Relationship between visceral adipose tissue and triglyceride glucose index with liver status in men and women with metabolic syndrome features: sex differences

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Research

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Abstract

Background

Metabolic syndrome (MetS) comprises several clinical and metabolic risk factors that increase the risk to develop cardiovascular disease and other comorbidities. Meanwhile, ageing and biological differences between men and women play an important role in body fat distribution and health status. The current study aimed to evaluate the association of visceral adipose tissue (VAT) and the triglyceride and glucose index (TyG) with metabolic and liver risk factors among subjects diagnosed with MetS with emphasis on differences attributable to sex.

Methods

A cross-sectional study was performed including 326 individuals with MetS (54–75 years) from the PREDIMED-Plus study. Liver markers, visceral fat (VAT) and the triglyceride glucose index (TyG) were assessed. Participants were stratified according to VAT and TyG tertiles. Receiver operating characteristic (ROC) curve was used to analyze the efficiency of TyG for VAT.

Results

Subjects with greater visceral fat depots showed worse lipid profile, higher HOMA-R, TyG, ALT, fibroblast growth factor-21 (FGF-21), fatty liver index (FLI) and hepatic steatosis index (HSI) compared to participants in the 1st tertile. The multi-adjusted linear regression analyses indicated that individuals in the third tertile of TyG (> 9.1–10.7 units) had a positive association with HOMA-IR (β = 3.07 [95% CI, 2.28–3.86]; p-trend < 0.001), ALT (β = 7.43 [95%CI, 2.23–12.63]; p-trend = 0.005), GGT (β = 14.12 [95% CI, 3.64–24.61; p-trend = 0.008]), FGF-21 (β = 190.69 [95% CI, 93.13-288.25; p-trend < 0.001]), FLI (β = 18.65 [95%CI, 14.97–22.23]; p-trend < 0.001) and HSI (β = 3.46 [95% CI, 2.23–4.68; p-trend < 0.001]) versus participants from the first tertile. Interestingly, the TyG showed the largest AUC for women (AUC = 0.713; 95%CI, 0.62–0.79) compared to men (AUC = 0.570; 95%CI, 0.48–0.66), despite the differences in VAT content associated to sex.

Conclusions

Both, VAT and the TyG are strongly associated with cardiometabolic and liver risk factors linked with non-alcoholic fatty liver disease in men and women diagnosed with MetS. The TyG could be used as suitable marker estimator of VAT in women.

Trial registration

Retrospectively registered at the International Standard Randomized Controlled Trial (http://www.isrctn.com/ISRCTN89898870), registration date: 24 July 2014.

Introduction

The metabolic syndrome (MetS) encompasses a cluster of cardiometabolic features like impaired glucose metabolism, dyslipidaemia, abdominal obesity, and elevated blood pressure [1]. The strong association between MetS and an increased risk of cardiovascular disease (CVD) as well as all-cause mortality is well documented [2, 3]. Ageing and sex differences should be considered as well since it is a major contributor to the prevalence of cardiovascular and metabolic risk factors that constitute the syndrome [1, 4]. On the other hand, non-alcoholic fatty liver disease (NAFLD) is recognized as the hepatic manifestation of MetS [5]. NAFLD is a highly prevalent chronic liver illness, whose incidence linearly increases with body mass index (BMI) and adiposity [6]. This condition is quite common in obese individuals with central adiposity [7, 8]. The distribution of adipose tissue is of great importance since abdominal obesity is a key factor in the development of the MetS [9] and NAFLD [10] when differences between men and women are usually reported [11, 12]. Insulin resistance is considered the primary triggering mechanism for the development of diabetes, NAFLD and MetS when fat accumulates in intra-abdominal depots [9, 10] Thus, body fat distribution in older adults and the differences between men and women is critical for determining how susceptible they are or will be to developing NAFLD and/or other CVD [11, 13–15] being partly attributed to sex differences in fat content [11, 15]. In ageing, adipose tissues exhibited changes in quantity, distribution, hormone production, inflammatory status, and others. [11]. In this context, consistent evidence suggests that sex hormones play a major role in the activity and fat distribution, which has differential harm effects on metabolic status in male and female [11, 13]. Central obesity is often quantified using waist circumference. But, it can be confounded by varying levels of subcutaneous fat in the waist, and may not accurately reflect visceral fat in all individuals [16]. The dual-energy X-ray absorptiometry (DXA) with CoreScan is a practical and valuable tool to assess visceral fat mass [17]. Nevertheless, the DXA equipment is expensive and might not be easy to access. In this sense, the identification of non-invasive markers able to discriminate subjects with higher visceral adiposity and higher susceptibility to develop NAFLD would be of great interest. Indeed, liver biopsy is the gold standard for the NAFLD diagnosis [7], but it is an invasive technique not suitable for routine screening and monitoring [7]. Several non-invasive markers related to liver status and insulin resistance have been proposed to characterize NAFLD [7, 18–20]. A novel potential marker is the triglyceride glucose index (TyG), which has demonstrated a better predictive value compared to fasting plasma glucose (FPG) for the risk of type 2 diabetes in normoglycemic individuals as well as to be related with insulin resistance [21]. In the present study, we hypothesized that the TyG index could be a good determinant in men and women with higher VAT and thus higher susceptibility to develop NAFLD. Therefore, the objective of the present work was to analyze the potentially detrimental effect of VAT accumulation on metabolic status and to assess the potential association of the TyG index with VAT, metabolic risk factors, NAFLD scores and serum markers in overweight/obese men and women with MetS and the differences between both sexes.

Methods

Study population and design

This research is a cross-sectional study concerning baseline data from al participant of the Navarra-Nutrition centre within the PREDIMED-Plus trial (ISRCTN89898870) (http://www.isrctn.com/ISRCTN89898870). PREDIMED-Plus is a multicenter, parallel-group, randomized trial carried out in Spain, aiming to evaluate the effectiveness of an energy-restricted traditional Mediterranean diet, physical activity promotion, and behavioural support (intervention group) on the primary prevention of CVD, in comparison with general advised energy-unrestricted Mediterranean diet (control group). Detailed methods and protocols of the study have been published previously [22,23]. In brief, 6874 individuals were recruited in 23 Spanish centres. Eligible participants were men (55–75 years) and women (60–75 years) with a BMI ≥27 and <40 kg/m^2 and fulfilling at least three criteria for the MetS: waist circumference (WC) in Caucasian people ≥102 cm for men and ≥ 88 cm for women, elevated triglycerides levels ≥150 mg/dL or drug treatment for hyperlipidemia; reduced HDL-c <40 mg/dL in men and <50 mg/dL in women or drug treatment; elevated blood pressure systolic ≥ 130 and/or diastolic ≥85 mmHg or current use of antihypertensive medication; elevated fasting glucose ≥100 mg/dL or drug treatment, according to guidelines from the International Diabetes Federation/National Heart, Lung and Blood Institute/American Heart Association (2009)[24]. As described elsewhere, exclusion criteria included a background of alcohol overuse, liver injury, history of previous CVD, gastrointestinal or other disorders, infectious processes, therapy with immunosuppressive drugs, cytotoxic agents or systemic corticosteroids. The protocol and procedures were approved for the Research Ethic Committee for clinical investigations of the University of Navarra according to the Declaration of Helsinki. All participants provided written informed consent. At Navarra-Nutrition centre, a total of 422 participants were registered in the pre-inclusion period and 331 were included in the study, after excluding individuals, who did not meet inclusion criteria (n=2) and participants who refused to participate (n=89). For this study, we also excluded participants who did not have data for measured non-invasive biomarkers (n=5) and patients without DXA values (n=77) (Figure 1).

