the coronary arteries. All of these factors may also contribute to EAT thickness, however we could not explore all possible interactions in this study. The interactions may be clarified by further laboratory studies. Second, the changes in diastolic function were subclinical, and no clear cutoff values of E/e′ or e′/a′ velocity ratio to indicate diastolic dysfunction in children and adolescents have been reported in previous studies. Third, this cross-sectional study only included longitudinal follow-up changes in EAT thickness, LV hypertrophy, and LV diastolic function. The findings will be more convincing if the reversibility of EAT thickness and diastolic dysfunction are observed after a reduction in body weight in adolescents. However, our findings provide a direction for further studies. That is, longitudinal changes in BMI z-score, EAT thickness, and LV diastolic function from adolescence to young adulthood are worth researching.

CONCLUSIONS

In the enrolled adolescents, BMI z-score, insulin resistance, total cholesterol/HDL-C ratio, and hs-CRP were positively associated with increased EAT thickness. EAT thickness was strongly associated with LV hypertrophy and reduction in LV diastolic function. Although the effects of EAT on LV geometry and diastolic dysfunction are not surprising in adults, our results highlight that these effects can occur as early as in adolescence, which has not been addressed in other studies. This emphasizes the importance of preventing obesity and EAT deposition early in life.

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DECLARATIONS OF CONFLICT OF INTEREST

All the authors declare no conflict of interest.

REFERENCES

1. Marchington JM, Mattacks CA, Pond CM. Adipose tissue in the mammalian heart and pericardium: structure, foetal development and biochemical properties. *Comp Biochem Physiol B* 1989; 94:225-32.
2. McGavock JM, Victor RG, Unger RH, Szczepaniak LS; American College of Physicians and the American Physiological Society. Adiposity of the heart, revisited. *Ann Intern Med* 2006;144:517-24.
3. Tansey DK, Aly Z, Sheppard MN. Fat in the right ventricle of the normal heart. *Histopathology* 2005;46:98-104.
4. Iacobellis G, Ribaudo MC, Assael F, et al. Echocardiographic epicardial adipose tissue is related to anthropometric and clinical parameters of metabolic syndrome: a new indicator of cardiovascular risk. *J Clin Endocrinol Metab* 2003;88:5163-8.
5. Christensen RH, von Scholten BJ, Hansen CS, et al. Epicardial adipose tissue predicts incident cardiovascular disease and mortality in patients with type 2 diabetes. *Cardiovasc Diabetol* 2019; 18:114.
6. Morales-Portano JD, Peraza-Zaldivar JÁ, Suárez-Cuenca JA, et al. Echocardiographic measurements of epicardial adipose tissue and comparative ability to predict adverse cardiovascular outcomes in patients with coronary artery disease. *Int J Cardiovasc Imaging* 2018;34:1429-37.
7. Mahabadi AA, Berg MH, Lehmann N, et al. Association of epicardial fat with cardiovascular risk factors and incident myocardial infarction in the general population: the Heinz Nixdorf Recall Study. *J Am Coll Cardiol* 2013;61:1388-95.
8. Mancio J, Azevedo D, Fragao-Marques M, et al. Meta-analysis of relation of epicardial adipose tissue volume to left atrial dilation and to left ventricular hypertrophy and functions. *Am J Cardiol* 2019;123:523-31.
9. Eren H, Omar MB, Kaya Ü, et al. Increased epicardial adipose tissue thickness is associated with microalbuminuria in hypertensive patients with left ventricular hypertrophy. *Clin Exp Hypertens* 2021;43:18-25.
10. Packer M. Drugs that ameliorate epicardial adipose tissue inflammation may have discordant effects in heart failure with a preserved ejection fraction as compared with a reduced ejection fraction. *J Card Fail* 2019;25:986-1003.
11. Canadian Paediatric Society. Age limits and adolescents. *Paediatr Child Health* 2003;8:577-8.
12. Chen W, Chang MH. New growth charts for Taiwanese children and adolescents based on World Health Organization standards and health-related physical fitness. *Pediatr Neonatol* 2010;51: 69-79.
13. Flynn JT, Kaelber DC, Baker-Smith CM, et al. Clinical practice guideline for screening and management of high blood pressure in children and adolescents. *Pediatrics* 2017;140:e20171904.
14. Bonora E, Formentini G, Calcaterra F, et al. HOMA-estimated insulin resistance is an independent predictor of cardiovascular

