ASSOCIATION OF NOVEL ANTHROPOMETRIC INDEXES WITH METABOLIC SYNDROME

EDITED BY: Ozra Tabatabaei-Malazy, Roya Kelishadi, Patricia Khashayar and Mostafa Qorbani
PUBLISHED IN: Frontiers in Endocrinology

frontiers Research Topics

frontiers

ISSN 1664-8714
ISBN 978-2-88976-990-2
DOI 10.3389/978-2-88976-990-2

About Frontiers

Frontiers is more than just an open-access publisher of scholarly articles: it is a pioneering approach to the world of academia, radically improving the way scholarly research is managed. The grand vision of Frontiers is a world where all people have an equal opportunity to seek, share and generate knowledge. Frontiers provides immediate and permanent online open access to all its publications, but this alone is not enough to realize our grand goals.

Frontiers Journal Series

The Frontiers Journal Series is a multi-tier and interdisciplinary set of open-access, online journals, promising a paradigm shift from the current review, selection and dissemination processes in academic publishing. All Frontiers journals are driven by researchers for researchers; therefore, they constitute a service to the scholarly community. At the same time, the Frontiers Journal Series operates on a revolutionary invention, the tiered publishing system, initially addressing specific communities of scholars, and gradually climbing up to broader public understanding, thus serving the interests of the lay society, too.

Dedication to Quality

Each Frontiers article is a landmark of the highest quality, thanks to genuinely collaborative interactions between authors and review editors, who include some of the world's best academicians. Research must be certified by peers before entering a stream of knowledge that may eventually reach the public - and shape society; therefore, Frontiers only applies the most rigorous and unbiased reviews.
Frontiers revolutionizes research publishing by freely delivering the most outstanding research, evaluated with no bias from both the academic and social point of view. By applying the most advanced information technologies, Frontiers is catapulting scholarly publishing into a new generation.

What are Frontiers Research Topics?

Frontiers Research Topics are very popular trademarks of the Frontiers Journals Series: they are collections of at least ten articles, all centered on a particular subject. With their unique mix of varied contributions from Original Research to Review Articles, Frontiers Research Topics unify the most influential researchers, the latest key findings and historical advances in a hot research area! Find out more on how to host your own Frontiers Research Topic or contribute to one as an author by contacting the Frontiers Editorial Office: frontiersin.org/about/contact

ASSOCIATION OF NOVEL ANTHROPOMETRIC INDEXES WITH METABOLIC SYNDROME

Topic Editors:
Ozra Tabatabaei-Malazy, Tehran University of Medical Sciences, Iran
Roya Kelishadi, Isfahan University of Medical Sciences, Iran
Patricia Khashayar, Ghent University, Belgium
Mostafa Qorbani, Alborz University of Medical Sciences, Iran

Citation: Tabatabaei-Malazy, O., Kelishadi, R., Khashayar, P., Qorbani, M., eds. (2022). Association of Novel Anthropometric Indexes With Metabolic Syndrome. Lausanne: Frontiers Media SA. doi: 10.3389/978-2-88976-990-2

Table of Contents

frontiers | Frontiers in Endocrinology

TYPE Editorial
PUBLISHED 08 August 2022
DOI 10.3389/fendo.2022.951571

OPEN ACCESS

EDITED AND REVIEWED BY
Katherine Samaras,
St Vincent's Hospital Sydney, Australia

*CORRESPONDENCE
Ozra Tabatabaei-Malazy
tabatabaeiml@sina.tums.ac.ir
Mostafa Qorbani
mqorbani1379@yahoo.com

SPECIALTY SECTION
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

RECEIVED 24 May 2022
ACCEPTED 04 July 2022
PUBLISHED 08 August 2022

CITATION
Khashayar P, Tabatabaei-Malazy O, Qorbani M and Kelishadi R (2022) Editorial: Association of novel anthropometric indexes with metabolic syndrome.
Front. Endocrinol. 13:951571.
doi: 10.3389/fendo.2022.951571

Editorial: Association of novel anthropometric indexes with metabolic syndrome

Patricia Khashayar[1], Ozra Tabatabaei-Malazy[2*], Mostafa Qorbani[3*] and Roya Kelishadi[4]

[1]Center for Microsystems Technology, Imec & Ghent University, Zwijnaarde-Gent, Belgium, [2]Non-Communicable Diseases Research Center, Endocrinology and Metabolism Population Sciences Institute, Tehran University of Medical Sciences, Tehran, Iran, [3]Non-Communicable Diseases Research Center, Alborz University of Medical Sciences, Karaj, Iran, [4]Department of Pediatrics, Child Growth and Development Research Center, Research Institute for Primordial Prevention of Non-Communicable Disease, Isfahan University of Medical Sciences, Isfahan, Iran

KEYWORDS

obesity, anthropometry, waist-to-height ratio, waist circumference, body mass index, adiposity, metabolic syndrome, cardiometabolic risk factors

Editorial on the Research Topic
Association of novel anthropometric indexes with metabolic syndrome

The prevalence of being overweight and obese has been increasing in people living in both developed and developing countries (1). Obesity is linked to cardiometabolic risk factors, including insulin resistance, type 2 diabetes, hyperlipidemia, and hypertension, all of which can lead to cardiometabolic diseases (2, 3). Body Mass Index (BMI), defined as weight (kg)/height (m)2 has been used since 1970 to classify obesity in adults. One of the main limitations of BMI is the fact that it measures excess weight rather than excess fat (4). Recently, novel anthropometric measures, such as body shape index, hip index, body surface area, vertical trunk circumference, and visceral adiposity index, have been developed to overcome the BMI limitations (5). Due to the strong association between obesity and a long list of complications, which all are responsible for the increased risk of morbidity and mortality, the current Research Topic entitled "Association of Novel Anthropometric Indexes with Metabolic Syndrome" was initiated. Two main objectives of the current Research Topic are studying the link between novel anthropometric indices and cardiometabolic risk factors, and comparing the association between novel and traditional anthropometric indices and cardiometabolic risk factors.

Overall, the present Research topic collection is compiled from authors from various countries and examines the relationship between obesity and cardiometabolic risk factors such as insulin resistance, and type 2 diabetes.

In the first article of this Research Topic, by Xi et al., an association was found between being overweight and a 94% increased risk of hypertension in 30,617 twin individuals selected from the Chinese National Twin Registry (CNTR). They comment that common genetic predisposition and early-life environments are linked with obesity

and hypertension; the effect of the environment however proved to be less significant. In a study by Kim et al. non-diabetic subjects scheduled to undergo bariatric surgery, showed that increased adiposity, visceral fat area (VFA), and the homeostatic model for insulin resistance (HOMA-IR) were independent risk factors of polyneuropathy (PN). As for the participants with diabetes, however, the role of fibrosis detected as increased expression of fibrotic genes such as TIMP1 was more significant. These studies, along with many others, highlight the need for a more accurate tool to diagnose the high-risk obese individuals who may benefit from intervention. Despite the common usage of BMI in determining obesity, many studies have pointed out its shortcomings, such as incapability in distinguishing adipose mass from muscle and thus differentiating between low- and high-risk phenotypes of obesity. As a result, several articles in this Research Topic (Liu et al., Khonsari et al., Wang et al., Sun et al., Guo et al., Li et al.) investigated the association between novel anthropometric indexes and cardiometabolic risk factors for screening and the management of obesity.

In this regard, some articles have used a combination of existing markers with BMI to improve its efficacy. In the Liu et al. study, BMI is combined with waist circumference to assess 35,557 adults (51.1% women with a total mean age of 44.9 years) from the National Health and Nutrition Examination Survey (NHANES 1999-2014). The authors confirm waist-BMI ratio to be a promising marker for determining the high-risk phenotype of obesity. They conclude this ratio to be a better discriminatory proxy of mortality (all cause and cardiovascular mortality) compared with each of the markers alone.

Another novel anthropometric index is normal weight obesity (NWO), which shows normal BMI but with high-fat percentage. Khonsari et al. in their systematic review/meta-analysis study, pooled the results of the studies assessing the correlation between NWO and cardiometabolic risk factors. In their meta-analysis, a significant association is observed between NWO and cardiometabolic risk factors. They assert body fat percentage to be a better index than BMI for obesity risk assessment.

Liu et al. reported an increased risk of diabetes in hypertensive individuals with high adiposity index, defined by high waist-to-height ratio (WHtR). They suggest WHtR as a non-invasive, cost-saving public health tool to assess diabetes risk among the hypertensive adult population. As pediatric obesity is also associated with increased incidence of cardiometabolic risk factors, the usefulness of WHtR is also assessed in children and adolescents. In a cross-sectional population-based study, WHtR is used to identify the presence of left ventricular hypertrophy (LVH) and geometric (LVG) as markers of cardiac structural damage (Wang et al.). They report WHtR to be a similar or better predictive tool compared with BMI and both to be stronger than waist circumference in identifying children at risk of subclinical cardiac structural damage in adulthood.

Others, however, have focused on newer markers to assess body fat and replace BMI. In a systematic review, Sun et al. studied the value of tri-ponderal mass index, calculated as weight (kg)/height (m3) and defined it as a new indicator for adiposity and obesity-related cardiovascular risk factors (CVRFs) in children and adolescents. The results of this systematic review revealed that tri-ponderal mass index had a similar or better ability to predict body fat compared with BMI. Despite being similar to BMI in identifying MetS, tri-ponderal mass index is suggested to be a useful tool when used in combination with other indicators (e.g., BMI and waist circumference). In addition, limited evidence shows that tri-ponderal mass index does not perform better than BMI in identifying specific CVRFs, including insulin resistance, high blood pressure, dyslipidemia, and inflammation in children and adolescents, as well as CVRFs in adults.

