Atherosclerosis 290 (2019) 87–93

Contents lists available at ScienceDirect

Atherosclerosis

journal homepage: www.elsevier.com/locate/atherosclerosis

Visceral adipose tissue volume is associated with premature atherosclerosis in early type 2 diabetes mellitus independent of traditional risk factors

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HIGHLIGHTS

- T2DM patients with increased VAT volume are at risk for premature atherosclerosis.
- VAT volume is positively associated with arterial inflammation in early T2DM.
- Premature atherosclerosis was imaged with FDG and quantified as $_{mean}$TBR.
- The association of VAT with $_{mean}$TBR was independent of VAT dysfunction biomarkers.

ARTICLE INFO

Keywords:
Cardiovascular risk
Type 2 diabetes mellitus
Arterial inflammation
Abdominal visceral adipose tissue
FDG-PET

ABSTRACT

Background and aims: Type 2 diabetes mellitus (T2DM) is commonly associated with abdominal obesity, predominantly with high visceral adipose tissue (VAT), and is accompanied by premature atherosclerosis. However, the association between VAT and subcutaneous adipose tissue (SAT) with premature atherosclerosis and (i.e. arterial) inflammation is not completely understood. To provide more insight into this association, we investigated the association between arterial ^{18}F-fluordeoxyglucose (FDG) positron emission tomography (PET) uptake, as a measure of arterial inflammation, and metabolic syndrome (MetS) markers in early T2DM patients.
Methods: Forty-four patients with early T2DM, without glucose lowering medication, were studied (median age 63 [IQR 54–66] years, median BMI 30.4 [IQR 27.5–35.8]). Arterial inflammation was quantified using glucose corrected maximum standardized uptake value (SUV_{max}) FDG of the aorta, carotid, iliac, and femoral arteries, and corrected for background activity (blood pool) as target-to-background ratio ($_{mean}$TBR). VAT and SAT volumes (cm^3) were automatically segmented using computed tomography (CT) between levels L1-L5. Non-alcoholic fatty liver disease (NAFLD) was assessed by liver function test and CT.
Results: VAT volume, but not SAT volume, correlated with $_{mean}$TBR ($r = 0.325$, $p = 0.031$). Linear regression models showed a significant association, even after sequential adjustment for potentially influencing MetS components. Interaction term VAT volume * sex and additional components including HbA1c, insulin resistance, NAFLD, adiponectin, leptin, and C- reactive protein (CRP) did not change the independent association between VAT volume and $_{mean}$TBR.
Conclusions: CT-assessed VAT volume is positively associated with FDG-PET assessed arterial inflammation, independently of factors thought to potentially mediate these effects. These findings suggest that VAT in contrast to SAT is linked to early atherosclerotic changes in T2DM patients.

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https://doi.org/10.1016/j.atherosclerosis.2019.09.016
Received 7 May 2019; Received in revised form 27 August 2019; Accepted 25 September 2019
Available online 25 September 2019
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1. Introduction

Obesity is associated with an increased risk of developing type 2 diabetes mellitus (T2DM) and accelerates development of premature cardiovascular disease (CVD) [1]. Although T2DM is associated with a strongly increased CVD risk, the process of irreversible subclinical vascular damage already starts during the pre-diabetes stage [2]. Hyperglycaemia alone does not fully explain this risk, suggesting that other factors are likely to be involved.

Abdominal adipose tissue can be divided into visceral adipose tissue (VAT) and subcutaneous adipose tissue (SAT). Both are key contributors to abdominal obesity but their associated atherosclerotic risk profiles differ significantly [3,4]. VAT secretes pro-inflammatory (i.e. C-reactive protein, leptin) and anti-inflammatory (i.e. adiponectin) adipokines. The pro- inflammatory adipokines promote endothelial dysfunction, insulin resistance and, therefore, atherosclerosis [5]. In contrast, adiponectin is inversely associated with insulin resistance and non-alcoholic fatty liver disease (NAFLD), protecting from metabolic syndrome (MetS) and T2DM [5,6]. These MetS factors are supposed to be linked with increased cardiovascular risk in T2DM [7–9]. VAT volume assessed by computed tomography (CT) was shown to be associated with subclinical atherosclerosis assessed by whole body magnetic resonance angiography (MRI) [10]. Importantly, in non-obese populations, VAT volume appeared to be associated with 18F-fluoro- 2-deoxy-D-glucose positron emission tomography (FDG-PET)-assessed arterial inflammation [11], suggesting that VAT creates a pro-inflammatory environment even in the absence of MetS and T2DM. FDG-PET-assessed arterial inflammation is a marker of premature atherosclerosis [12] and predicts cardiovascular events independent of traditional risk factors in asymptomatic adults [13]. MetS, including HOMA-IR as a proxy for insulin resistance, seems an important determinant of arterial inflammation [12], potentially linking visceral adiposity with enhanced arterial inflammation in diabetes.