Study assessment

Clinical and biochemical measurements

At baseline, participants completed an administered survey, which included questions about socio-demographic characteristics, lifestyle behaviours, diseases history, and medication. Smoking habit was classified into never, former, or current smoker as described elsewhere [22]. Blood pressure was measured in triplicate using a validated semiautomatic oscillometer (Omron HEM-705CP). Diabetes was established as previous diagnosis of diabetes or glycated haemoglobin (HbA1c) ≥6.5%, use of antidiabetic medication or fasting glucose ≥126 mg/dl according to the American Diabetes Association (ADA) guidelines [25]. After overnight fasting for at least 12-h, a blood sample was obtained from each participant. Serum and plasma were collected and frozen at -80˚C. All biochemical measurements, including plasma glucose, HbA1c, insulin, total cholesterol, high-density lipoprotein cholesterol (HDL-c), triglyceride, alanine aminotransferase (ALT), aspartate aminotransferase (AST) and gamma-glutamyl transferase (GGT) were performed using standard laboratory enzymatic methods and following validated protocols [22]. The fibroblast growth factor 21 (FGF-21) plasma concentrations were measured using

human FGF-21 Quantikine ELISA Kit (R&D Systems, Minneapolis, MN, USA) with an autoanalyzer system (Triturus, Grifols SA, Barcelona, Spain) following the manufacturer's instructions. Low-density lipoprotein cholesterol (LDL-c) concentration was calculated by Friedewald's formula and the very-low-density lipoprotein cholesterol (VLDL-c) were calculated as triglycerides/5 [26]. Also, homeostatic model assessment for insulin (HOMA-IR) was calculated according to the formula: fasting insulin (mIU/L) x fasting glucose (nmol/L)/22.5 [27].

Dietary variables

Trained dietitians face-to-face administered a semi-quantitative 143-item food frequency questionnaire to estimate energy intake and alcohol consumption [28]. Also, a 17-item questionnaire, which is a modified version of the previously validated questionnaire used in the PREDIMED study to assess the participant´s adherence to the Mediterranean diet was implemented [29].

Physical activity measurement

Physical activity was assessed using the short REGICOR (Registre Gironi del Cor) questionnaire that showed high reliability and sensitivity to detect changes in moderate and vigorous intensity [30,31]. This tool was validated in the Spanish adult population, which is a version of the Minnesota Leisure Time [30]. This questionnaire evaluated the total energy expenditure in leisure- time physical activity using Metabolic Equivalent Tasks (METs) in minutes/week. Physical activities were classified into light-intensity (<4 METs), moderate- intensity (4-5.5 METs), and vigorous- intensity (≥6 METs) as detailed in the report [30]. Sedentary lifestyles were evaluated considering a validated Nurses' Health Study questionnaire [32]. For the present study, physical activity was expressed as MET/hours/week.

Anthropometry and body composition measurements

Anthropometric measurements were performed by trained dietitians following standardized PREDIMED-Plus protocols [22]. Weight, height and WC, were measured using a calibrated scale, a stadiometer and an anthropometric tape, respectively. BMI was conventionally calculated as weight in kilograms divided by the height in square meters (kg/m^2). VAT was estimated using dual-energy X-ray absorptiometry (Lunar iDXA ™, software version 6.0, Madison, WI, USA) connected with enCore™ software, which was assessed by trained operators according to standard procedures supplied by the manufacturer.

Non-invasive markers

The TyG index is a newly described marker that has been reported as a useful screening tool for insulin resistance [21,33,34], NAFLD [35] and as an early predictor of MetS features [36]. This marker was calculated using biochemical data according to the following formula [33],

$$\mathrm{TyG} = \mathrm{Ln}\,[\mathrm{Triglyceride\ (mg/dL)} * \mathrm{glucose\ (mg/dL)/2}].$$

The HSI was validated in a cohort of patients with NAFLD diagnosed by ultrasonography [37]. The HSI equation: $$\mathrm{HSI} = 8 \times \mathrm{ALT/AST\ ratio} + \mathrm{BMI\ (+2, if\ diabetes;\ +2, if\ female)}$$ [37,38] it was also computed to estimate liver status. Another liver marker as an indicator of NAFLD is the FLI,

which was calculated as $\frac{e^{0.953\times\log(\text{triglycerides})+0.139\times\text{BMI}+0.718\times\log(\text{GGT})+0.053\times\text{waist circumference}-15.745}}{1+e^{0.953\times\log(\text{triglycerides})+0.139\times\text{BMI}+0.718\times\log(\text{GGT})+0.053\times\text{waist circumference}-15.745}}\times 100$ as previously described [39].

Statistical analyses

Continuous variables are presented as means ± SD and categorical variables as numbers (n) and percentages (%). One-way analysis of variance (ANOVA) and chi-square tests were used to assess differences between groups, as appropriate. The ANCOVA test after adjustment for the following potential confounders: age (years), physical activity (MET/min/week), energy intake (kcal/d), alcohol intake (g) and smoking status (never, former, current). Bonferroni correction was applied to assess differences in metabolic and liver parameters according to sex-specific VAT tertiles. Crude and multiple linear regression models adjusted by age (years), sex (male and female), physical activity (MET/min/week), energy intake (kcal/d), alcohol intake (g) and smoking status (never, former, current) were fitted to analyze the association between NAFLD biomarkers and the TyG index. Tests of linear trend were assessed assigning the median value of each tertile of TyG and then using it as a continuous variable and correlation was assessed using the Pearson's coefficient. The area under the ROC curve (AUC) was performed to quantify the diagnostic accuracy of TyG index as a predictor of VAT considering as references values of 50th percentile of VAT by sex. All tests were two-sided and the cut-off level of significance was defined at 0.05. Statistical analyses were carried out with Stata 12.0 software (StataCorp LP, College Station, TX).

Results

Study sample characteristics

Baseline characteristics of men and women according to VAT sex- specific tertiles are summarized in Table 1. As expected, BMI and WC increased across VAT tertiles in men and women. No significant differences were found in the frequency of diabetes, hypertension and smoking habits among tertiles in both sexes. Likewise, blood pressure (SBP and DBP) measurements, energy intake, alcohol consumption, adherence to the Mediterranean diet score, and physical activity did not differ statistically.

Crosstalk between VAT, TyG index and NAFLD risk factors in both sexes

Anthropometric, metabolic profile and liver status of participants are reported in Table 2. The adjusted analysis revealed that BMI and WC were significantly increasing through VAT tertiles specific by sex. Moreover, insulin, TyG and HOMA-IR increased with VAT tertiles reaching statistical differences among them (all p<0.001). Glucose and HbA1c did not show differences between tertiles. As concerns lipidic markers, the T3 group presented significantly higher levels of VLDL-c [mean 32.4 mg/dL (95% CI, 29.7-35.2)], triglycerides [mean 162.1 mg/dL (95% CI, 148.3-175.9)] and TG/HDL-c ratio [mean 3.8 mg/dL (95% CI, 3.4-4.3)] than T1 participants, while no associations were found regarding total cholesterol, LDL-c and HDL-c serum levels. Men and women in the highest VAT tertile showed significantly higher ALT levels, HSI

and FLI scores as compared with participants in the lowest tertile of VAT. No significant differences were found in AST and FGF-21 levels in VAT tertiles.