Visceral adipose (VA) tissue defined as an "ectopic fat" can increase the risk of metabolic syndrome (MetS) by stimulating systemic inflammation, insulin resistance, and metabolic profiles. From among the VA tissue deposits, perirenal fat thickness can be easily measured using ultrasound, CT, and MRI scanning. In addition to its special anatomical structure, perirenal fat thickness can modulate the metabolism system. It can, thus, be a promising surrogate marker in identifying MetS. Guo et al. reported a significant association between perirenal fat thickness and MetS, as well as its components in individuals with newly diagnosed diabetes.

In the ninth article, Li et al. assessed the association between four anthropometric indexes including lipid accumulation products, waist-triglyceride index, visceral obesity index, triglyceride and glucose index, and MetS in the National Health and Nutrition Examination Survey (NHANES). The NHANES, a population-based study conducted between 1996-2006, is representative of the American adults. The authors concluded that the products of lipid accumulation were the strongest predictor of MetS in both sexes, suggesting them to be a more suitable tool to predict MetS in the clinical setting.

In another attempt, researchers looked into circulating biomarkers. In the article by Hu et al., increased serum levels of C1q/TNF-related protein 7 (CTRP7) were reported in MetS patients, suggesting they are a possible biomarker for detecting metabolic diseases. This is in line with previous animal studies that had linked CTRP7 with energy metabolism. In this study, serum levels of CTRP7 were confirmed to be significantly higher in MetS patients, showing a positive correlation with waist circumference, blood pressure, fasting blood glucose, 2h-blood glucose and triglyceride, but a negative correlation with HDL-C and adiponectin. They also confirmed a strong link between CTRP7 and metabolism-related genes and signal pathways by performing interventional studies (HEC, OGTT and lipid infusion) in healthy individuals.

In the eleventh article, an abnormal amino acid profile is reported in a cohort on the Chinese Han population (Sun et al.). This finding is the continuation of studies linking certain plasma amino acids with visceral obesity, insulin resistance, future development of diabetes, and cardiovascular diseases. Targeted liquid chromatography/tandem mass spectrometry (LC/MS) combined with principal component analysis (PCA) suggests a profile consisting of 12 amino acids (isoleucine, leucine, valine, tyrosine, tryptophan, glutamic acid, aspartic acid, alanine, histidine, methionine, asparagine, and proline) as capable of assessing and monitoring of MetS risk. Reduced taurine levels show promising results for early diagnosis of the disease.

Galectin-3-binding protein (GAL-3BP) is a glycoprotein known for its functions in innate immunity and is a potential mediator of adipose inflammation in obesity. The study by Zhen et al. showed a positive association between serum GAL-3BP and MetS in the Chinese population. This association is more significant among postmenopausal women.

Childhood obesity, regardless of age, results in adulthood obesity. One of the factors associated with early childhood obesity is maternal smoking. Hirai et al. reported urinary cotinine concentration as an accurate and quantitative marker for maternal smoking and childhood obesity among 89,617 mother-infant singleton pairs. They used the concentrations of cotinine in the mothers' urine, rather than their smoking classes, to predict childhood obesity in a dose-dependent manner.

Infrared thermography (IRT) is a non-contact and non-invasive technique, in which infrared emanated radiation from the body is captured and converted into temperature. This is then used for early screening of numerous diseases such as breast cancer and diabetic neuropathy. The skin temperature affected by local blood perfusion can, to some extent, determine tissue activity. For example, skin temperature of the anterior supraclavicular can reflect metabolic changes by detecting activated brown adipose tissue (BAT). Gao et al. assess the association of IRT and temperature distribution of the face, palms, feet, and the trunk area with MetS. They report a positive correlation between the temperature of face, palms, and dorsum of feet and the number of MetS components. As for the temperature of the anterior trunk, they show a negative association.

To conclude, all articles in this Research Topic collection propose novel anthropometric indexes as well as their association with MetS. This Research Topic has a great scientific impact and points out the importance of early detection of obesity in all ages and both genders. Moreover, they highlight current research gaps, paving the way for future research on the topic as well as practical strategies to overcome the existing shortcomings.

Author contributions

All authors listed have made a substantial, direct, and intellectual contribution to the work and approved it for publication.

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

References

1. Ng M, Fleming T, Robinson M, Thomson B, Graetz N, Margono C, et al. Global, regional, and national prevalence of overweight and obesity in children and adults during 1980–2013: A systematic analysis for the global burden of disease study 2013. *Lancet* (2014) 384:766–81. doi: 10.1016/S0140-6736(14)60460-8

2. Batty GD, Shipley MJ, Jarrett RJ, Breeze E, Marmot MG, Davey Smith G. Obesity and overweight in relation to disease-specific mortality in men with and without existing coronary heart disease in London: The original Whitehall study. *Heart* (2006) 92(7):886–92. doi: 10.1136/hrt.2005.072637

3. Bakhtiyari M, Kazemian E, Kabir K, Hadaegh F, Aghajanian S, Mardi P, et al. Contribution of obesity and cardiometabolic risk factors in developing cardiovascular disease: A population-based cohort study. *Sci Rep* (2022) 12:1544. doi: 10.1038/s41598-022-05536-w

4. Li M, McDermott RA. Using anthropometric indices to predict cardio-metabolic risk factors in Australian indigenous populations. *Diabetes Res Clin Pract* (2010) 87:401–6. doi: 10.1016/j.diabres.2009.12.004

5. Payab M, Qorbani M, Shahbal N, Motlagh ME, Hasani-Ranjbar S, Zahedi H, et al. Association of anthropometric indices with metabolic phenotypes of obesity in children and adolescents: The CASPIAN-V study. *Front Endocrinol (Lausanne)* (2019) 10:786. doi: 10.3389/fendo.2019.00786

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 31 May 2021
doi: 10.3389/fendo.2021.697437

Quotient of Waist Circumference and Body Mass Index: A Valuable Indicator for the High-Risk Phenotype of Obesity

Xiao-cong Liu, Yu Huang, Kenneth Lo, Yu-qing Huang, Ji-yan Chen and Ying-qing Feng *

Department of Cardiology, Guangdong Cardiovascular Institute, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou, China

OPEN ACCESS

Edited by:
Mostafa Qorbani, Alborz University of Medical Sciences, Iran

Reviewed by:
Hoda Zahedi, Tehran University of Medical Sciences, Iran
Mahdi Shadnoush, Shahid Beheshti University of Medical Sciences, Iran

***Correspondence:**
Ying-qing Feng
fyq1819@163.com

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *19 April 2021*
Accepted: *10 May 2021*
Published: *31 May 2021*

Citation:
Liu XC, Huang Y, Lo K, Huang YQ, Chen JY and Feng YQ (2021) Quotient of Waist Circumference and Body Mass Index: A Valuable Indicator for the High-Risk Phenotype of Obesity. Front. Endocrinol. 12:697437. doi: 10.3389/fendo.2021.697437

Objective: Measuring the body mass index (BMI) or waist circumference (WC) alone is insufficient for assessing possible health risks due to obesity. This study aimed to investigate whether the quotient of WC and BMI can be used as a proxy of the high-risk phenotype of obesity.

Methods: Data for analysis were derived from the National Health and Nutrition Examination Survey (NHANES 1999-2014). The Waist-BMI Ratio was defined as WC divided by BMI. The associations between Waist-BMI Ratio and mortality were estimated using Cox regression models. Restricted cubic spline and two-piecewise linear regression models were used to identify non-linear relationships. The discriminative abilities of different anthropometric measures were compared using receiver operating characteristic curves (ROC).

Results: This study is based on data from 35557 adults (51.1% female, mean age 44.9 years). During an average follow-up of 101.8 months, 3680 participants died, including 807 of cardiovascular causes. In fully adjusted models, Waist-BMI Ratio was independently associated with overall (hazard ratio [HR], 1.78; 95% confidence interval [CI], 1.48-2.13) and cardiovascular (HR, 1.77; 95% CI, 1.25-2.52) mortality. Spline analyses revealed that dose-response relationships existed between Waist-BMI Ratio and death. The mortality risk rises dramatically above the cut-off point of the Waist-BMI Ratio (HR, 3.22; 95% CI, 2.43-4.26 for overall mortality and HR, 3.07; 95% CI, 1.71-5.52 for cardiovascular mortality). ROC curve analysis suggested that Waist-BMI Ratio was a better discriminator of mortality (AUC 0.637 for overall and 0.639 for cardiovascular mortality) than BMI, WC, and waist-to-height ratio (Delong's test all P <0.001).

Conclusions: Waist-BMI Ratio was independently associated with overall and cardiovascular mortality in a J-shaped pattern, offering an immense potential risk marker for obesity in the clinical setting.

Keywords: body mass index, waist circumference, waist-BMI ratio, obesity, mortality

INTRODUCTION

Obesity has been recognized as one of the three gravest threats to human health and survival (1). It is responsible for 40% of cases of cardiovascular disease, most cases of type 2 diabetes, and more than 10% of gastrointestinal as well as urogenital cancer (2). Although increasing attention is being given to the problem, the prevalence of overweight and obesity has doubled since 1980 around the world and has shown a continuous increase in most countries (3). To reverse this growth and reduce the healthcare burden, accurate assessments of obesity are essential in order to identify high-risk individuals and thus implement appropriate behavior modifications and early therapeutic intervention.