We hypothesize that VAT volume is an important determinant in the pathogenesis of arterial inflammation in patients with T2DM, independent of the classical risk factors. Therefore, we studied the association of CT-assessed abdominal VAT and SAT volumes with FDG-PET-assessed arterial inflammation in a homogenous obese population of T2DM patients without glucose lowering treatment and without history of CVD. Furthermore, we assessed the association with sex, additional MetS components (hypertension, dyslipidaemia, and HbA1c), HOMA-IR as an index of insulin resistance, liver function tests and CT-assessed NAFLD, the adipokine adiponectin, leptin, and CRP.

2. Materials and methods

2.1. Study design

This single centre, cross-sectional study included participants from a previously conducted study, the RELEASE trial [14,15]. This study was performed in compliance with the principles of the Declaration of Helsinki. All patients gave written informed consent. The protocol was reviewed and approved by the Institutional Review Board of the UMCG (number 2013-080).

2.2. Study population

Eligibility criteria were described in detail previously [14]. Briefly, potentially eligible early T2DM patients without glucose lowering drug treatment and aged between 30 and 70 years were included. T2DM was defined according to the criteria formulated by the American Diabetes Association. Exclusion criteria were current glucose-lowering drug use, uncontrolled hypertension (SBP > 160 mmHg or DBP > 100 mmHg), history of CVD defined as stable coronary artery disease or acute coronary syndrome, stroke or transient ischemic attack, or peripheral artery disease.

2.3. Clinical and laboratory assessments

The following clinical data were evaluated: age, sex, medical history, drug use, and cardiovascular history. Blood pressure, weight, height and waist circumference (WC) were also determined. BMI was calculated by dividing weight by height squared (kg/m^2). All blood samples were obtained in the morning after at least 8 h of overnight fasting, and plasma glucose, insulin, HbA1c, lipid profile, alanine aminotransferase (ALT), aspartate aminotransferase (AST), and gamma-glutamyl transferase (GGT) and CRP were measured using routine automated assays. Estimated Glomerular Filtration Rate (eGFR) was measured as marker of kidney function using CKD-EPI formula. Liver function tests ALT, AST, and GGT were measured as an indication of NAFLD. Total plasma adiponectin and leptin were determined with an Enzyme Linked Immuno Sorbent Assay (ELISA) kit (Linco Research, St Charles, Mo, USA; EZHADP-61K and EZHL-80SK). Insulin resistance was estimated with the HOMA-IR: fasting insulin * fasting glucose/22.5) [16].

2.4. FDG-PET/CT imaging and arterial FDG uptake

Patients fasted for a minimum of 6 h, and blood glucose concentrations were ensured to be less than 11 mmol/L before 3 MBq FDG/kg body weight was administered intravenously. All FDG-PET/CT scans were performed approximately 60 min after FDG administration on a Siemens Biograph 64 slice PET/CT scanner (Siemens Medical Systems, Knoxville, Tn, USA) according to the European Association of Nuclear Medicine (EANM) procedure guidelines for FDG imaging [17]. Low-dose CT was performed before the PET-emission for attenuation correction and anatomic mapping with 100 kV and 30 mAs. PET emission data were acquired from the skull to knee, 3 min per bed position.

Arterial inflammation was quantified as the maximum standardized

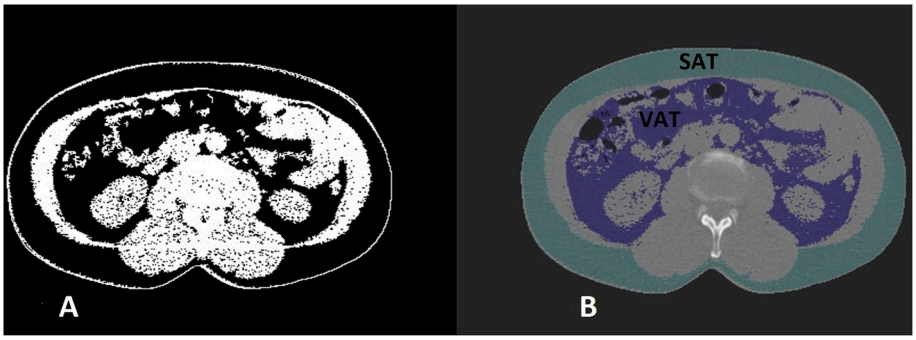

Fig. 1. Analysis of abdominal adipose tissue using semi-automated method [18].
(A) Closed muscle layer separating visceral adipose tissue (VAT) and subcutaneous adipose tissue (SAT). (B) Differentiated adipose tissue VAT (purple colored) and SAT (green colored).

Table 1
Characteristics of the total early type 2 diabetes mellitus population.