disease in type 2 diabetic subjects: prospective data from the Verona Diabetes Complications Study. *Diabetes Care* 2002;25: 1135-41.
15. Lang RM, Badano LP, Mor-Avi V, et al. Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *Eur Heart J Cardiovasc Imaging* 2015;16:233-70.
16. Iacobellis G, Willens HJ. Echocardiographic epicardial fat: a review of research and clinical applications. *J Am Soc Echocardiogr* 2009;22:1311-418.
17. Iacobellis G, Assael F, Ribaudo MC, et al. Epicardial fat from echocardiography: a new method for visceral adipose tissue prediction. *Obes Res* 2003;11:304-10.
18. Iacobellis G, Willens HJ, Barbaro G, Sharma AM. Threshold values of high-risk echocardiographic epicardial fat thickness. *Obesity (Silver Spring)* 2008;16:887-92.
19. Khoury PR, Mitsnefes M, Daniels SR, Kimball TR. Age-specific reference intervals for indexed left ventricular mass in children. *J Am Soc Echocardiogr* 2009;22:709-14.
20. Hietalampi H, Pahkala K, Jokinen E, et al. Left ventricular mass and geometry in adolescence: early childhood determinants. *Hypertension* 2012;60:1266-72.
21. Yang MC, Liu HK, Su YT, et al. Serum apoptotic marker M30 is positively correlated with early diastolic dysfunction in adolescent obesity. *PLoS One* 2019;14:e0217429.
22. Marchington JM, Pond CM. Site-specific properties of pericardial and epicardial adipose tissue: the effects of insulin and high-fat feeding on lipogenesis and the incorporation of fatty acids in vitro. *Int J Obes* 1990;14:1013-22.
23. Barber MC, Ward RJ, Richards SE, et al. Ovine adipose tissue monounsaturated fat content is correlated to depot-specific expression of the stearoyl-CoA desaturase gene. *J Anim Sci* 2000; 78:62-8.
24. Tomášová P, Čermáková M, Pelantová H, et al. Lipid profiling in epicardial and subcutaneous adipose tissue of patients with coronary artery disease. *J Proteome Res* 2020;19:3993-4003.
25. Kankaanpää M, Lehto HR, Pärkkä JP, et al. Myocardial triglyceride content and epicardial fat mass in human obesity: relationship to left ventricular function and serum free fatty acid levels. *J Clin Endocrinol Metab* 2006;91:4689-95.
26. Ahmed AM, Ragab SH, Ismail NA, et al. Echocardiographic assessment of epicardial adipose tissue in obese children and its relation to clinical parameters of metabolic syndrome. *J Saudi Heart Assoc* 2013;25:108.
27. Kocaman SA, Baysan O, Çetin M, et al. An increase in epicardial adipose tissue is strongly associated with carotid-intima media thickness and atherosclerotic plaque, but LDL only with the plaque. *Anatol J Cardiol* 2017;17:56-63.
28. Iacobellis G, Ribaudo MC, Zappaterreno A, et al. Relation between epicardial adipose tissue and left ventricular mass. *Am J Cardiol* 2004;94:1084-7.
29. Poirier P, Giles TD, Bray GA, et al. Obesity and cardiovascular disease: pathophysiology, evaluation, and effect of weight loss. *Arterioscler Thromb Vasc Biol* 2006;26:968-76.
30. Manzella D, Barbieri M, Rizzo MR, et al. Role of free fatty acids on cardiac autonomic nervous system in noninsulin-dependent diabetic patients: effects of metabolic control. *J Clin Endocrinol Metab* 2001;86:2769-74.
31. Sacks HS, Fain JN. Human epicardial adipose tissue: a review. *Am Heart J* 2007;153:907-17.
32. Mazurek T, Zhang L, Zalewski A, et al. Human epicardial adipose tissue is a source of inflammatory mediators. *Circulation* 2003; 108:2460-6.
33. Meléndez GC, McLarty JL, Levick SP, et al. Interleukin 6 mediates myocardial fibrosis, concentric hypertrophy, and diastolic dysfunction in rats. *Hypertension* 2010;56:225-31.