Although multiple methods have been developed to assess obesity, each method has its own strengths and weaknesses. Imaging-based methods, such as dual x-ray absorptiometry and magnetic resonance imaging, can offer precise assessments and body fat quantifications (4). However, these technologically complex methods are too expensive and time-consuming for regular screening. The body mass index (BMI) is a simple anthropometric measure that has been routinely used to identify overweight individuals and estimate body fat (5). Nevertheless, BMI fails to describe body fat distribution and distinguish lean mass from fat mass, which has sparked the controversy related to "obesity paradox" (6–8). Prior studies have demonstrated that visceral adipose tissue (VAT) has an adverse impact on the cardiovascular and metabolic systems (4, 9), while certain types of peripheral fat could actually be metabolically, immunologically, and mechanically protective, and act as a cushion for potential health shocks (10, 11). Waist circumference (WC) and waist-to-height ratio (WtHR) are more accurate anthropometric measures of VAT but there are limitations with the use of either measure alone (12, 13).

Integration of BMI with WC/WtHR in clinical assessment has been recommended, as it may be able to discriminate the higher-risk phenotype of obesity (14, 15); it has been generally implemented by BMI stratification or introducing both variables into regression models. Studies have revealed that WC is positively correlated with mortality after adjustment for BMI, and patients with higher WtHR and lower BMI are at the highest risk of developing cardiovascular events (16–18). However, no study has yet focused on whether the quotient of WC and BMI (Waist-BMI Ratio) can be used to distinguish the high-risk phenotype of obesity. In this study, we evaluated the relationship of the Waist-BMI Ratio with cause-specific mortality and compared its predictive capacity with traditional anthropometric measures, including BMI, WC, and WtHR.

MATERIALS AND METHODS

Study Design and Participants

The study population was drawn from the National Health and Nutrition Examination Survey (NHANES). The NHANES is a national, cross-sectional, multistage, probability sampling survey used to provide representative samples of the non-institutionalized US resident population (19). The survey protocols were approved by the ethics review board of the National Center for Health Statistics, and informed consent was obtained from all participants. Our analysis involved all participants who were ≥ 18 years old from NHANES 1999-2000 to NHANES 2013-2014. After excluding those with missing data and with cancer at baseline, the final study size comprised 35557 individuals (**Figure 1**).

Anthropometric Measurements

Height, weight, and waist circumference were measured by trained personnel following a standard protocol (19). Waist circumferences were measured at the uppermost lateral border of the right ilium to the nearest 1 millimeter. BMI was calculated as weight in kilograms divided by height in meters squared. Waist-to-height ratio was calculated as waist circumference in centimeters divided by height in centimeters. Waist-BMI Ratio was defined as waist circumference in centimeters divided by BMI:

$$Waist-BMI\ ratio = waist\ circumference\,(cm)/body\ mass\ index\,(kg/m^2)\,.$$

Outcomes

Mortality status for NHANES participants was ascertained through probabilistic record matching with the National Death Index (20). The primary outcome of this study was overall mortality and the secondary outcome was cardiovascular mortality, assessed *via* underlying causes of death with International Classification of Diseases, 10th Revision (ICD-10) codes I00–I09, I11, I13, I20–I51, and I60–I69 (21).

Covariates

Demographic information including age, gender, and race/ethnicity (categorized as non-Hispanic white, non-Hispanic black, other Hispanic, Mexican American, and other) was collected by standard questionnaires. Past medical history, prescription medication use, and smoking status were self-reported. Blood pressure was measured by trained personnel. Lipid profile data were derived from laboratory measurements. The estimated glomerular filtration rate (eGFR) was calculated using the Modification of Diet in Renal Disease formula (22). Cardiovascular disease (CVD) was defined as self-reported coronary artery disease, angina, heart attack, or stroke. Diabetes was defined as a self-reported history of diabetes, diabetes medication use, fasting blood glucose level of at least 7.0 mmol/L, or a hemoglobin A1c (HbA1c) level of at least 6.5% (23). Hypertension was defined as a self-reported history of hypertension, receiving blood pressure control medication, systolic blood pressure of at least 140 mmHg, or diastolic blood pressure of at least 90 mmHg (24).

Statistical Analysis

To account for the complex survey design of NHANES, appropriate sampling weights were used to reconstitute data on the US non-institutionalized population. Participants were divided into quintile groups by the Waist-BMI Ratio. The mean or percentage, with standard error (SE), was provided by

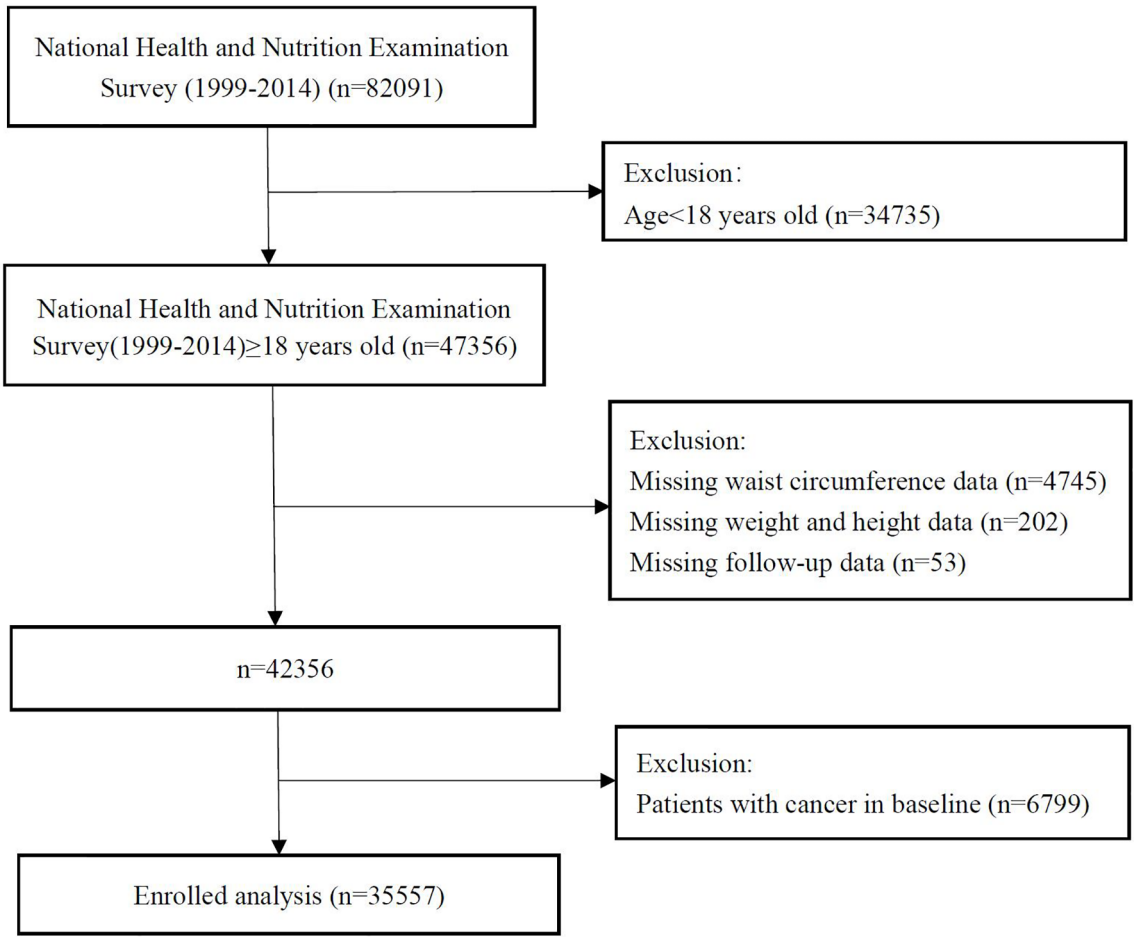


FIGURE 1 | Study cohort.

quintile groups. The linear trend for baseline characteristics was tested by linear or logistic regression whenever appropriate. Kaplan-Meier survival analyses were performed to evaluate the incidence rate of mortality among Waist-BMI Ratio groups, and discrepancies among groups were evaluated by log-rank test. Three sets of Cox proportional hazard models were constructed to evaluate associations with mortality from the date of medical examination to the date of either death or censoring (December 31, 2015), whichever came first. Tests for linear trends were also performed, by entering the mean value of each quintile group of Waist-BMI Ratio as a continuous variable. Restricted cubic regression splines were employed to examine the associations of different anthropometric measures with mortality. We used a two-piecewise linear regression model to evaluate the nonlinear relationships between Waist-BMI Ratio and mortality, and the optimal cut-off points were set by testing all possible values and selecting the cut-off values with the highest likelihood. The difference between one-line linear regression models and two-piecewise linear regression models was assessed by means of logarithmic likelihood ratio tests. To compare the effects of different anthropometric measures, we used the receiver operating characteristic (ROC) curve and the area under the ROC curve (AUC) to identify the ability of baseline BMI, WC, WtHR, and Waist-BMI Ratio to predict mortality events. The statistical significance of the differences in AUC was calculated with Delong's test. All analyses were conducted in R version 4.0.3 (R Foundation for Statistical Computing), including the rms, survey, pROC, and survminer packages. Two-sided $P < 0.05$ was regarded as a significant difference.