	Total population (N = 44)
Male (%)	27 (61%)
Age (years)	63 (54-66)
T2DM duration (years)	1 (0-3.8)
BMI (kg/m^2)	30.4 (27.5–35.8)
Manually measured waist circumference (cm)	101 (94-108)
Systolic blood pressure (mmHg)	137 (127-147)
Estimated glomerular filtration rate (mL/min/1.73 m^2)	85 ± 14
HbA1c (%)	6.3 ± 0.43
HOMA-IR	5.1 (3.1–8.5)
Total cholesterol (mmol/L)	4.8 ± 0.95
HDL (mmol/L)	1.3 (1.1–1.5)
LDL (mmol/L)	3.1 ± 1.0
Triglycerides (mmol/L)	1.4 (0.91–2.0)
Alanine transaminase (U/L)	26 (20-44)
Aspartate transaminase (U/L)	26 (22-33)
Gamma-glutamyl transferase (U/L)	34.5 (25.0–46.5)
Computed tomography-assessed non-alcoholic fatty liver disease (%)	17 (39%)
Glucose (mmol/l)	7.4 ± 0.97[a]
Adiponectin (ng/ml)	7.9 (6.3–10.5)
Leptin (ng/ml)	13.2 (6.4–25.6)
C-reactive protein (mg/l)	1.2 (0.70–3.1)
Visceral adipose tissue volume (cm^3)	7.97 (6.55–10.58)
Subcutaneous adipose tissue volume (cm^3)	8.86 (5.57–13.31)

Data are presented as mean ± SD (when normally distributed) or as median with IQR (when not normally distributed). [a]N = 43.

FDG uptake value (SUV_{max}) in the arteries as described earlier [14]. In short, SUV_{max} was calculated by dividing radioactive concentration (kBq/mL) by (activity injected (kBq)/weight (g)). The SUV_{max} in arteries was measured by manually drawing three-dimensional Volumes of Interest (VOIs). VOIs were separately drawn around the carotid arteries, ascending aorta and aortic arch, descending and abdominal aorta, and iliac and femoral arteries and adjusted for the pre-scan glucose value and background activity by the caval veins blood pool activity, yielding a mean arterial target-to- background ratio ($_{mean}$TBR).

2.5. Adipose tissue volume analysis

All measurements were performed using MATLAB software (version R2015b The MathWorks, Inc, Natick, MA, USA) as previously described [18]. In order to analyse the entire abdomen, all slices from vertebral levels L1 to L5 were manually selected. Adipose tissue was initially segmented by thresholding the CT images between −174 and −24 Hounsfield Units (HU) [18]. Adipose tissue volumes were determined using a semi-automated method [18]. The abdominal muscular layer was used as a boundary to separate VAT from SAT, see Fig. 1.

2.6. CT-assessed NAFLD analyses

Fatty liver analyses were performed on CT as a proxy for NAFLD. These analyses were done with the method of Zeb et al. [19], based on CT images. In brief, ratio of liver-to-spleen HU was used to assess the severity of NAFLD. HU decreases in the development of NAFLD. Ratio was calculated as the mean of three regions of interest of mean HU value of the liver, divided by the mean HU of one region of interest of the spleen. HU < 1.0 was used to diagnose NAFLD. The measurements of liver and spleen HU attenuations were shown to be highly reproducible in this previous study.

2.7. Statistical analysis

Discrete variables are presented as numbers with percentages. Quantitative variables with a normal distribution are presented as mean ± SD and not normally distributed as median with interquartile range (IQR). All statistical analyses were performed with IBM Statistical Package for Social Sciences (SPSS) version 23. Independent *t*-test in normally distributed variables, a Mann-Whitney *U* test in not-normally

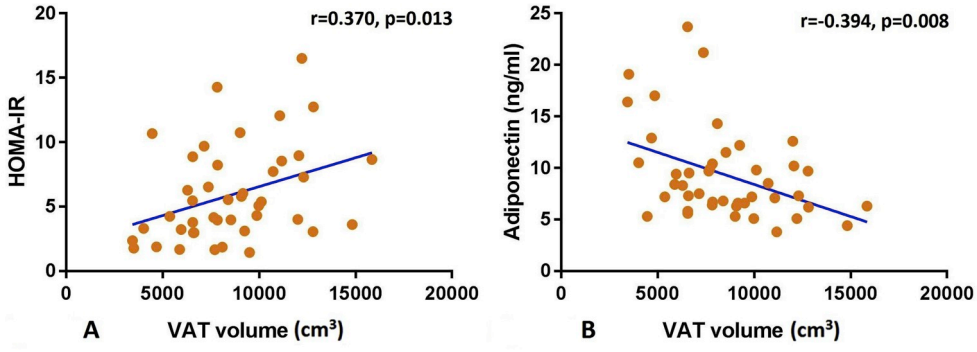

Fig. 2. Relation of visceral adipose tissue volume (cm^3) with HOMA insulin resistance (A) and adiponectin (ng/ml) (B).

distributed variables and a one-way ANOVA in categorical variables were used to compare groups. Univariate associations with VAT and SAT volumes were assessed and a possible relation between $_{mean}$TBR and potential confounders was investigated with Spearman's correlation coefficient (R).