The association of the TyG index with variables related to liver health was explored (Table 3). Linear regression models were fitted considering NAFLD related markers as dependent factors and the TyG index as the independent variable (Table 3). A fully adjusted model including sex revealed that individuals in the 3rd TyG tertile (>9.1-10.7) were significantly associated with higher WC (β=2.62, 95% CI: 0.41 to 4.84, p for trend<0.020), HOMA-IR (β=3.07, 95% CI: 2.28 to 3.86, p for trend<0.001), ALT (β=7.43, 95% CI: 2.23 to 12.63, p for trend=0.005), GGT (β=14.12, 95% CI: 3.64 to 24.61, p for trend=0.008), FGF-21 levels (β=190.69, 95% CI: 93.13 to 288.25, p for trend <0.001), FLI units (β=18.65, 95% CI: 14.97 to 22.33, p for trend<0.001), HSI units (β=3.46, 95% CI: 2.23 to 4.68, p for trend<0.001) than participants in the first TyG tertile. Furthermore, variables related with glucose-insulin homeostasis were significantly correlated with VAT in both sexes expect for men in glucose levels (Figure 2). Glucose (men: r= 0.093, p= 0.292; women: r= 0.264, p= 0.004) (Figure 2A), triglycerides (men: r= 0.291, p<0.001; women: r= 0.220, p=0.016) (Figure 2B), HOMA-IR (men: r= 0.345, p>0.001; women: r= 0.500, p<0.001) (Figure 2C), and TyG index (men: r=0.266, p= 0.002; women: r= 0.322, p<0.001) (Figure 2D).

Receiver operator characteristic (ROC) analyses for the TyG index to predict VAT in male and female

ROC curves were applied to assess the capacity of the TyG index to identify elevated VAT accumulation in both sexes (Figure 3). The AUCs of the TyG index for prediction of VAT were 0.570 (95% CI: 0.48 to 0.66) for men and 0.713 (95% CI: 0.62 to 0.79) for women, which appears as a relevant outcome of their analyses.

Discussion

In this cross-sectional study, VAT and the TyG index were associated with relevant metabolic and liver risk factors linked to NAFLD in male and female with MetS. Moreover, the TyG index could be a reliable indicator of visceral adipose dysfunction in women (AUC: 0.570), but not in men (AUC: 0.713). Many metabolic abnormalities related to insulin resistance are often occurring in obese individuals with higher amount of VAT [9, 40]. In our study, men and women with increased VAT showed higher levels of serum variables related to NAFLD. Interestingly, the TyG index and atherogenic lipid profile (VLDL-c, triglycerides and TG/HDL-c ratio) were significantly increased across tertiles of VAT specific by sex independently of confounder factors. In line with our results, Lee and colleagues observed that VAT and triglycerides were independent risk factors for hepatic steatosis [41]. VAT as the main source of free fatty acids (FFAs) and other biological compounds, which entering portal circulation into the liver contribute to hepatic fat accumulation [40]. Younger women have the ability to partition FFA's towards ketone body production rather than VLDL- triacylglycerol, but is affected in postmenopausal women that have a negative impact on liver status [42]. Moreover, a statically significant increase of liver markers (ALT, FLI and HSI) in both sexes with excessive accumulation of VAT depots was found. In agreement with our results, Chung et al. indicated that increased VAT was associated with higher ALT levels [43] and NASH or significant fibrosis

in subjects diagnosed with NAFLD [44]. Based on these data, central adiposity plays a key role in NAFLD pathogenesis [10, 15, 45] promoting liver damage [44], insulin resistance and disrupted lipid metabolism [46].

Nowadays, NAFLD has become a public health problem with a negative impact over the individual's health, socioeconomic and health care system [6, 47]. In this context, early screening is crucial in the NAFLD pathogenesis [7]. Liver biopsy is the gold standard for NAFLD diagnosis [7]. However, it has several limitations such as sampling error, cost, medical complications, and technical difficulties [48]. In this regard, many techniques have been used in the detection and featuring NAFLD that showed to be relatively effective, inexpensive and useful in a primary health care setting [20, 48–50]. The TyG index is a novel marker that has exhibited a good accuracy for recognizing insulin resistance [21, 51]. Furthermore, this marker showed highly sensitive for detecting NAFLD [35]. Interestingly, the multivariable regression analysis evidenced that individuals with higher TyG (> 9.1) value were associated with higher levels of HOMA-IR ALT, GGT, FGF-21, and HSI units compared with lower TyG values (≤ 8.7 units), after adjusting for sex and potential confounders. These results could be partially explaining by that NAFLD is prevalent in men but this trend increases in postmenopausal women, which is related to regional adiposity and hormones effects in ageing [15, 42]. Serum aminotransferase levels might associated with insulin resistance [52]. Bonnet et al. found that increased levels of ALT and GGT are strongly associated with hepatic insulin resistance and decreased hepatic insulin clearance [53]. The FGF-21 is primarily produced in hepatocytes and is implicated in the regulation of glucose-lipid metabolism, insulin sensitivity, inflammation and energy homeostasis [54]. Several clinical studies and reviews have documented that disrupted adipose tissue and excessive intrahepatic fat accumulation may trigger FGF-21 resistance [54, 55]. Thus, Shen and colleagues [56] found that NAFLD patients showed significantly higher serum FGF-21 levels compared with subjects without NAFLD [56]. The FLI was positively associated with the third tertile of the TyG index, in the same manner that the HSI, indicating that subjects with higher values of TyG had 3.46 more units of HSI compared to the reference (lower values). Taking together, these results can be explained by the fact that insulin resistance is a major feature of NAFLD by increasing *de novo* lipogenesis and FFAs flux to the liver through decreased inhibition of lipolysis [5] promoting inflammation, oxidative stress [57] and hepatocyte injure [58].

DXA has been considered the gold standard for body composition measurements [17]. Nevertheless, this imaging technic for assessing adipose tissue distribution is expensive and not feasible for community screening. In our results, we observed a close relationship between insulin resistance and dysfunctional VAT. Interestingly, men had higher amounts of VAT than women. Meanwhile, women and men with ≥ VAT median had similar TyG values (data not shown). Moreover, the ROC curves indicate a moderate predictive ability of the TyG to discriminate VAT in women (AUC = 0.713), but it was weak for men (AUC = 0.570).

The connection between body fat distribution and adipose tissue biology with insulin resistance differ by sexes, age and other factors [13]. In general, women have more total body fat mass and men presented higher abdominal/visceral fat mass [13]. However, decreased levels of estrogen and adipose tissue re-

distribution by increased depots of VAT are characterized in postmenopausal women [11, 13]. Estrogen and testosterone are involved in glucose and lipid metabolism. A disbalance of sex hormone levels promote insulin resistance and an atherogenic lipid profile, which increased the risk of CVD in older women [13]. Interestingly, some studies have suggested that obese women are more insulin sensitivity than men despite a higher amount of VAT [11]. However, the plausible mechanism is still unclear. Recently, the Netherlands Epidemiology of Obesity Study (NEO) showed that in obese women, VAT was differently associated with cardiometabolic risk factors as compared obese men [12]. But, this is in contrast with Ferrara et al. who reported that older obese men are more insulin resistant compared with older women even adjusted for differences in abdominal fat distribution measured by DXA [14]. This result can be attributed to the limited number of participants. One possible explanation for our results could be that women exhibit a greater amount of FFAs delivery derived from VAT lipolysis [59]. Thus, there are differences between sexes in the lipid storage capacity and function [57]. Serra et al. showed that postmenopausal women (overweight or obese) diagnosed with MetS had lower adipose tissue LPL activity and limited capacity for lipid accumulation in subcutaneous abdominal adipose tissue leading to higher rates of lipid, accumulation of VAT and insulin resistance [60]. These results reinforce that a high amount of VAT has a negative impact over metabolic and liver status leading to elevated insulin levels and hepatic insulin resistance [5, 45]. In this context, the improvement of the knowledge of these interrelationships in male and female with MetS should be useful to easily identify individuals with a high risk of NAFLD, which may allow early intervention and prevention of NAFLD complications.