RESULTS

Baseline Characteristics

Data were analyzed for 35557 adults from the continuous NHANES survey (1999–2014); 51.1% were females, and the mean age was 44.9 years. Baseline characteristics stratified by Waist-BMI Ratio are presented in **Table 1**. Briefly, during the average follow-up period of 101.8 months, 3680 deaths were recorded and 807 of them were attributed to cardiovascular

TABLE 1 | Baseline characteristics according to Waist-BMI Ratio quintiles.

Variables	Total	Waist-BMI Ratio					*P* for trend
		Q1	Q2	Q3	Q4	Q5	
Number	35557	7112	7111	7111	7112	7111	
Age, years	44.9 (0.19)	42.7 (0.26)	42.4 (0.27)	44.1 (0.25)	45.7 (0.31)	49.4 (0.31)	<0.001
Gender-female, %	51.1 (0.27)	78.8 (0.60)	56.5 (0.86)	44.9 (0.67)	40.2 (0.62)	37.2 (0.85)	<0.001
Race, %							
Mexican American	8.6 (0.61)	10.5 (0.80)	11.1 (0.86)	9.4 (0.69)	7.6 (0.54)	4.7 (0.43)	<0.001
Other Hispanic	5.7 (0.54)	7.7 (0.77)	6.8 (0.72)	5.5 (0.54)	5.1 (0.54)	3.5 (0.46)	<0.001
Non-Hispanic White	67.7 (1.17)	56.6 (1.52)	62.5 (1.51)	68.2 (1.26)	72.1 (1.08)	78.2 (1.04)	<0.001
Non-Hispanic Black	11.6 (0.64)	20.6 (1.14)	13.5 (0.79)	10.2 (0.65)	7.8 (0.48)	6.6 (0.44)	<0.001
Other	6.4 (0.32)	4.6 (0.39)	6.1 (0.47)	6.7 (0.49)	7.5 (0.46)	7.0 (0.49)	<0.001
Smoking, %	46.2 (0.56)	36.7 (0.70)	42.8 (0.85)	45.6 (0.80)	49.9 (0.97)	55.4 (1.04)	<0.001
Systolic blood pressure, mmHg	121.6 (0.18)	122.5 (0.28)	121.1 (0.29)	121.1 (0.24)	121.3 (0.28)	122.2 (0.33)	0.548
Diastolic blood pressure, mmHg	71.0 (0.16)	71.7 (0.24)	71.2 (0.25)	71.1 (0.23)	70.8 (0.22)	70.2 (0.21)	<0.001
eGFR, mg/min/1.73m^2	87.6 (0.32)	90.5 (0.49)	89.1 (0.43)	86.7 (0.45)	86.2 (0.45)	85.9 (0.48)	<0.001
Total cholesterol, mg/dL	197.9 (0.37)	197.0 (0.59)	198.2 (0.66)	198.7 (0.69)	199.0 (0.65)	196.5 (0.74)	0.903
HDL-cholesterol, mg/dL	52.7 (0.16)	51.2 (0.27)	51.2 (0.28)	51.9 (0.25)	53.6 (0.26)	55.4 (0.26)	<0.001
Body Measures							
Waist circumference, cm	97.4 (0.18)	107.2 (0.36)	98.9 (0.30)	96.4 (0.24)	93.9 (0.23)	91.3 (0.20)	<0.001
Body mass index, kg/m2	28.4 (0.07)	36.1 (0.14)	30.0 (0.09)	27.7 (0.07)	25.8 (0.06)	23.3 (0.05)	<0.001
Waist-Height Ratio	0.58 (0.001)	0.66 (0.002)	0.59 (0.002)	0.57 (0.001)	0.55 (0.001)	0.53 (0.001)	<0.001
Waist-BMI Ratio	3.48 (0.003)	2.99 (0.003)	3.30 (0.001)	3.48 (0.001)	3.65 (0.001)	3.93 (0.003)	<0.001
Comorbidities, %							
Diabetes	11.2 (0.22)	14.9 (0.54)	11.6 (0.54)	10.9 (0.45)	8.9 (0.39)	10.2 (0.43)	<0.001
Hypertension	36.1 (0.46)	41.7 (0.86)	34.7 (0.77)	33.7 (0.70)	34.0 (0.70)	36.8 (0.84)	<0.001
Cardiovascular disease	6.4 (0.19)	5.1 (0.31)	5.2 (0.36)	6.1 (0.33)	6.9 (0.39)	8.9 (0.43)	<0.001
Medicine use, %							
Antihypertensive drugs	20.7 (0.4)	25.7 (0.81)	18.9 (0.64)	19.1 (0.60)	18.5 (0.64)	21.7 (0.70)	<0.001
Hypoglycemic agents	6.0 (0.17)	8.2 (0.41)	6.3 (0.42)	5.5 (0.31)	4.5 (0.29)	5.5 (0.27)	<0.001
Lipid-lowering drugs	11.1 (0.28)	9.3 (0.49)	9.7 (0.47)	11.1 (0.50)	12.0 (0.51)	13.4 (0.58)	<0.001
Antiplatelet drugs	1.2 (0.08)	1.0 (0.17)	0.9 (0.11)	1.0 (0.17)	1.3 (0.16)	2.0 (0.19)	<0.001
Outcomes, %							
Cardiovascular disease mortality	1.5 (0.07)	0.9 (0.11)	1.0 (0.13)	1.1 (0.11)	1.6 (0.17)	2.6 (0.22)	<0.001
Overall mortality	7.2 (0.21)	5.0 (0.33)	4.8 (0.28)	6.0 (0.31)	7.7 (0.39)	12.3 (0.52)	<0.001

Q, quintiles; eGFR, estimated glomerular filtration rate; HDL, high density lipoprotein.
Values are mean or percent with standard error.
P for trend was tested by linear or logistic regression.

disease. The ranges of Waist-BMI Ratio were: Q1: 1.80-3.20; Q2: 3.20-3.39; Q3: 3.39-3.56; Q4:3.56-3.75; Q5: 3.75-5.56. Individuals in the upper Waist-BMI Ratio quintiles tended to be older, male, and have higher rates of smoking and CVD. High density lipoprotein (HDL) cholesterol levels and mortality risk tended to be higher with the increase of Waist-BMI Ratio while eGFR and WtHR were decreased with increasing Waist-BMI Ratio quintiles (all *P* for trend < 0.001).

The Association of Waist-BMI Ratio and Other Anthropometric Measures With Overall and Cardiovascular Mortality

As shown in **Table 2**, the crude incidence rate per 1000 person-years of overall and cardiovascular mortality rose dramatically with increasing Waist-BMI Ratio categories. Kaplan-Meier curves for mortality showed significant differences among the Waist-BMI Ratio quintile groups (**Figure 2**, both log-rank $P < 0.001$). After adjustment for all covariables, including age, gender, race, smoking, systolic blood pressure, HDL-cholesterol, total cholesterol, eGFR, hypertension, diabetes, CVD, antihypertensive drugs, hypoglycemic agents, lipid-lowering drugs, and antiplatelet drugs, Waist-BMI Ratio was positively associated with overall (hazard ratio [HR], 1.78; 95% confidence interval [CI], 1.48-2.13; $P < 0.001$) and cardiovascular (HR, 1.77; 95% CI, 1.25-2.52; $P = 0.001$) mortality as a continuous linear variable. When using the lowest Waist-BMI Ratio quintile (Q1) as the reference, significant association with overall death can be seen for the upper quintile (HR, 1.40; 95% CI, 1.17-1.67; $P < 0.001$). However, the relationship between the highest Waist-BMI Ratio quintile (Q5) and cardiovascular death was not significant (HR, 1.34; 95% CI, 0.95-1.88; $P = 0.094$).

Restricted cubic splines (**Figure 3**) demonstrated a J-shaped relationship of Waist-BMI Ratio with overall (non-linear $P < 0.001$) and cardiovascular mortality (non-linear $P = 0.017$), while other anthropometric measures, including BMI, WC, and WtHR, showed asymmetrical U-shaped relationships with mortality (all non-linear $P < 0.001$). Significant differences were detected between the linear regression models and the two-piecewise regression models (log-likelihood ratio test $P<0.001$ for overall

TABLE 2 | Multivariate Cox regression analysis of Waist-BMI Ratio with cause-specific mortality.

	Overall mortality				Cardiovascular mortality			
	Event rate/ 1000 person-years	Model I	Model II	Model III	Event rate/ 1000 person-years	Model I	Model II	Model III
Waist-BMI Ratio								
As continuous variables	13.65	3.55 (3.08, 4.09) <0.001	1.60 (1.34, 1.90) <0.001	1.78 (1.48, 2.13) <0.001	2.84	4.11 (3.07, 5.51) <0.001	1.30 (0.91, 1.85) 0.153	1.77 (1.25, 2.52) 0.001
As categorical variables (quintiles)								
Q1	7.47	Reference	Reference	Reference	1.37	Reference	Reference	Reference
Q2	8.30	0.97 (0.82, 1.15) 0.760	0.89 (0.77, 1.03) 0.105	0.95 (0.81, 1.11) 0.525	1.76	1.09 (0.76, 1.57) 0.631	0.91 (0.65, 1.28) 0.587	1.05 (0.73, 1.51) 0.796
Q3	11.12	1.23 (1.06, 1.43) 0.006	0.92 (0.80, 1.06) 0.262	0.96 (0.82, 1.12) 0.579	1.97	1.19 (0.93, 1.54) 0.171	0.74 (0.56, 0.99) 0.042	0.87 (0.66, 1.15) 0.326
Q4	14.80	1.57 (1.36, 1.83) <0.001	1.03 (0.89, 1.18) 0.710	1.10 (0.95, 1.28) 0.198	3.30	1.77 (1.29, 2.43) <0.001	0.90 (0.64, 1.26) 0.539	1.19 (0.85, 1.68) 0.312
Q5	27.84	2.64 (2.28, 3.06) <0.001	1.27 (1.08, 1.51) 0.005	1.40 (1.17, 1.67) <0.001	6.11	3.02 (2.30, 3.96) <0.001	1.02 (0.73, 1.42) 0.913	1.34 (0.95, 1.88) 0.094
P for trend		<0.001	<0.001	<0.001		<0.001	0.490	0.026

Data are hazard ratios (HRs), 95% confidence intervals (95% CIs), and P-value.
Model I adjust for none.
Model II adjust for age, gender, and race.
Model III adjust for age, gender, race, smoking, SBP, HDL-cholesterol, total cholesterol, eGFR, comorbidities (hypertension, diabetes, and cardiovascular disease), and medicine use (antihypertensive drugs, hypoglycemic agents, lipid-lowering drugs, and antiplatelet drugs).