34. Akyol B, Boyraz M, Aysoy C. Relationship of epicardial adipose tissue thickness with early indicators of atherosclerosis and cardiac functional changes in obese adolescents with metabolic syndrome. *J Clin Res Pediatr Endocrinol* 2013;5:156-63.
35. Nerlekar N, Muthalaly RG, Wong N, et al. Association of volumetric epicardial adipose tissue quantification and cardiac structure and function. *J Am Heart Assoc* 2018;7:e009975.
36. Bakkum MJ, Danad I, Romijn MA, et al. The impact of obesity on the relationship between epicardial adipose tissue, left ventricular mass and coronary microvascular function. *Eur J Nucl Med Mol Imaging* 2015;42:1562-73.
37. Cavalcante JL, Tamarappoo BK, Hachamovitch R, et al. Association of epicardial fat, hypertension, subclinical coronary artery disease, and metabolic syndrome with left ventricular diastolic dysfunction. *Am J Cardiol* 2012;110:1793-8.
38. Fernando RR, Sayegh B, Syed MA, et al. Epicardial adipose tissue volume by cardiac magnetic resonance imaging predicts abnormal myocardial relaxation in patients with atrial fibrillation. *J Cardiovasc Magn Reson* 2015;17(Suppl 1):P352.
39. Doesch C, Streitner F, Bellm S, et al. Epicardial adipose tissue assessed by cardiac magnetic resonance imaging in patients with heart failure due to dilated cardiomyopathy. *Obesity (Silver Spring)* 2013;21:E253-61.
40. Doesch C, Haghi D, Flüchter S, et al. Epicardial adipose tissue in patients with heart failure. *J Cardiovasc Magn Reson* 2010;12:40.
41. Fontes-Carvalho R, Fontes-Oliveira M, Sampaio F, et al. Influence of epicardial and visceral fat on left ventricular diastolic and systolic functions in patients after myocardial infarction. *Am J Cardiol* 2014;114:1663-9.
42. Lai YH, Hou CJ, Yun CH, et al. The association among MDCT-derived three-dimensional visceral adiposities on cardiac diastology and dyssynchrony in asymptomatic population. *BMC Cardiovasc Disord* 2015;15:142.
43. Cavalcante JL, Tamarappoo BK, Hachamovitch R, et al. Association of epicardial fat, hypertension, subclinical coronary artery disease, and metabolic syndrome with left ventricular diastolic dysfunction. *Am J Cardiol* 2012;110:1793-8.
44. Kankaanpää M, Lehto HR, Pärkkä JP, et al. Myocardial triglyceride content and epicardial fat mass in human obesity: relationship to

left ventricular function and serum free fatty acid levels. *J Clin Endocrinol Metab* 2006;91:4689-95.
45. Sade LE, Eroglu S, Bozbaş H, et al. Relation between epicardial fat thickness and coronary flow reserve in women with chest pain and angiographically normal coronary arteries. *Atherosclerosis* 2009;204:580-5.
46. Yilmaz Y, Kurt R, Gurdal A, et al. Circulating vaspin levels and epicardial adipose tissue thickness are associated with impaired coronary flow reserve in patients with nonalcoholic fatty liver disease. *Atherosclerosis* 2011;217:125-9.
47. Mak GJ, Ledwidge MT, Watson CJ, et al. Natural history of markers of collagen turnover in patients with early diastolic dysfunction and impact of eplerenone. *J Am Coll Cardiol* 2009;54:1674-82.
48. Kapur NK. Transforming growth factor-β: governing the transition from inflammation to fibrosis in heart failure with preserved left ventricular function. *Circ Heart Fail* 2011;4:5-7.
49. Meléndez GC, McLarty JL, Levick SP, et al. Interleukin 6 mediates myocardial fibrosis, concentric hypertrophy, and diastolic dysfunction in rats. *Hypertension* 2010;56:225-31.