The strengths of this study include the relatively large sample size of individuals with MetS in the framework of PREDIMED-Plus. Also, VAT was objectively measured with a validated imaging technique. However, some limitations required considerations. First, the cross-sectional design cannot imply a causal relationship. Second, the lack of NAFLD diagnosis by liver biopsy or imaging techniques, but it is important to note that liver biopsy is not available or feasible in large epidemiological studies. On the other hand, validated non-invasive markers were used to estimate hepatic fat accumulation [38].

Conclusion

A higher accumulation of VAT is associated with cardiometabolic and liver risk factors linked to NAFLD in older men and women with overweight/obese and MetS. Moreover, we reinforce that in addition to anthropometric measurements such as WC or DXA approach, the TyG index could be a useful simple marker to identify dysfunctional VAT phenotype in women with MetS better than in men.

Abbreviations

MetS
Metabolic syndrome
CVD
Cardiovascular disease
NAFLD

non-alcoholic fatty liver disease
VAT
Visceral adipose tissue
TyG
Triglyceride and glucose index
FGF-21
Fibroblast growth factor-21
FLI
Fatty liver index
HSI
Hepatic steatosis index

Declarations

Ethics approval and consent to participate

Research Ethics Committees from all recruitment centers approved the study protocol following the rules of the Declaration of Helsinki on July 24, 2014 (approval number ISRCTN898988709). All participants were informed of procedures and signed an informed consent.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Author's contribution

VB-V and MAZ contributed equally in this study. VB-V, IA, MAZ, and JAM were involved in the study design, collecting data, analysis, interpretation of the results and draft preparation. JAT, JK, DR, XP, EC, MAM-G, CS-O, ET, DC, MM-G, FT, MF, RE, ER, JS-S, LD and CMM were contributed to intellectual content and commented on drafts. All authors approved the final version.

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Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

References

1. Dominguez LJ, Barbagallo M. The biology of the metabolic syndrome and aging. Curr Opin Clin Nutr Metab Care. 2016;19:5–11.
2. Rani V, Deep G, Singh RK, Palle K, Yadav UCS. Oxidative stress and metabolic disorders: Pathogenesis and therapeutic strategies. Life Sci Elsevier Inc. 2016;148:183–93.
3. Mansourian M, Babahajiani M, Jafari-Koshki T, Roohafza H, Sadeghi M, Sarrafzadegan N. Metabolic Syndrome Components and Long-Term Incidence of Cardiovascular Disease in Eastern Mediterranean Region: A 13-Year Population-Based Cohort Study. Metab Syndr Relat Disord. 2019;17:362–6.

4. Schroeder EB, Bayliss EA, Daugherty SL, Steiner JF. Gender differences in cardiovascular risk factors in incident diabetes. Women's Heal Issues Elsevier. 2014;24:e61–8.
5. Kitade H, Chen G, Ni Y, Ota T. Nonalcoholic fatty liver disease and insulin resistance: New insights and potential new treatments. Nutrients. 2017;9:1–13.
6. Younossi ZM. Non-alcoholic fatty liver disease – A global public health perspective. J Hepatol. 2019;70:531–44.
7. Chalasani N, Younossi Z, Lavine JE, Charlton M, Cusi K, Rinella M, et al. The diagnosis and management of nonalcoholic fatty liver disease: Practice guidance from the American Association for the Study of Liver Diseases. Hepatology. 2018;67:328–57.
8. Rinella ME. Nonalcoholic Fatty Liver Disease. JAMA. 2015;313:2263.
9. González-Muniesa P, Mártinez-González M-A, Hu FB, Després J-P, Matsuzawa Y, Loos RJF, et al. Obesity Nat Rev Dis Prim. 2017;3:17034.
10. Bosy-Westphal A, Braun W, Albrecht V, Müller MJ. Determinants of ectopic liver fat in metabolic disease. Eur J Clin Nutr. 2019;73:209–14.
11. Frank AP, de Souza Santos R, Palmer BF, Clegg DJ. Determinants of body fat distribution in humans may provide insight about obesity-related health risks. J Lipid Res. 2019;60:1710–9.
12. Elffers TW, de Mutsert R, Lamb HJ, de Roos A, Willems van Dijk K, Rosendaal FR, et al Body fat distribution, in particular visceral fat, is associated with cardiometabolic risk factors in obese women. He M, editor. PLoS One. 2017;12:e0185403.
13. Karastergiou K, Smith SR, Greenberg AS, Fried SK. Sex differences in human adipose tissues – the biology of pear shape. Biol Sex Differ. 2012;3:13.
14. Ferrara CM, Goldberg AP, Nicklas BJ, Sorkin JD, Ryan AS. Sex differences in insulin action and body fat distribution in overweight and obese middle-aged and older men and women. Appl Physiol Nutr Metab. 2008;33:784–90.
15. Suzuki A, Abdelmalek MF. Nonalcoholic Fatty Liver Disease in Women. Women's Health. 2009;5:191–203.
16. Kuk JL, Saunders TJ, Davidson LE, Ross R. Age-related changes in total and regional fat distribution. Ageing Res. 2009;8:339–48.
17. Micklesfield LK, Goedecke JH, Punyanitya M, Wilson KE, Kelly TL. Dual-Energy X-Ray Performs as Well as Clinical Computed Tomography for the Measurement of Visceral Fat. Obesity. 2012;20:1109–14.
18. Wu P, Chen Q, Chen L, Zhang P, Xiao J, Chen X, et al. Dose-response relationship between alanine aminotransferase levels within the reference interval and metabolic syndrome in Chinese adults. Yonsei Med J. 2017;58:158–64.
19. Cheung C-L, Lam KS, Wong IC, Cheung BM. Non-invasive score identifies ultrasonography-diagnosed non-alcoholic fatty liver disease and predicts mortality in the USA. BMC Med. 2014;12:154.