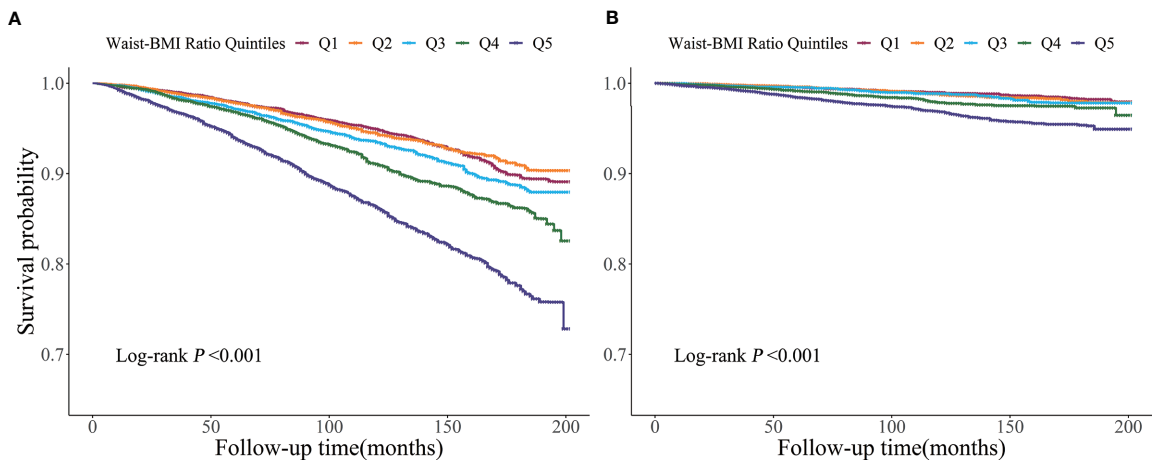


FIGURE 2 | Kaplan-Meier survival curve for overall **(A)** and cardiovascular **(B)** mortality by Waist-BMI Ratio quintiles.

mortality and *P*=0.008 for cardiovascular mortality). The cut-off points were estimated by piecewise regression models to be at a Waist-BMI Ratio of 3.72 for overall mortality and 3.66 for cardiovascular mortality (**Table 3**). Above the cut-off points, the risk of overall and cardiovascular death rose steeply with the increase of Waist-BMI Ratio (HR, 3.22; 95% CI, 2.43-4.26; $P < 0.001$ and HR, 3.07; 95% CI, 1.71-5.52; $P < 0.001$, respectively). Nonetheless, no significant association was found below the cut-off points (HR, 1.16; 95% CI, 0.93-1.44; $P = 0.188$ and HR,1.00; 95% CI, 0.58-1.73; $P = 0.999$, respectively).

The Predictive Value of Waist-BMI Ratio and Other Anthropometric Measures in Overall and Cardiovascular Mortality

The ROC curve analysis comparing the predictive ability of different anthropometric measures demonstrated that Waist-BMI Ratio was the strongest predictor of overall mortality (AUC, 0.637; 95% CI, 0.627-0.647). As shown in **Figure 4**, the AUC of BMI, WC, and WtHR for predicting overall mortality were 0.523 (95% CI, 0.513-0.533), 0.552 (95% CI, 0.543-0.562), and 0.572 (95% CI, 0.562-0.581), respectively. Similar results

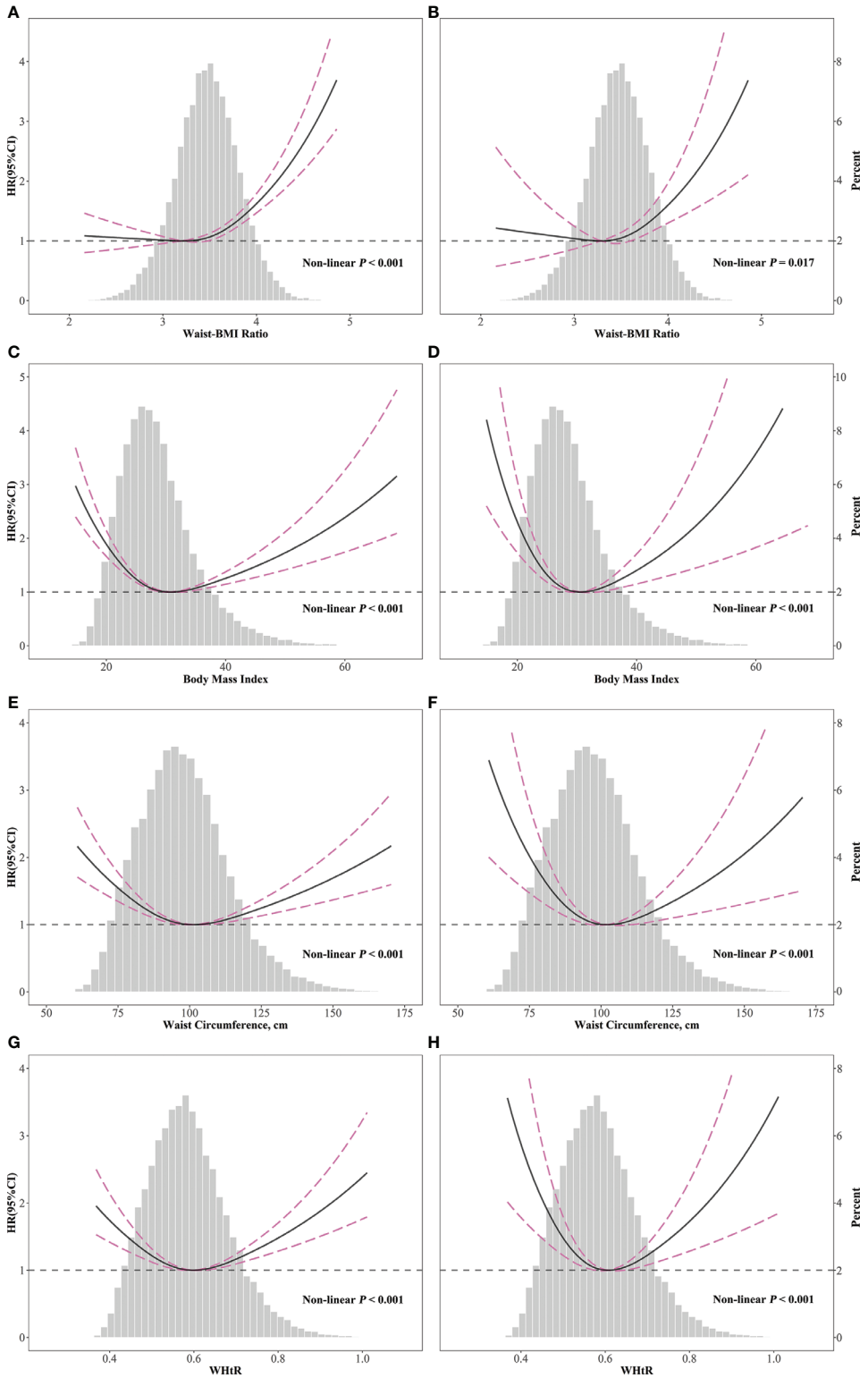


FIGURE 3 | Spline analyses of overall **(A, C, E, G)** and cardiovascular **(B, D, F, H)** mortality by Waist-BMI Ratio **(A, B)**, body mass index **(C, D)**, waist circumference **(E, F)**, and Waist-Height Ratio **(G, H)** and the probability distribution histogram is represented in the background. (Spline analyses were adjusted for age, gender, race, smoking, SBP, HDL-cholesterol, total cholesterol, eGFR, hypertension, diabetes, and cardiovascular disease, antihypertensive drugs, hypoglycemic agents, lipid-lowering drugs, and antiplatelet drugs).

TABLE 3 | The results of two-piecewise linear regression model between Waist-BMI Ratio and cause-specific mortality.