Multiple linear regressions were used to examine factors associated with the $_{mean}$TBR and to assess whether VAT volume was associated with $_{mean}$TBR independently of anticipated factors. Five models were constructed to sequentially evaluate the addition of MetS on the association of VAT volume with $_{mean}$TBR (Model 1). Because of sex differences in adipose tissue distribution, the interaction of sex vs. VAT volume was tested and all models were corrected for sex as well as for the interaction term of VAT volume * sex. After including VAT volume, sex, and the interaction term (Model 2), components of MetS were added sequentially: HbA1c, or BMI, or triglycerides and HDLc, or SBP and DBP separately (Model 3), HOMA-IR and ALT (Model 4), HOMA-IR and CT-assessed NAFLD (Model 5), and adiponectin, leptin, and CRP (Model 6). $p < 0.05$ was considered statistically significant.

3. Results

3.1. Patient characteristics

In total, 44 early T2DM patients participated in the study, of which 27 males and 17 females. Clinical characteristics are presented in Table 1.

VAT volume was associated with BMI, waist circumference, eGFR, ALT, AST, GGT, HOMA- IR, adiponectin, and leptin. Relation between VAT volume with HOMA-IR and adiponectin is shown in Fig. 2. VAT and SAT volumes were not interrelated. No associations of VAT volume with MetS components (lipids, blood pressure, and HbA1c) or CRP were found, as shown in Table 2. Additionally, SAT volume did not correlate with liver function tests and CT-assessed NAFLD, neither with lipids, blood pressure, and HbA1c. However, SAT volume was associated with leptin levels (r = 0.817, $p < 0.001$).

3.2. Association of VAT and SAT volume with arterial FDG uptake

VAT volume (r = 0.348, $p = 0.022$) correlated significantly with $_{mean}$TBR, see Fig. 3. In contrast, SAT volume did not correlate significantly with $_{mean}$TBR (r = 0.045, $p = 0.773$). $_{mean}$TBR showed to be significantly associated with HbA1c (r = 0.434, $p = 0.003$) and systolic blood pressure (r = 0.338, $p = 0.029$) as shown in Table 2.

The association between VAT volume and $_{mean}$TBR was modified by sex (interaction VAT volume * sex $p = 0.026$). Therefore, we included this interaction variable in the multiple linear regression models. Furthermore, adding MetS components BMI, SBP and DBP, and triglycerides and HDLc separately, did not alter the association between VAT volume and $_{mean}$TBR (data not shown). All models showed that VAT volume remained significantly associated with $_{mean}$TBR, as shown in Table 3. While HbA1c was positively associated with the association between $_{mean}$TBR and VAT volume, the models with HOMA-IR and NAFLD (ALT or CT-assessed NAFLD) or adiponectin, leptin and C-reactive protein were not.

4. Discussion

In this cross-sectional study, we demonstrated that VAT volume, contrary to SAT volume, was associated with arterial FDG-PET as a marker of arterial inflammation in early T2DM. This association remained present even after adjustment for sex, HbA1c, insulin resistance (HOMA- IR) and NAFLD (ALT and CT-assessed NAFLD), and levels of adipokines (adiponectin, leptin, and C-reactive protein). These findings confirm previous reports that VAT is linked with arterial inflammation, which is considered an early step in the atherosclerosis process. This process possibly results in premature atherosclerosis as observed in T2DM, even early in the course of the disease with only mild hyperglycaemia. We found a significant correlation of arterial inflammation with well-known components of MetS. Sex did play an important role in this association. Therefore, we included interaction variable VAT volume * Sex. The correlation showed to be independent of sex. Moroever, the MetS factors did not influence the association of VAT with arterial inflammation in the current study, suggesting that other unknown factors may mediate the development of premature atherosclerosis.

To the best of our knowledge, this is the first study in an early T2DM cohort to analyse the effect of adipose tissue volumes on arterial FDG uptake. We differentiated between VAT and SAT in relation to arterial

Table 2
Relation of VAT volume and $_{mean}$TBR with T2DM associated risk factors.

	VAT volume		$_{mean}$TBR	
	Correlation coefficient r	*p*-value	Correlation coefficient r	*p*-value
BMI	0.47	**0.001**	0.30	0.051
Waist circumference	0.64	< **0.001**	0.22	0.159
Systolic blood pressure	0.90	0.569	0.34	**0.029**
Estimated glomerular filtration rate	0.37	**0.013**	0.25	0.109
HbA1c	0.19	0.207	0.43	**0.003**
HOMA-IR	0.37	**0.015**	0.10	0.506
Total cholesterol	−0.11	0.476	0.10	0.526
HDL	−0.17	0.272	−0.21	0.171
LDL	−0.14	0.385	0.09	0.579
Triglycerides	0.27	0.077	0.23	0.134
Alanine transaminase	0.57	**0.000**	0.28	0.071
Aspartate transaminase	0.42	**0.005**	0.53	0.098
Gamma-glutamyl transferase	0.33	**0.028**	0.17	0.263
Adiponectin	−0.39	**0.008**	0.085	0.584
Leptin	0.08	0.631	−0.064	0.680
C-reactive protein	−0.023	0.880	−0.23	0.137
Visceral adipose tissue volume			0.33	**0.031**
Subcutaneous adipose tissue volume	0.58	0.400	0.041	0.793