20. Cantero I, Elorz M, Abete I, Marin BA, Herrero JI, Monreal JI, et al. Ultrasound/elastography techniques, lipidomic and blood markers compared to magnetic resonance imaging in non-alcoholic fatty liver disease adults. Int J Med Sci. 2019;16:75–83.
21. Navarro-González D, Sánchez-Íñigo L, Pastrana-Delgado J, Fernández-Montero A, Martinez JA. Triglyceride–glucose index (TyG index) in comparison with fasting plasma glucose improved diabetes prediction in patients with normal fasting glucose: The Vascular-Metabolic CUN cohort. Prev Med (Baltim). 2016;86:99–105.
22. Martínez-González MA, Buil-Cosiales P, Corella D, Bulló M, Fitó M, Vioque J, et al. Cohort Profile: Design and methods of the PREDIMED-Plus randomized trial. Int J Epidemiol. 2019;48:387–8o.
23. Sayón-Orea C, Razquin C, Bulló M, Corella D, Fitó M, Romaguera D, et al. Effect of a Nutritional and Behavioral Intervention on Energy-Reduced Mediterranean Diet Adherence Among Patients With Metabolic Syndrome. JAMA. 2019;322:1486.
24. Alberti KGMM, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA, et al. Harmonizing the Metabolic Syndrome Circulation. 2009;120:1640–5.
25. Atroshenko SA, Korolyov IA, Didenko N. Standards of Medical Care in Diabetes–2014. Diabetes Care. 2014;37:14–80.
26. Bairaktari ET, Seferiadis KI, Elisaf MS. Evaluation of Methods for the Measurement of Low-Density Lipoprotein Cholesterol. J Cardiovasc Pharmacol Ther. 2005;10:45–54.
27. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and ?-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia. 1985;28:412–9.
28. Fernández-Ballart JD, Piñol JL, Zazpe I, Corella D, Carrasco P, Toledo E, et al. Relative validity of a semi-quantitative food-frequency questionnaire in an elderly Mediterranean population of Spain. Br J Nutr. 2010;103:1808–16.
29. Schröder H, Fitó M, Estruch R, Martínez-González MA, Corella D, Salas-Salvadó J, et al. A Short Screener Is Valid for Assessing Mediterranean Diet Adherence among Older Spanish Men and Women. J Nutr. 2011;141:1140–5.
30. Molina L, Sarmiento M, Peñafiel J, Donaire D, Garcia-Aymerich J, Gomez M, et al. Validation of the Regicor Short Physical Activity Questionnaire for the Adult Population. PLoS One. 2017;12:e0168148.
31. Rosique-Esteban N, Babio N, Díaz-López A, Romaguera D, Alfredo Martínez J, Sanchez VM, et al. Leisure-time physical activity at moderate and high intensity is associated with parameters of body composition, muscle strength and sarcopenia in aged adults with obesity and metabolic syndrome from the PREDIMED-Plus study. Clin Nutr. 2019;38:1324–31.
32. Martínez-González MA, López-Fontana C, Varo JJ, Sánchez-Villegas A, Martinez JA. Validation of the Spanish version of the physical activity questionnaire used in the Nurses' Health Study and the Health Professionals' Follow-up Study. Public Health Nutr. 2005;8:920–7.

33. Fedchuk L, Nascimbeni F, Pais R, Charlotte F, Housset C, Ratziu V. Performance and limitations of steatosis biomarkers in patients with nonalcoholic fatty liver disease. Aliment Pharmacol Ther. 2014;40:1209–22.
34. Zheng R, Du Z, Wang M, Mao Y, Mao W. A longitudinal epidemiological study on the triglyceride and glucose index and the incident nonalcoholic fatty liver disease. Lipids Health Dis Lipids in Health Disease. 2018;17:1–9.
35. Zhang S, Du T, Zhang J, Lu H, Lin X, Xie J, et al. The triglyceride and glucose index (TyG) is an effective biomarker to identify nonalcoholic fatty liver disease. Lipids Health Dis Lipids in Health Disease. 2017;16:1–8.
36. Moon S, Park JS, Ahn Y. The Cut-off Values of Triglycerides and Glucose Index for Metabolic Syndrome in American and Korean Adolescents. J Korean Med Sci. 2017;32:427–33.
37. Lee J-H, Kim D, Kim HJ, Lee C-H, Yang JI, Kim W, et al. Hepatic steatosis index: A simple screening tool reflecting nonalcoholic fatty liver disease. Dig Liver Dis. 2010;42:503–8.
38. Bugianesi E, Rosso C, Cortez-Pinto H. How to diagnose NAFLD in 2016. J Hepatol. 2016;65:643–4.
39. Bedogni G, Bellentani S, Miglioli L, Masutti F, Passalacqua M, Castiglione A, et al. The Fatty Liver Index: a simple and accurate predictor of hepatic steatosis in the general population. BMC Gastroenterol. 2006;6:33.
40. Parker R. The role of adipose tissue in fatty liver diseases. Liver Res Elsevier BV. 2018;2:35–42.
41. Lee HW, Kim KJ, Jung KS, Chon YE, Huh JH, Park KH, et al. The relationship between visceral obesity and hepatic steatosis measured by controlled attenuation parameter. PLoS One. 2017;12:e0187066.
42. Ballestri S, Nascimbeni F, Baldelli E, Marrazzo A, Romagnoli D, Lonardo A. NAFLD as a Sexual Dimorphic Disease: Role of Gender and Reproductive Status in the Development and Progression of Nonalcoholic Fatty Liver Disease and Inherent Cardiovascular Risk. Adv Ther Springer Healthcare. 2017;34:1291–326.
43. Chung GE, Kim D, Kwark MS, Kim W, Yim JY, Kim YJ, et al. Visceral Adipose Tissue Area as an Independent Risk Factor for Elevated Liver Enzyme in Nonalcoholic Fatty Liver Disease. Medicine. 2015;94:e573.
44. Yu SJ, Kim W, Kim D, Yoon J-H, Lee K, Kim JH, et al. Visceral Obesity Predicts Significant Fibrosis in Patients With Nonalcoholic Fatty Liver Disease. Medicine. 2015;94:e2159.
45. Qureshi K, Abrams GA. Metabolic liver disease of obesity and role of adipose tissue in the pathogenesis of nonalcoholic fatty liver disease. World J Gastroenterol. 2007;13:3540–53.
46. Khan RS, Bril F, Cusi K, Newsome PN. Modulation of Insulin Resistance in Nonalcoholic Fatty Liver Disease. Hepatology. 2019;70.
47. Younossi Z, Anstee QM, Marietti M, Hardy T, Henry L, Eslam M, et al. Global burden of NAFLD and NASH: Trends, predictions, risk factors and prevention. Nat Rev Gastroenterol Hepatol. 2018;15:11–20.

48. Zhou JH, She ZG, Li HL, Cai JJ. Noninvasive evaluation of nonalcoholic fatty liver disease: Current evidence and practice. World J Gastroenterol. 2019;25:1307–26.
49. Martínez-González MA, Bastarrika G. Mediterranean diet as the ideal model for preventing non-alcoholic fatty liver disease (NAFLD). [Epub ahead of print] Hepatobiliary Surg Nutr Publ 2019.
50. Bullón-Vela V, Abete I, Tur JA, Pintó X, Corbella E, Martínez-González MA, et al. The influence of lifestyle factors and staple foods from the Mediterranean Diet on non-alcoholic fatty liver disease among elder with metabolic syndrome features. Nutrition. 2019;71:110620.
51. Du T, Yuan G, Zhang M, Zhou X, Sun X, Yu X. Clinical usefulness of lipid ratios, visceral adiposity indicators, and the triglycerides and glucose index as risk markers of insulin resistance. Cardiovasc Diabetol. 2014;13:146.
52. Zhao L, Cheng J, Chen Y, Li Q, Han B, Chan Y, et al. Serum alanine aminotransferase/aspartate aminotransferase ratio is one of the best markers of insulin resistance in the Chinese population. Nutr Metab. 2017;14:64.
53. Bonnet F, Ducluzeau PH, Gastaldelli A, Laville M, Anderwald CH, Konrad T, et al. Liver enzymes are associated with hepatic insulin resistance, insulin secretion, and glucagon concentration in healthy men and women. Diabetes. 2011;60:1660–7.
54. Liu J, Xu Y, Hu Y, Wang G. The role of fibroblast growth factor 21 in the pathogenesis of non-alcoholic fatty liver disease and implications for therapy. Metabolism. Elsevier Inc.; 2015;64:380–90.
55. Inagaki T. Research Perspectives on the Regulation and Physiological Functions of FGF21 and its Association with NAFLD. Front Endocrinol (Lausanne). 2015;6:1–7.
56. Shen Y, Ma X, Zhou J, Pan X, Hao Y, Zhou M, et al. Additive relationship between serum fibroblast growth factor 21 level and coronary artery disease. Cardiovasc Diabetol Cardiovascular Diabetology. 2013;12:124.
57. Koliaki C, Szendroedi J, Kaul K, Jelenik T, Nowotny P, Jankowiak F, et al. Adaptation of Hepatic Mitochondrial Function in Humans with Non-Alcoholic Fatty Liver Is Lost in Steatohepatitis. Cell Metab Elsevier Inc. 2015;21:739–46.
58. Bullón-Vela MVanessa, Abete I, Martínez J, Alfredo, Zulet MA. Obesity and nonalcoholic fatty liver disease: role of oxidative stress. In: Marti del Moral A, García CMA, edi- tors. Obesity: oxidative stress and dietary antioxidants. London: Elsevier; 2018. pp. 111–33.
59. Nielsen S, Guo Z, Johnson CM, Hensrud DD, Jensen MD. Splanchnic lipolysis in human obesity. J Clin Invest. 2004;113:1582–8.
60. Serra MC, Ryan AS, Goldberg AP. Reduced LPL and subcutaneous lipid storage capacity are associated with metabolic syndrome in postmenopausal women with obesity. Obes Sci Pract. 2017;3:106–14.