	Overall mortality	Cardiovascular mortality
Cutoff value	3.72	3.66
<Cut-off value	1.16 (0.93, 1.44) 0.188	1.00 (0.58, 1.73) 0.999
≥Cut-off value	3.22 (2.43, 4.26) <0.001	3.07 (1.71, 5.52) <0.001
P for log likelihood ratio test	<0.001	0.008

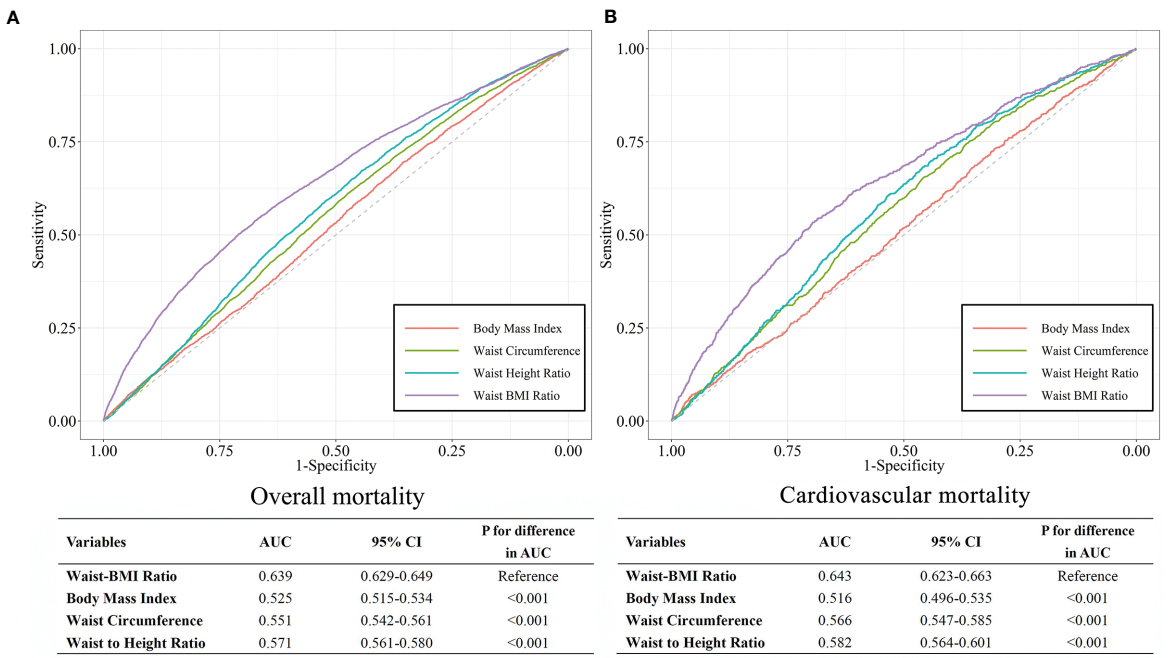
Data are hazard ratios (HRs), 95% confidence intervals (95% CIs), and P-value.
The two-piecewise linear regression model were adjusted for age, gender, race, smoking, SBP, HDL-cholesterol, total cholesterol, eGFR, comorbidities (hypertension, diabetes, and cardiovascular disease), and medicine use (antihypertensive drugs, hypoglycemic agents, lipid-lowering drugs, and antiplatelet drugs).

were observed for cardiovascular death. When compared with other indexes, Waist-BMI Ratio showed significantly better performance (AUC, 0.643; 95% CI, 0.623-0.663) than BMI (AUC, 0.516; 95% CI, 0.496-0.535), WC (AUC, 0.566; 95% CI, 0.547-0.585), and WtHR (AUC, 0.582; 95% CI, 0.564-0.601) in predicting cardiovascular death (all *P* for difference in AUC < 0.001). The optimal value of the Waist-BMI Ratio was 3.60 for predicting overall mortality, with a sensitivity of 53.8% and a specificity of 67.4%; as the optimal value was 3.64 for predicting cardiovascular mortality, with a sensitivity of 53.4% and specificity of 69.6%.

DISCUSSION

In this study, we retrospectively investigated the relationship between a newly defined anthropometric measure and mortality. The results demonstrated that the Waist-BMI Ratio was positively associated with overall and cardiovascular mortality in a J-shaped pattern. Compared with traditional obesity indices, the Waist-BMI Ratio can more adequately predict the risk of death and could be a valuable indicator of the higher-risk phenotype of obesity.

For decades, BMI has been used as an indicator of obesity and has been introduced into various predictive models as a cardiovascular risk factor (25–27). However, a proportion of people with normal BMI have a series of metabolic risk factors, and may be described as "metabolically with obesity but of normal weight" (28), while some individuals with obesity seem to be protected from or more resistant to the development of metabolic abnormalities, and are known as "metabolically healthy but with obesity" (29). One of the reasons may lie in the inability to describe visceral fat and ectopic fat deposition (8), because information from a single anthropometric measure cannot provide sufficient insights into body fat distribution (12). Adverse metabolic effects of excess body fat, including insulin resistance and dyslipidemia, are mainly linked to dysfunctional abdominal subcutaneous adipose tissue and



Variables	AUC	95% CI	P for difference in AUC
Waist-BMI Ratio	0.639	0.629-0.649	Reference
Body Mass Index	0.525	0.515-0.534	<0.001
Waist Circumference	0.551	0.542-0.561	<0.001
Waist to Height Ratio	0.571	0.561-0.580	<0.001

Variables	AUC	95% CI	P for difference in AUC
Waist-BMI Ratio	0.643	0.623-0.663	Reference
Body Mass Index	0.516	0.496-0.535	<0.001
Waist Circumference	0.566	0.547-0.585	<0.001
Waist to Height Ratio	0.582	0.564-0.601	<0.001

FIGURE 4 | ROC curves for Body mass index, Waist circumference, Waist to Height Ratio, and Waist-BMI Ratio for predicting overall **(A)** and cardiovascular **(B)** mortality.

visceral adipose tissue accumulation (9). Experimental models have shown that visceral adipose tissue produces potentially proinflammatory adipokines and macrophage signals, which may be involved in myocardial hypertrophy, fibrosis, and injury (30).

A meta-analysis has revealed that BMI fails to identify half of individuals with excess body fat (31). Chanchal et al. found that the joint use of BMI and WtHR could be conducive to recognizing patients with the highest risk of the composite outcomes (16). A Consensus Statement from the IAS and ICCR Working Group claimed that, although waist circumference is closely linked to overall and cardiovascular death, the full strength of these associations is revealed only after adjustment for BMI (12). However, as a statistical term, "adjustment" might not be easy to understand and use in clinical applications, which limits the combination of WC and BMI. Clinicians have been recommended to evaluate WC variation among patients with similar BMI values, whereas the current obesity-risk classification system still uses the same WC threshold values for all BMI categories (32, 33).

Consistent with previous studies, our present analysis illustrated that individuals in the upper Waist-BMI Ratio quantile, which corresponds to individuals with high WC but low BMI, had the highest risk of mortality. Moreover, patients with low absolute BMI and WC are prone to a higher risk of mortality (34, 35). Studies have indicated that underweight is correlated with undernutrition, inflammation, and other underlying wasting diseases that potentially explain the enhanced risk of death (11, 36, 37). Therefore, U-shape relationships and an "obesity paradox" have often been reported (6, 38). This flaw is circumvented by using Waist-BMI Ratio as an adiposity indicator and thus J-shape relationships were observed. Meanwhile, people of normal weight generally pay less attention to their health indices and do not take preventive measures against obesity-related diseases. Compared with costly and time-consuming imaging-based methods, anthropometric methods such as the Waist-BMI Ratio form a more convenient, comprehensible, and even home self-testing monitoring system for early identification of high-risk individuals and for disease prevention.

Our study had several notable advantages. First, the study was based on the NHANES dataset, a nationally representative survey with rigorous methodology and comprehensive quality control procedures; the large sample size was sufficient to provide good statistical power. Second, our analysis was adjusted for study weights and the complex survey design to reduce estimation errors. Third, the newly defined parameter was easily obtained and calculated, with a clear and unilateral risk threshold. Nevertheless, several limitations pertain to our study. First, we were unable to obtain accurate information about body composition. Therefore, we could not calculate the correlation coefficient between Waist-BMI Ratio and abdominal or visceral adipose tissue. Second, definitive causal inferences cannot be drawn because of the observational nature of this study. Third, although our analyses controlled for important confounding variables, the possibility of residual confounding variables remains. For example, we were unable to adjust for alcohol consumption and physical activity level due to missing covariate data. Fourth, death certificates may not precisely represent the real cause of death. Fifth, NHANES data are representative of the United States population, thus probably limiting the applicability of our results to other regions and ethnic populations.

CONCLUSION

As a newly defined anthropometric measure, Waist-BMI Ratio was independently associated with overall and cardiovascular mortality after mutual adjustment. Compared with other traditional anthropometric measures, Waist-BMI Ratio had a better predictive ability and a more certain risk threshold value for mortality. Because it is convenient, easy to access, and virtually free cost, Waist-BMI Ratio can be used as a valuable indicator for the high-risk phenotype of obesity.

DATA AVAILABILITY STATEMENT

Publicly available datasets were analyzed in this study. This data can be found here: https://www.cdc.gov/nchs/nhanes/index.htm.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Institutional Review Board of the Centers for Disease Control and Prevention (Protocol 98–12, 2005–06 and 2011–17). The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

Conceptualization, XCL and YQH. Methodology, XCL. Validation, YQF, YQH and JYC. Formal Analysis, XCL. Investigation, KL. Resources, YQF. Data Curation, XCL. Writing — Original Draft Preparation, XCL. Writing – Review and Editing, YH. Visualization, XCL. Supervision, JYC. Project Administration, YQF. Funding Acquisition, YQF. All authors contributed to the article and approved the submitted version.

FUNDING

This research was funded by the National Key Research and Development Program of China (No. 2017YFC1307603), the Science and Technology Plan Program of Guangzhou (No. 201803040012), the Key Area R&D Program of Guangdong Province (No. 2019B020227005), Guangdong Provincial People's

Hospital Clinical Research Fund (Y012018085), the Fundamental and Applied Basic Research Foundation Project of Guangdong Province (2020A1515010738), and the Climbing Plan of Guangdong Provincial People's Hospital (DFJH2020022).