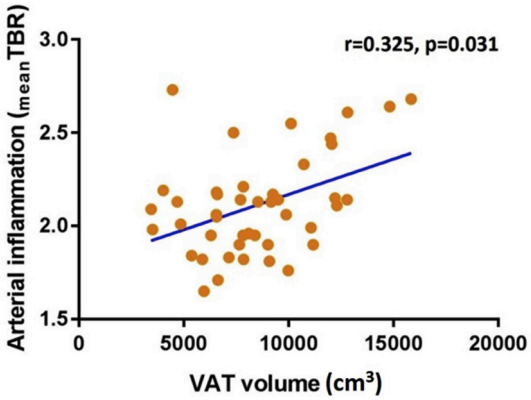

Fig. 3. Relation of visceral adipose tissue (VAT) volume (cm^3) to arterial FDG uptake ($_{mean}$TBR).

FDG uptake and studied the effect of pathophysiological confounders potentially mediating the association between VAT and arterial inflammation. This study, therefore, improves and refines the understanding of the relation between adiposity and cardiovascular risk in T2DM, although the precise underlying mechanism of adiposity-enhanced arterial inflammation needs to be identified.

Arterial inflammation in relation with VAT volume was studied before [20,21]. However, the study population differed from our study. While we studied a prospectively selected, well defined, homogeneous, high risk cohort with obesity and early T2DM with a median BMI of 30.4, Hong et al. scanned healthy check-up subjects with a median BMI of 24.8. Figueroa et al. scanned patients who came for oncological evaluation with a median BMI of 26.4. Our study therefore expanded the results of these studies in the target population. Nevertheless, a positive association of waist circumference with carotid FDG uptake has been found in patients with MetS [12]. In the study of Tahara et al., abdominal waist circumference was studied as a proxy for VAT. When in fact, this measure also includes SAT. Our results now show that VAT volume is indeed an independent factor associated with arterial inflammation, whereas SAT is not. Furthermore, waist circumference in our study did not correlate with arterial inflammation. These results are in line with the concept that VAT is known to produce more pro-inflammatory adipokines compared to SAT [4]. Furthermore, VAT is associated with CVD risk factors [8,22] and future CVD events, even after adjustment for CVD risk factors and generalized adiposity [23,24]. Our study not only confirms these studies, but also clarifies that these effects also occur at a very early disease stage.

Several studies investigated VAT in relation to proxies of subclinical atherosclerosis. A positive relation was found between VAT with ultrasound assessed common carotid intima- media thickness [25,26] and coronary and abdominal aortic calcium [27]. Different technical methods were used for measuring VAT, including ultrasonography [25], MRI [26], and CT [27]. In contrast to these methods which visualize later (irreversible) stages, we used FDG-PET for measuring one of the earliest stages of atherosclerosis that can be assessed *in vivo*. This method is intended to detect the most premature stages of atherosclerosis, when there are still no detectable structural changes. We previously demonstrated that *ex vivo* FDG-uptake in carotid plaques strongly correlates with macrophage plaque infiltration [28]. Furthermore, FDG-PET assessed arterial inflammation is strongly associated with pulse wave velocity, which is considered a clinical proxy for endothelial dysfunction in early T2DM [14]. Although arterial inflammation was measured with FDG-PET before in the carotid artery [12,20] and ascending aorta [21], our study extends these findings as we measured FDG-uptake in ten different artery segments within the individual patients, thereby providing a more accurate estimation of the total body arterial inflammation.

The underlying mechanism of VAT leading to atherosclerosis is incompletely understood. In a previous study, MetS was shown to be an important determinant of arterial inflammation [12]. Remarkably, we did not find significant associations between VAT volume and MetS components lipids, systolic blood pressure and HbA1c. This could potentially be due to the use of statins, blood pressure lowering drugs and HbA1c levels. HbA1c in our study did not correlate with VAT. But HbA1c was relatively low in this treatment naive T2DM population. Importantly, a clear association with insulin resistance was indeed observed. Furthermore, VAT volume proved to be associated with other biomarkers of VAT dysfunction, i.e. total plasma adiponectin, liver function tests and CT-assessed NAFLD. In this study, we also investigated the effect of several plausible pathophysiological confounders, which has not been studied before. Influence of insulin resistance, NAFLD, and adipokines on the association between arterial inflammation and VAT volume was not investigated before [12,20,21]. Moreover, in previous studies, results were not corrected for sex, while sex is a key factor in adipose tissue distribution [29]. Although we can only speculate, our result suggests that other factors play a role when VAT volume increases. Unassessed cytokines, such as TNF-α, which are secreted by adipose tissue after macrophage recruitment, oxidative stress, or hormonal factors could play a role [30]. The underlying mechanism of the influence of adipose tissue in the development of atherosclerosis still needs further investigation.