Tables

Table 1. Clinical and lifestyle characteristics of subjects with MetS according to tertiles of VAT by sex

	Tertiles of visceral adipose tissue (kg)								
	Men (n=133)				Women (n=121)				
	T1 (n=45) (1.29 to ≤2.42)	T2 (n=44) (>2.42 to ≤3.10)	T3 (n=44) (>3.10 to 5.45)	p-value†	T1 (n=41) (0.77 to ≤1.60)	T2 (n=40) (>1.60 to ≤2.06)	T3 (n=40) (>2.06 to 3.59)	p-value†	p-value¶
ears)	63.9 ± 5.8	64.5 ± 5.8	64.6 ± 5.1	0.818	67.7 ± 3.5	67.0 ± 4.2	67.5 ± 4.4	0.751	<0.001
kg/m^2)	30.0 ± 2.2	31.5 ± 2.2	34.0 ± 2.9	<0.001	30.6 ± 2.9	32.4 ± 3.6	34.4 ± 3.2	<0.001	0.128
m)	103.5 ± 5.8	109.5 ± 6.2	117.3 ± 7.6	<0.001	97.1 ± 6.5	102.8 ± 6.9	109.5 ± 7.2	<0.001	<0.001
kg)	2.0 ± 0.3	2.8 ± 0.2	3.8 ± 0.5	<0.001	1.3 ± 0.2	1.8 ± 0.1	2.5 ± 0.4	<0.001	<0.001
tes, n (%)	19 (42.2)	16 (36.4)	17 (38.6)	0.849	10 (24.4)	16 (40.0)	19 (47.5)	0.089	0.755
tension, n (%)	40 (88.9)	42 (95.5)	42 (95.5)	0.362	39 (95.1)	38 (95.0)	37 (92.5)	0.851	0.362
mmHg)	147.6 ± 18.5	142.3 ± 13.8	143.4 ± 14.7	0.250	145.8 ± 16.9	143.5 ± 15.8	142.0 ± 16.2	0.578	0.734
mmHg)	88.5 ± 9.9	88.6 ± 8.5	88.0 ± 6.7	0.941	85.1 ± 8.9	87.6 ± 9.9	86.7 ± 8.6	0.455	0.082
ng habits, n (%)				0.087				0.674	<0.001
smoker	12 (26.7)	7 (15.9)	4 (9.1)		30 (73.2)	23 (57.5)	25 (62.5)		
er smoker	23 (51.1)	26 (59.1)	34 (77.3)		8 (19.5)	13 (32.5)	11 (27.5)		
nt smoker	10 (22.2)	11 (25.0)	6 (13.6)		3 (7.3)	4 (10.0)	4 (10.0)		
ol intake (g/d)	15.2 ± 14.3	18.5 ± 20.3	23.3 ± 20.6	0.119	2.4 ± 6.2	5.3 ± 8.3	2.6 ± 4.9	0.102	<0.001
y intake (kcal/d)	2655.4 ± 580.2	2670.0 ± 439.7	2789.0 ± 550.2	0.428	2451.5 ± 549.5	2407.0 ± 511.9	2474.2 ± 413.4	0.827	<0.001
ence to MedDiet (0-17)	9 ± 2.7	8.5 ± 2.2	8.8 ± 2.5	0.636	9.2 ± 2.5	9.0 ± 2.8	9.4 ± 2.5	0.767	0.169
cal activity /hours/week)	59.0 ± 57.6	61.2 ± 49.6	46.9 ± 41.1	0.356	43.2 ± 33.6	39.4 ± 32.9	36.0 ± 26.5	0.588	0.002

5 is considered statistically significant. Data are expressed as mean ± SD.

e† for differences between tertiles of visceral fat mass by sex was calculated by chi-square or ANOVA as appropriate.

e¶ for differences between men and women was calculated by Student's t- test.

viations: BMI, Body mass index; WC, waist circumference; VAT, visceral adipose tissue; SBP, Systolic blood pressure; DBP, diastolic pressure; MedDiet, Mediterranean diet; MET, metabolic equivalent.

Table 2. Anthropometric, body composition, metabolic profile and liver status in subjects with MetS according to tertiles of VAT specific by sex

	Tertiles of visceral adipose tissue (kg)			
	T1	T2	T3	p-value[†]
	(n=86)	(n=84)	(n=84)	
	(0.77 to 2.42)	(1.60 to 3.10)	(2.06 to 5.45)	
Anthropometric and body composition				
BMI (kg/m^2)	30.3 (29.7-30.9)[a,b,c]	32.0 (31.4-32.6)[b,c]	34.1 (33.5-34.8)	<0.001
WC (cm)	101.0 (99.5-102.6)[a,b,c]	106.3 (104.8-107.8)[b,c]	113.1 (111.5-114.6)	<0.001
VAT (Kg)	1.7 (1.6-1.8)[a,b,c]	2.3 (2.2-2.4)[b,c]	3.1 (3.0-3.2)	<0.001
Glucose profile				
Glucose (mg/dL)	115.1 (108.0-122.2)	118.2 (111.2-125.2)	123.6 (1116.6-130.6)	0.247
HbA1c (%)	6.0 (5.8-6.2)	6.2 (6.0-6.4)	6.2 (6.0-6.5)	0.281
TyG index	8.8 (8.7-8.9)[a,b,c]	9.0 (8.9-9.1)	9.1 (9.0-9.2)	0.001
Insulin (mU/L)	10.2 (8.5-11.9)[a,b,c]	13.8 (12.1-15.4)	16.5 (14.9-18.2)	<0.001
HOMA-IR	2.9 (2.3-3.4)[a,b,c]	4.1 (3.5-4.6)	5.0 (4.5-5.5)	<0.001
Lipid profile				
Total cholesterol (mg/dL)	198.0 (190.2-205.8)	201.2 (193.4-209.0)	205.6 (197.7-213.5)	0.411
LDL-c (mg/dL)	125.5 (118.5-132.5)	125.8 (118.7-132.9)	129.1 (121.8-136.4)	0.747
HDL-c (mg/dL)	47.8 (45.6-49.9)	45.5 (43.3-47.6)	45.9 (43.7-48.1)	0.315
VLDL-c (mg/dL)	25.0 (22.2-27.7)[a,b,c]	30.2 (27.4-32.9)	32.4 (29.7-35.2)	<0.001
Triglycerides (mg/dL)	124.8 (111.0-138.5) [a,b,c]	150.9 (137.2-164.5)	162.1 (148.3-175.9)	<0.001
TG/HDL-c ratio	2.9 (2.4-3.3)[a,c]	3.6 (3.1-4.0)	3.8 (3.4-4.3)	0.008
Liver status				
ALT (U/L)	23.3 (19.1-27.4)[a,c]	29.2 (25.1-33.3)	32.0 (27.9-36.1)	0.013
AST (U/L)	21.8 (19.0-24.6)	24.2 (21.4-26.9)	25.0 (22.2-27.7)	0.268
GGT (U/L)	37.2 (28.7-45.6)	46.8 (38.5-55.2)	40.7 (32.2-49.2)	0.272
FGF-21 (pg/mL)	378.5 (294.5-462.5)	484.7 (403.8-565.6)	430.5 (346.7-514.4)	0.207
FLI (arbitrary units)	66.6 (63.9-69.3)[a,b,c]	79.8 (77.1-82.5)[b,c]	86.9 (84.2-89.6)	<0.001
HSI (arbitrary units)	40.2 (39.4-41.1)[a,b,c]	43.2 (42.2-44.1)[b,c]	45.9 (45.1-46.8)	<0.001