ACKNOWLEDGMENTS

We are grateful to Dr. YS Liu for her invaluable advices that helped improve this article.

REFERENCES

1. Swinburn BA, Kraak VI, Allender S, Atkins VJ, Baker PI, Bogard JR, et al. The Global Syndemic of Obesity, Undernutrition, and Climate Change: The Lancet Commission Report. *Lancet* (2019) 393:791–846. doi: 10.1016/s0140-6736(18)32822-8
2. Kumanyika S, Dietz WH. Solving Population-Wide Obesity - Progress and Future Prospects. *N Engl J Med* (2020) 383:2197–200. doi: 10.1056/NEJMp2029646
3. Collaborators TGO. Health Effects of Overweight and Obesity in 195 Countries Over 25 Years. *N Engl J Med* (2017) 377:13–27. doi: 10.1056/NEJMoa1614362
4. Neeland IJ, Poirier P, Despres JP. Cardiovascular and Metabolic Heterogeneity of Obesity: Clinical Challenges and Implications for Management. *Circulation* (2018) 137:1391–406. doi: 10.1161/circulationaha.117.029617
5. World Health Organization. *Obesity*. Available at: https://www.who.int/health-topics/obesity (Accessed January 15, 2021).
6. Antonopoulos AS, Oikonomou EK, Antoniades C, Tousoulis D. From the BMI Paradox to the Obesity Paradox: The Obesity-Mortality Association in Coronary Heart Disease. *Obes Rev* (2016) 17:989–1000. doi: 10.1111/obr.12440
7. Iliodromiti S, Celis-Morales CA, Lyall DM, Anderson J, Gray SR, Mackay DF, et al. The Impact of Confounding on the Associations of Different Adiposity Measures With the Incidence of Cardiovascular Disease: A Cohort Study of 296 535 Adults of White European Descent. *Eur Heart J* (2018) 39:1514–20. doi: 10.1093/eurheartj/ehy057
8. Elagizi A, Kachur S, Lavie CJ, Carbone S, Pandey A, Ortega FB, et al. An Overview and Update on Obesity and the Obesity Paradox in Cardiovascular Diseases. *Prog Cardiovasc Dis* (2018) 61:142–50. doi: 10.1016/j.pcad.2018.07.003
9. Piche ME, Poirier P, Lemieux I, Despres JP. Overview of Epidemiology and Contribution of Obesity and Body Fat Distribution to Cardiovascular Disease: An Update. *Prog Cardiovasc Dis* (2018) 61:103–13. doi: 10.1016/j.pcad.2018.06.004
10. Tchkonia T, Thomou T, Zhu Y, Karagiannides I, Pothoulakis C, Jensen Michael D, et al. Mechanisms and Metabolic Implications of Regional Differences Among Fat Depots. *Cell Metab* (2013) 17:644–56. doi: 10.1016/j.cmet.2013.03.008
11. Liu XC, Liu L, Yu YL, Huang JY, Chen CL, Lo K, et al. The Association of Subscapular Skinfold With All-Cause, Cardiovascular and Cerebrovascular Mortality. *Risk Manag Healthc Policy* (2020) 13:955–63. doi: 10.2147/rmhp.S262300
12. Ross R, Neeland IJ, Yamashita S, Shai I, Seidell J, Magni P, et al. Waist Circumference as a Vital Sign in Clinical Practice: A Consensus Statement From the IAS and ICCR Working Group on Visceral Obesity. *Nat Rev Endocrinol* (2020) 16:177–89. doi: 10.1038/s41574-019-0310-7
13. Khoury M, Manlhiot C, McCrindle BW. Role of the Waist/Height Ratio in the Cardiometabolic Risk Assessment of Children Classified by Body Mass Index. *J Am Coll Cardiol* (2013) 62:742–51. doi: 10.1016/j.jacc.2013.01.026
14. Wharton S, Lau DCW, Vallis M, Sharma AM, Biertho L, Campbell-Scherer D, et al. Obesity in Adults: A Clinical Practice Guideline. *CMAJ* (2020) 192:E875–E91. doi: 10.1503/cmaj.191707
15. Clinical Guidelines on the Identification, Evaluation, and Treatment of Overweight and Obesity in Adults–The Evidence Report. National Institutes of Health. *Obes Res* (1998) 6 Suppl 2:51s–209s. doi: 10.1002/j.1550-8528.1998.tb00690.x
16. Chandramouli C, Tay WT, Bamadhaj NS, Tromp J, Teng TK, Yap JJL, et al. Association of Obesity With Heart Failure Outcomes in 11 Asian Regions: A Cohort Study. *PloS Med* (2019) 16:e1002916. doi: 10.1371/journal.pmed.1002916
17. Janssen I, Katzmarzyk PT, Ross R. Body Mass Index Is Inversely Related to Mortality in Older People After Adjustment for Waist Circumference. *J Am Geriatr Soc* (2005) 53:2112–8. doi: 10.1111/j.1532-5415.2005.00505.x
18. Cerhan JR, Moore SC, Jacobs EJ, Kitahara CM, Rosenberg PS, Adami HO, et al. A Pooled Analysis of Waist Circumference and Mortality in 650,000 Adults. *Mayo Clin Proc* (2014) 89:335–45. doi: 10.1016/j.mayocp.2013.11.011
19. Centers for Disease Control and Prevention. *Nhanes - National Health and Nutrition Examination Survey Homepage* (2020). Available at: https://www.cdc.gov/nchs/nhanes/index.htm (Accessed March 15, 2020).
20. National Center for Health Statistics. *The Linkage of National Center for Health Statistics Survey Data to the National Death Index—2015 Linked Mortality File (LMF): Methodology Overview and Analytic Considerations. Centers for Disease Control and Prevention* (2019) Geneva,Switzerland:World Health Organization. Available at: https://www.cdc.gov/nchs/data/datalinkage/LMF2015_Methodology_Analytic_Considerations.pdf (Accessed March 15, 2020).
21. Liu X-C, He G-D, Lo K, Huang Y-Q, Feng Y-Q. The Triglyceride-Glucose Index, An Insulin Resistance Marker, Was Non-Linear Associated With All-Cause and Cardiovascular Mortality in the General Population. *Front Cardiovasc Med* (2021) 7:628109. doi: 10.3389/fcvm.2020.628109
22. Inker LA, Schmid CH, Tighiouart H, Eckfeldt JH, Feldman HI, Greene T, et al. Estimating Glomerular Filtration Rate From Serum Creatinine and Cystatin C. *N Engl J Med* (2012) 367:20–9. doi: 10.1056/NEJMoa1114248
23. Chamberlain JJ, Johnson EL, Leal S, Rhinehart AS, Shubrook JH, Peterson L. Cardiovascular Disease and Risk Management: Review of the American Diabetes Association Standards of Medical Care in Diabetes 2018. *Ann Intern Med* (2018) 168:640–50. doi: 10.7326/m18-0222
24. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL Jr, et al. The Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure: The JNC 7 Report. *JAMA* (2003) 289:2560–72. doi: 10.1001/jama.289.19.2560
25. Wu Y, Liu X, Li X, Li Y, Zhao L, Chen Z, et al. Estimation of 10-Year Risk of Fatal and Nonfatal Ischemic Cardiovascular Diseases in Chinese Adults. *Circulation* (2006) 114:2217–25. doi: 10.1161/CIRCULATIONAHA.105.607499
26. Hajifathalian K, Ueda P, Lu Y, Woodward M, Ahmadvand A, Aguilar-Salinas CA, et al. A Novel Risk Score to Predict Cardiovascular Disease Risk in National Populations (Globorisk): A Pooled Analysis of Prospective Cohorts and Health Examination Surveys. *Lancet Diabetes Endocrinol* (2015) 3:339–55. doi: 10.1016/s2213-8587(15)00081-9
27. Hippisley-Cox J, Coupland C, Brindle P. Development and Validation of QRISK3 Risk Prediction Algorithms to Estimate Future Risk of Cardiovascular Disease: Prospective Cohort Study. *BMJ* (2017) 357:j2099. doi: 10.1136/bmj.j2099
28. Hinnouho GM, Czernichow S, Dugravot A, Nabi H, Brunner EJ, Kivimaki M, et al. Metabolically Healthy Obesity and the Risk of Cardiovascular Disease and Type 2 Diabetes: The Whitehall II Cohort Study. *Eur Heart J* (2015) 36:551–9. doi: 10.1093/eurheartj/ehu123
29. Primeau V, Coderre L, Karelis AD, Brochu M, Lavoie ME, Messier V, et al. Characterizing the Profile of Obese Patients Who Are Metabolically Healthy. *Int J Obes (Lond)* (2011) 35:971–81. doi: 10.1038/ijo.2010.216
30. Murase T, Hattori T, Ohtake M, Abe M, Amakusa Y, Takatsu M, et al. Cardiac Remodeling and Diastolic Dysfunction in DahlS.Z-Lepr(fa)/Lepr(fa) Rats: A New Animal Model of Metabolic Syndrome. *Hypertens Res* (2012) 35:186–93. doi: 10.1038/hr.2011.157
31. Okorodudu DO, Jumean MF, Montori VM, Romero-Corral A, Somers VK, Erwin PJ, et al. Diagnostic Performance of Body Mass Index to Identify Obesity as Defined by Body Adiposity: A Systematic Review and Meta-Analysis. *Int J Obes (Lond)* (2010) 34:791–9. doi: 10.1038/ijo.2010.5
32. Despres JP. Excess Visceral Adipose Tissue/Ectopic Fat the Missing Link in the Obesity Paradox? *J Am Coll Cardiol* (2011) 57:1887–9. doi: 10.1016/j.jacc.2010.10.063
33. World Health Organisation. *Obesity: Preventing and Managing the Global Epidemic: Report of a WHO Consultation (World Health Organisation Technical Report Series 894)*. Geneva, Switzerland: WHO (2000).