Another factor that could possibly stimulate atherosclerosis in T2DM is leptin. Although leptin reduces appetite [31], obese individuals show resistance to leptin, which results in elevated levels failing to control appetite and therefore, weight gain [32]. The exact effect of leptin in T2DM is unclear; it could be either a cause or effect of adipose tissue volume. In our study, we found a significant association between SAT volume and leptin levels. However, leptin did not influence the association between VAT volume and arterial inflammation.

This study has some limitations. First, the relative small observational cohort limits the power in our study. Second, almost 55% of the patients already used a statin. Statins affect the arterial inflammation [33] and lipid levels. However, patients were only included if they had been on a stable dose of statins. Our main associations might be an underestimation of the true effect, at least in statin users. Third, we performed a low dose (ld) CT with lower resolutions compared to diagnostic CT scans. The quality of the ldCT scans was appropriate to differentiate adipose tissue from surrounding tissues and the radiation dose is much less compared to diagnostic CT scans. Furthermore, the value of HOMA-IR is not uniform and depends on many factors including demography, BMI, age and sex. HOMA-IR is a value for whole body insulin resistance whereas insulin resistance could vary substantially between different organs and tissues. HOMA-IR is commonly used as a marker of insulin resistance and is the most accurate assessment of insulin resistance at this moment.

In conclusion, CT-assessed VAT volume is associated with FDG-PET assessed arterial inflammation as marker of premature atherosclerosis in early T2DM. This association is independent of sex and traditional MetS risk factors in T2DM patients, including pathophysiologically relevant biomarkers for VAT dysfunction. More insight in this association may help to identify T2DM patients at particularly high risk of CVD, allowing early interventions.

Trial registry number

NCT02015299 (clinicaltrials.gov).

Financial support

This study was supported by Boehringer Ingelheim (Alkmaar, the Netherlands). Boehringer Ingelheim was not involved in the design and results of the study; collection, management, analysis and

Table 3
Arterial inflammation and VAT volume: multivariate linear regression models.

	Model 1 (R^2 = 0.177, p = 0.004)		Model 2 (R^2 = 0.311, p = 0.002)		Model 3 (R^2 = 0.532, p < 0.001)		Model 4 (R^2 = 0.540, p < 0.001)		Model 5 (R^2 = 0.500, p < 0.001)		Model 6 (R^2 = 0.590, p < 0.001)	
Dependent: $_{mean}$target to background ratio	St. β	*p*-value	St. β	*p*-value	St. β	*p*-value	St. β	*p*-value	St. β	*p*-value	St. β	*p*-value
VAT volume	0.42	**0.004**	1.56	**0.002**	1.30	**0.002**	1.34	**0.002**	1.34	**0.003**	1.36	**0.002**
Sex			1.21	**0.011**	0.97	**0.015**	1.06	**0.014**	1.00	**0.023**	1.08	**0.007**
Visceral adipose tissue volume * sex			- 1.10	**0.020**	- 0.92	**0.022**	- 1.03	**0.02**	- 0.99	**0.029**	- 0.81	**0.046**
HbA1c					0.48	< **0.001**	0.47	< **0.001**	0.43	**0.001**	0.47	< **0.001**
HOMA-IR							0.11	0.452	- 0.16	0.417		
Alanine transaminase							- 0.04	0.077				
CT-assessed NAFLD									- 0.17	0.342		
Adiponectin											0.14	0.274
Leptin											- 0.23	0.229
C-reactive protein											- 0.05	0.694

interpretation of data, writing of the report or the decision to submit the paper for publication.

Author contributions

M.R. collected, analysed, interpreted data, and wrote the manuscript, S.A.d.B. collected, analysed, interpreted data, conceived and designed the study and reviewed the manuscript critically for intellectual content, D.S.S. designed the technical method and reviewed the manuscript critically for intellectual content, J.D.L. conceived and designed the study and reviewed the manuscript critically for intellectual content, H.J.L.H. analysed, interpreted data, conceived and designed the study and reviewed the manuscript critically for intellectual content, R.B. analysed, interpreted data and reviewed the manuscript critically for intellectual content, M.J.W.G. validated CT-assessed NAFLD analyses and designed the study and reviewed the manuscript critically for intellectual content, J.L.H. conceived and designed the study and reviewed the manuscript critically for intellectual content, R.J.H.B. analysed, interpreted data and reviewed the manuscript critically for intellectual content, R.H.J.A.S. analysed data, validated CT-assessed NAFLD analyses, conceived and designed the study and reviewed the manuscript critically for intellectual content, D.J.M. collected, analysed, interpreted data, conceived and designed the study and reviewed the manuscript critically for intellectual content. M.R. and D.J.M. are the guarantors of this work and, as such, had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. All authors approved the final version.

Declaration of competing interest

The authors declared they do not have anything to disclose regarding conflict of interest with respect to this manuscript.

Acknowledgements

We wish to thank the study participants, the general practitioner practices, and MG Piersma- Wichers and HL Lutgers from CERTE Groningen.