*Tertiles of visceral adipose tissue specific by sex.

p<0.05 is considered statistically significant. Data are expressed as mean (95% CI) Data is adjusted by age (years), physical activity(MET/hours/week), energy intake (kcal/d), alcohol intake (g) and smoking status (never, former, current). FGF-21 available in 211 patients.

a,b significant differences between T1 vs T2.

a,c significant differences between T1 vs T3.

b,c significant differences between T2 vs T3.

Abbreviations: BMI, boddy mass index; WC, waist circumference; VAT, visceral adipose tissue; HbA1c, glycated haemoglobin A1c; TyG, Triglyceride and glucose index; LDL-c, Low density lipoprotein cholesterol; HDL-c, High density lipoprotein cholesterol; TG/HDL ratio, triglycerides/ High density lipoprotein cholesterol ratio; VLDL, Very low-density lipoprotein cholesterol; ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyl transferase; FGF-21, fibroblast growth factor- 21; FLI, fatty liver index; HSI, hepatic steatosis index.

Table 3. Multivariable linear regression analyses evaluating the association between TyG index tertiles as independent variable and liver status as dependent variable

	Tertiles of TyG index			p for trend
	T1	T2	T3	
	(n=109)	(n=110)	(n=107)	
	(7.3-8.7)	(>8.7-9.1)	(>9.1-10.7)	
	β estimates (95% CI)	β estimates (95% CI)	β estimates (95% CI)	
WC (cm)				
Crude	(0 Ref.)	1.32 (-1.06 to 3.71)	3.21 (0.81 to 5.61)	0.009
Multivariable adjusted	(0 Ref.)	1.81 (-0.37 to 3.99)	2.62 (0.41 to 4.84)	0.020
HOMA-IR				
Crude	(0 Ref.)	1.21 (0.43 to 1.99)	3.09 (2.31 to 3.88)	<0.001
Multivariable adjusted	(0 Ref.)	1.25 (0.48 to 2.01)	3.07 (2.28 to 3.86)	<0.001
ALT (U/L)				
Crude	(0 Ref.)	3.79 (-1.48 to 9.07)	8.14 (2.83 to 13.45)	0.003
Multivariable adjusted	(0 Ref.)	4.79 (-0.32 to 9.90)	7.43 (2.23 to 12.63)	0.005
AST (U/L)				
Crude	(0 Ref.)	1.84 (-1.51 to 5.18)	2.56 (-0.80 to 5.93)	0.137
Multivariable adjusted	(0 Ref.)	2.29 (-1.00 to 5.57)	1.98 (-1.36 to 5.33)	0.246
GGT (U/L)				
Crude	(0 Ref.)	3.06 (-7.39 to 13.51)	16.72 (6.17 to 27.26)	0.002
Multivariable adjusted	(0 Ref.)	4.07 (-6.21 to 14.34)	14.12 (3.64 to 24.61)	0.008
FGF-21 (pg/mL)*				
Crude	(0 Ref.)	92.45 (-2.17 to 187.07)	195.11 (100.49 to 289.73)	<0.001
Multivariable adjusted	(0 Ref.)	91.26 (-5.09 to 187.62)	190.69 (93.13 to 288.25)	<0.001
FLI (arbitrary units)				
Crude	(0 Ref.)	11.18 (7.46 to 14.90)	19.60 (15.84 to 23.36)	<0.001
Multivariable adjusted	(0 Ref.)	11.78 (8.18 to 15.39)	18.65 (14.97 to 22.33)	<0.001
HSI (arbitrary units)†				
Crude	(0 Ref.)	1.76 (0.52 to 3.00)	3.35 (2.11 to 4.60)	<0.001
Multivariable adjusted	(0 Ref.)	1.95 (0.75 to 3.16)	3.46 (2.23 to 4.68)	<0.001

p<0.05 is considered statistically significant. Data are expressed as mean (95% CI). Data is adjusted by age (years), sex (male and female), physical activity (MET/hours/week), energy intake (kcal/d), alcohol intake (g) and smoking status (never, former, current). *FGF-21 available in 278 patients. †adjusted for all variables except for sex.

Abbreviations: WC, waist circumference; HOMA-IR, homeostatic model assessment for insulin; ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyl transferase; FGF-21, fibroblast growth factor-21; HSI, hepatic steatosis index; FLI, fatty liver index.