References

[1] A.E. Field, E.H. Coakley, A. Must, J.L. Spadano, N. Laird, W.H. Dietz, et al., Impact of overweight on the risk of developing common chronic diseases during a 10-year period, Arch. Intern. Med. 161 (13) (2001) 1581–1586 Jul 9.
[2] J.C. Pickup, Inflammation and activated innate immunity in the pathogenesis of type 2 diabetes, Diabetes Care 27 (3) (2004) 813–823 Mar.
[3] S.S. Dhaliwal, T.A. Welborn, Central obesity and multivariable cardiovascular risk as assessed by the Framingham prediction scores, Am. J. Cardiol. 103 (10) (2009) 1403–1407 May 15.
[4] C.S. Fox, J.M. Massaro, U. Hoffmann, K.M. Pou, P. Maurovich-Horvat, C.Y. Liu, et al., Abdominal visceral and subcutaneous adipose tissue compartments: association with metabolic risk factors in the Framingham Heart Study, Circulation 116 (1) (2007) 39–48 Jul 3.
[5] D.C. Lau, B. Dhillon, H. Yan, P.E. Szmitko, S. Verma, Adipokines: molecular links between obesity and atheroslcerosis, Am. J. Physiol. Heart Circ. Physiol. 288 (5) (2005) 2031 May.
[6] S.A. Polyzos, K.A. Toulis, D.G. Goulis, C. Zavos, J. Kountouras, Serum total adiponectin in nonalcoholic fatty liver disease: a systematic review and meta-analysis, Metabolism 60 (3) (2011) 313–326 Mar.
[7] H.W. Kwon, S.M. Lee, J.W. Lee, J.E. Oh, S.W. Lee, S.Y. Kim, Association between volume and glucose metabolism of abdominal adipose tissue in healthy population, Obes. Res. Clin. Pract. (2017) Jan 7.
[8] J. Liu, C.S. Fox, D.A. Hickson, W.D. May, K.G. Hairston, J.J. Carr, et al., Impact of abdominal visceral and subcutaneous adipose tissue on cardiometabolic risk factors: the Jackson heart study, J. Clin. Endocrinol. Metab. 95 (12) (2010) 5419–5426 Dec.
[9] O. Schnell, L. Ryden, E. Standl, A. Ceriello, D&CVD EASD Study Group, Current perspectives on cardiovascular outcome trials in diabetes, Cardiovasc. Diabetol. 15 (1) (2016) 139 Oct 1.
[10] T. Hansen, H. Ahlstrom, S. Soderberg, J. Hulthe, J. Wikstrom, L. Lind, et al., Visceral adipose tissue, adiponectin levels and insulin resistance are related to atherosclerosis as assessed by whole-body magnetic resonance angiography in an elderly population, Atherosclerosis 205 (1) (2009) 163–167 Jul.

[11] C.S. Hung, J.K. Lee, C.Y. Yang, H.R. Hsieh, W.Y. Ma, M.S. Lin, et al., Measurement of visceral fat: should we include retroperitoneal fat? PLoS One 9 (11) (2014) e112355Nov 17.
[12] N. Tahara, H. Kai, S. Yamagishi, M. Mizoguchi, H. Nakaura, M. Ishibashi, et al., Vascular inflammation evaluated by [18F]-fluorodeoxyglucose positron emission tomography is associated with the metabolic syndrome, J. Am. Coll. Cardiol. 49 (14) (2007) 1533–1539. Apr 10.
[13] S.H. Moon, Y.S. Cho, T.S. Noh, J.Y. Choi, B.T. Kim, K.H. Lee, Carotid FDG uptake improves prediction of future cardiovascular events in asymptomatic individuals, JACC Cardiovasc. Imaging 8 (8) (2015) 949–956. Aug.
[14] S.A. de Boer, M.C. Hovinga-de Boer, H.J. Heerspink, J.D. Lefrandt, A.M. van Roon, H.L. Lutgers, et al., Arterial stiffness is positively associated with 18F-fluorodeoxyglucose positron emission tomography-assessed subclinical vascular inflammation in people with early type 2 diabetes, Diabetes Care 39 (8) (2016) 1440–1447. Aug.
[15] S.A. de Boer, H.J. Heerspink, L.E. Juarez Orozco, A.M. van Roon, P.W. Kamphuisen, A.J. Smit, et al., Effect of linagliptin on pulse wave velocity in early type 2 diabetes: a randomized, double-blind, controlled 26-week trial (RELEASE), Diabetes Obes. Metab. (2017) Feb 28.
[16] D.R. Matthews, J.P. Hosker, A.S. Rudenski, B.A. Naylor, D.F. Treacher, R.C. Turner, Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man, Diabetologia 28 (7) (1985) 412 Jul.
[17] R. Boellaard, R. Delgado-Bolton, W.J. Oyen, F. Giammarile, K. Tatsch, W. Eschner, et al., FDG PET/CT: EANM procedure guidelines for tumour imaging: version 2.0, Eur. J. Nucl. Med. Mol. Imaging 42 (2) (2015) 328–354 Feb.
[18] S.A. de Boer, D.S. Spoor, R.H.J.A. Slart, D.J. Mulder, M. Reijrink, R.J.H. Borra, et al., Performance evaluation of a semi-automated method for [(18)F]FDG uptake in abdominal visceral adipose tissue, Mol. Imaging Biol. (2018) May 22.
[19] I. Zeb, D. Li, K. Nasir, R. Katz, V.N. Larijani, M.J. Budoff, Computed tomography scans in the evaluation of fatty liver disease in a population based study: the multi-ethnic study of atherosclerosis, Acad. Radiol. 19 (7) (2012) 811–818 Jul.
[20] H.C. Hong, S.Y. Hwang, S. Park, J.Y. Ryu, H.Y. Choi, H.J. Yoo, et al., Implications of pericardial, visceral and subcutaneous adipose tissue on vascular inflammation measured using 18FDG- PET/CT, PLoS One 10 (8) (2015) e0135294Aug 13.
[21] A.L. Figueroa, R.A. Takx, M.H. MacNabb, A. Abdelbaky, Z.R. Lavender, R.S. Kaplan, et al., Relationship between measures of adiposity, arterial inflammation, and subsequent cardiovascular events, Circ Cardiovasc. Imaging 9 (4) (2016) e004043Apr.
[22] H. He, Y. Ni, J. Chen, Z. Zhao, J. Zhong, D. Liu, et al., Sex difference in cardiometabolic risk profile and adiponectin expression in subjects with visceral fat obesity, Transl. Res. 155 (2) (2010) 71–77 Feb.
[23] R. Ladeiras-Lopes, F. Sampaio, N. Bettencourt, R. Fontes-Carvalho, N. Ferreira, A. Leite-Moreira, et al., The ratio between visceral and subcutaneous abdominal fat assessed by computed tomography is an independent predictor of mortality and cardiac events, Rev. Esp. Cardiol. (2016) Oct 17.
[24] K.A. Britton, J.M. Massaro, J.M. Murabito, B.E. Kreger, U. Hoffmann, C.S. Fox, Body fat distribution, incident cardiovascular disease, cancer, and all-cause mortality, J. Am. Coll. Cardiol. 62 (10) (2013) 921–925 Sep 3.
[25] L.S. Rallidis, K. Baroutsi, M. Zolindaki, M. Karagianni, C. Varounis, N. Dagres, et al., Visceral adipose tissue is a better predictor of subclinical carotid atherosclerosis compared with waist circumference, Ultrasound Med. Biol. 40 (6) (2014) 1083–1088 Jun.
[26] K.B. Gast, M. den Heijer, J.W. Smit, R.L. Widya, H.J. Lamb, A. de Roos, et al., Individual contributions of visceral fat and total body fat to subclinical atherosclerosis: the NEO study, Atherosclerosis 241 (2) (2015) 547–554. Aug.
[27] N.J. Alvey, A. Pedley, K.J. Rosenquist, J.M. Massaro, C.J. O'Donnell, U. Hoffmann, et al., Association of fat density with subclinical atherosclerosis, J. Am. Heart Assoc. 3 (4) (2014), https://doi.org/10.1161/JAHA.114.000788 Aug 28.
[28] M.G. Masteling, C.J. Zeebregts, R.A. Tio, J.C. Breek, U.J. Tietge, J.F. de Boer, et al., High-resolution imaging of human atherosclerotic carotid plaques with micro 18F-FDG PET scanning exploring plaque vulnerability, J. Nucl. Cardiol. 18 (6) (2011) 1066–1075 Dec.
[29] K. Karastergiou, S.K. Fried, Cellular mechanisms driving sex differences in adipose tissue biology and body shape in humans and mouse models, Adv. Exp. Med. Biol. 1043 (2017) 29–51.
[30] L.F. Van Gaal, I.L. Mertens, C.E. De Block, Mechanisms linking obesity with cardiovascular disease, Nature 444 (7121) (2006) 875–880 Dec 14.
[31] O.M. Farr, C. Fiorenza, P. Papageorgiou, M. Brinkoetter, F. Ziemke, B.B. Koo, et al., Leptin therapy alters appetite and neural responses to food stimuli in brain areas of leptin-sensitive subjects without altering brain structure, J. Clin. Endocrinol. Metab. 99 (12) (2014) 2529 Dec.
[32] P. Singh, T.E. Peterson, F.H. Sert-Kuniyoshi, J.A. Glenn, D.E. Davison, A. Romero-Corral, et al., Leptin signaling in adipose tissue: role in lipid accumulation and weight gain, Circ. Res. 111 (5) (2012) 599–603. Aug 17.
[33] N. Tahara, H. Kai, M. Ishibashi, H. Nakaura, H. Kaida, K. Baba, et al., Simvastatin attenuates plaque inflammation: evaluation by fluorodeoxyglucose positron emission tomography, J. Am. Coll. Cardiol. 48 (9) (2006) 1825–1831 Nov 7.