Figures

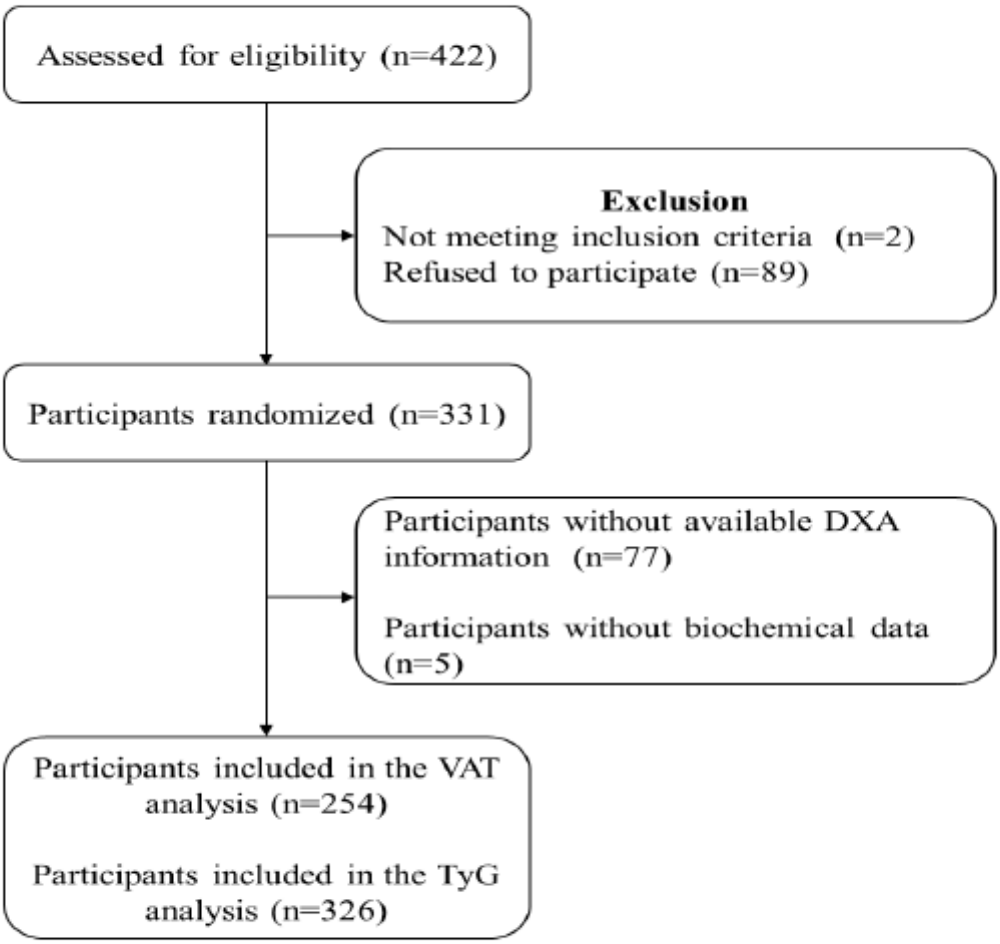

Figure 1

Study flow for the selection of the study population

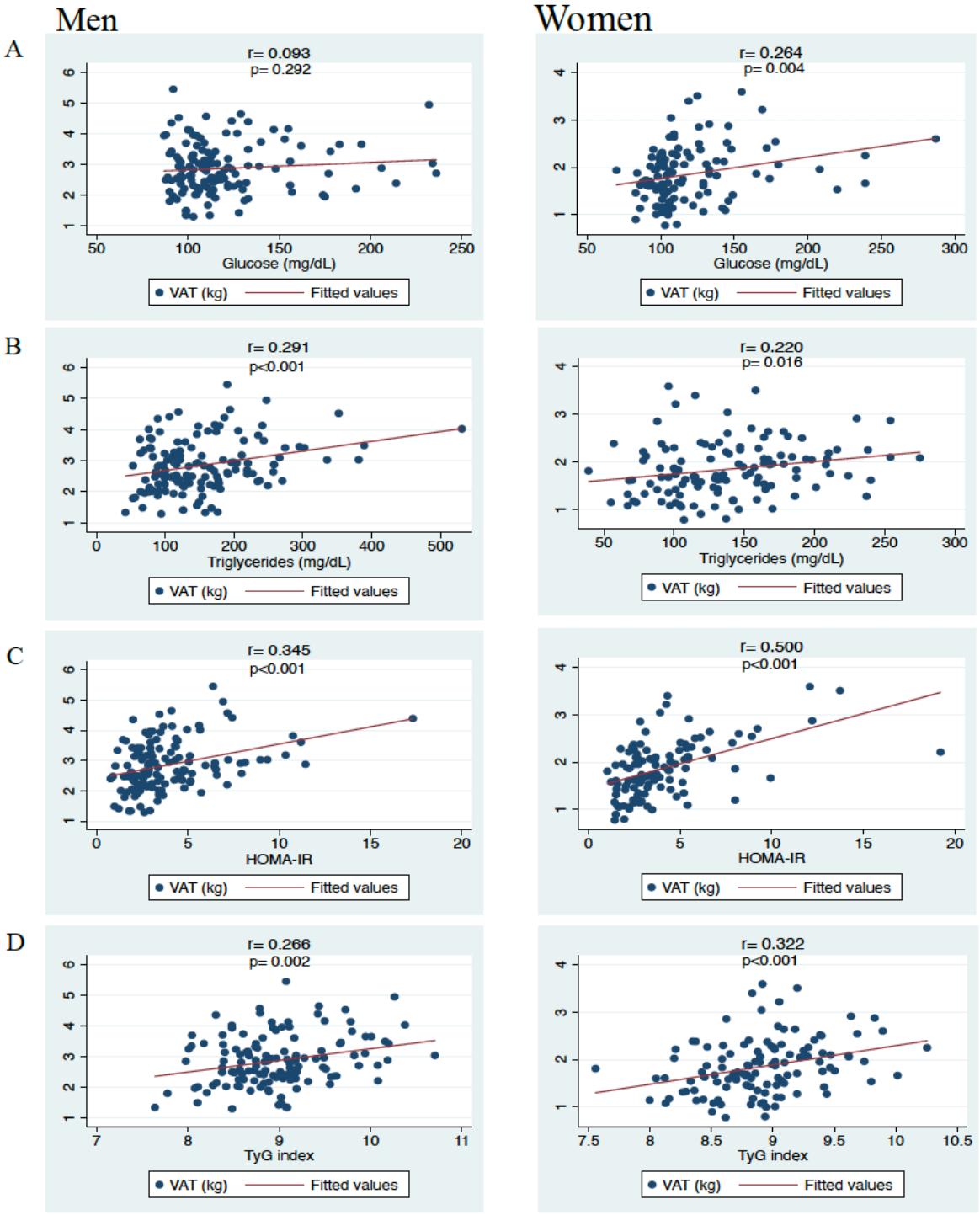

Figure 2

Correlations between VAT and parameters related to glucose and insulin homeostasis in men and women diagnosed with MetS

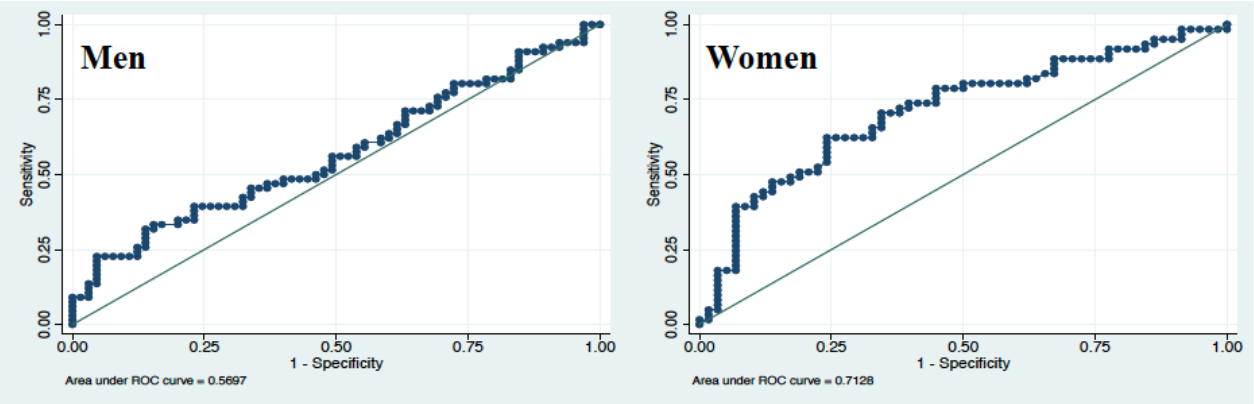

Figure 3

Receiver operating characteristics curve (ROC) analysis of predictive value of the TyG index in subjects with metabolic syndrome. Legend: VAT Cut-off men: ≥ 2.777 kg; VAT cutt-off women: ≥1.748 kg