34. Bhaskaran K, dos-Santos-Silva I, Leon DA, Douglas IJ, Smeeth L. Association of BMI With Overall and Cause-Specific Mortality: A Population-Based Cohort Study of 3·6 Million Adults in the UK. *Lancet Diabetes Endocrinol* (2018) 6:944–53. doi: 10.1016/s2213-8587(18)30288-2
35. Chen Y, Yang Y, Jiang H, Liang X, Wang Y, Lu W. Associations of BMI and Waist Circumference With All-Cause Mortality: A 22-Year Cohort Study. *Obesity (Silver Spring)* (2019) 27:662–9. doi: 10.1002/oby.22423
36. Wirth R, Streicher M, Smoliner C, Kolb C, Hiesmayr M, Thiem U, et al. The Impact of Weight Loss and Low BMI on Mortality of Nursing Home Residents - Results From the Nutrition Day in Nursing Homes. *Clin Nutr* (2016) 35:900–6. doi: 10.1016/j.clnu.2015.06.003
37. Nakajima K, Yamaoka H, Morita K, Ebata M, Eguchi S, Muneyuki T, et al. Elderly People With Low Body Weight May Have Subtle Low-Grade Inflammation. *Obesity (Silver Spring)* (2009) 17:803–8. doi: 10.1038/oby.2008.596
38. Lavie CJ, Laddu D, Arena R, Ortega FB, Alpert MA, Kushner RF. Healthy Weight and Obesity Prevention: JACC Health Promotion Series. *J Am Coll Cardiol* (2018) 72:1506–31. doi: 10.1016/j.jacc.2018.08.1037

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 05 October 2021
doi: 10.3389/fendo.2021.743962

The Roles of Genetic and Early-Life Environmental Factors in the Association Between Overweight or Obesity and Hypertension: A Population-Based Twin Study

Yu'e Xi[1], Wenjing Gao[1], Ke Zheng[1], Jun Lv[1], Canqing Yu[1], Shengfeng Wang[1], Tao Huang[1], Dianjianyi Sun[1], Chunxiao Liao[1], Yuanjie Pang[1], Zengchang Pang[2], Min Yu[3], Hua Wang[4], Xianping Wu[5], Zhong Dong[6], Fan Wu[7], Guohong Jiang[8], Xiaojie Wang[9], Yu Liu[10], Jian Deng[11], Lin Lu[12], Weihua Cao[1]* and Liming Li[1]*

[1] Department of Epidemiology and Biostatistics, School of Public Health, Peking University, Beijing, China, [2] Qingdao Municipal Center for Disease Control and Prevention , Qingdao, China, [3] Zhejiang Provincial Center for Disease Control and Prevention, Hangzhou, China, [4] Jiangsu Provincial Center for Disease Control and Prevention, Nanjing, China, [5] Sichuan Center for Disease Control and Prevention, Chengdu, China, [6] Beijing Center for Disease Prevention and Control, Beijing, China, [7] Shanghai Municipal Center for Disease Control and Prevention, Shanghai, China, [8] Tianjin Centers for Disease Control and Prevention, Tianjin, China, [9] Qinghai Center for Diseases Prevention and Control, Xining, China, [10] Heilongjiang Provincial Center for Disease Control and Prevention, Harbin, China, [11] Handan Center for Disease Control and Prevention, Handan, China, [12] Yunnan Center for Disease Control and Prevention, Kunming, China

OPEN ACCESS

Edited by:
Patricia Khashayar,
Ghent University, Belgium

Reviewed by:
Xiaocao Tian,
Qingdao Municipal Center for Disease Control and Prevention, China
Xiao-cong Liu,
Guangdong Academy of Medical Sciences, China

***Correspondence:**
Wenjing Gao
pkuepigwj@126.com
Weihua Cao
caoweihua60@163.com

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: 19 July 2021
Accepted: 15 September 2021
Published: 05 October 2021

Citation:
Xi Y'e, Gao W, Zheng K, Lv J, Yu C, Wang S, Huang T, Sun D, Liao C, Pang Y, Pang Z, Yu M, Wang H, Wu X, Dong Z, Wu F, Jiang G, Wang X, Liu Y, Deng J, Lu L, Cao W and Li L (2021) The Roles of Genetic and Early-Life Environmental Factors in the Association Between Overweight or Obesity and Hypertension: A Population-Based Twin Study. Front. Endocrinol. 12:743962. doi: 10.3389/fendo.2021.743962

Aims/Hypothesis: We aimed to explore whether and to what extent overweight or obesity could increase the risk of hypertension, and further to estimate the roles of genetic and early-life familial environmental factors in their association.

Methods: This prospective twin study was based on the Chinese National Twin Registry (CNTR), which collected information from self-report questionnaires. We conducted unmatched case-control analysis to examine the association between overweight or obesity and hypertension. And further to explore whether genetics and familiar environments shared within a twin pair, accounted for their association *via* co-twin matched case-control design. Generalized estimating equation (GEE) models and conditional logistic regressions were used in the unmatched and matched analyses, respectively. Then, we used logistic regressions to test the difference in odds ratios (ORs) between the unmatched and matched analyses. Finally, through bivariate twin model, the roles of genetic and environmental factors in the body mass index (BMI)- hypertension association were estimated.

Results: Overall, we included a total of 30,617 twin individuals, of which 7533 (24.6%) twin participants were overweight or obesity and 757 (2.5%) developed hypertension during a median follow-up time of 4.4 years. In the GEE model, overweight or obesity was associated with a 94% increased risk of hypertension (OR=1.94, 95% confidence interval (CI): 1.64~2.30). In the conditional logistic regression, the multi-adjusted OR was 1.80 (95% CI: 1.18~2.74). The difference in OR between unmatched and matched analyses

was significant (P=0.016). Specifically, overweight or obesity was not associated with hypertension risk in the co-twin design when we full controlled genetic and familiar environmental factors (OR=0.89, 95 CI: 0.46~1.72). After controlling for age and sex, we found the positive BMI-hypertension association was mainly explained by a genetic correlation between them (r_A= 0.59, 95% CI: 0.44~1.00).

Conclusions/Interpretation: Genetics and early-life environments shared by participants within a twin pair appear to account for the association between overweight or obesity and hypertension risk.

Keywords: BMI, hypertension, genetics, early-life environments, twin study

INTRODUCTION

Raised blood pressure remains the leading cause of death globally, high systolic blood pressure accounted for 10·8 million deaths in 2019 (1). Unfortunately, hypertension has high prevalence but low rate of control. It is estimated that 1 in 4 men and 1 in 5 women (1.13 billion people), living with hypertension in 2015, but less than 1/5 have their blood pressure under control (2).

In the worldwide, a large number of people suffer from higher body mass index (BMI), including overweight and obesity. In 2016, a total of 39% adults are overweight, with a BMI ≥ 25 kg/m^2, and 13% are obese (BMI ≥ 30 kg/m^2) (3). Higher BMI is a major risk factor of hypertension. In the original Framingham cohort, Wilson et al. (4) found overweight and obese status were associated with increased risk of hypertension: the multi-adjusted risk ratios (RRs) among the overweight was 1.48 in men and 1.70 in women, while among the obese was 2.23 in men and 2.63 in women. In another Framingham study, weight loss led to a 21%~29% reduction in long-term hypertension risk (5). Mendelian randomization (MR) analysis, using genetic variants as the instrumental variable, has also demonstrated the causality between obesity and hypertension (6).

Genetic and early-life environmental factors, including shared fetal environment, childhood socioeconomic situation and adolescent environment, might have long-term effects on the subsequent risk of obesity (7–9) and hypertension (10–12). However, due to the limitations of general population-based study, their roles in the obesity-hypertension association are uncertain. Co-twin case-control analysis could address part of this difficulty, by controlling for genetic background and key shared familial environmental factors associated with obesity and hypertension. Twins are generally raised together, so they share their early-life environmental factors. They also share the same genetic predisposition and intrauterine environments. Therefore, as naturally matched pairs, co-twin case-control analyses provide an opportunity to explore the role of genetic and early-life environmental factors in the association between overweight or obesity and hypertension (13, 14).

The purpose of this study was to examine the association between overweight or obesity and hypertension, and to explore whether the association could be explained by genetic and common environmental factors shared within a twin pair, based on information from the Chinese National Twin Registry (CNTR).

METHODS

Study Population

This prospective study enrolled twin participants from the CNTR (15), a twin population-based cohort study. Briefly, a total of 61,566 twin pairs, including 31,705 monozygotic (MZ) twins and 15,060 same-sex dizygotic (DZ) twins, from 11 provinces and cities in China were included at the baseline since 2001. Totally, the current study included 32,197 twins whose age was more than 18 years and participated in the resurvey.

We excluded 14 participants whose sex was missing. We also excluded 619 twins whose BMI was missing or with extreme outliers (under 3 or over 3-Z score of the BMI). In addition, we excluded those who were diagnosed with hypertension at baseline (892), and those who had no disease information at baseline or at the time of resurveys (55). Finally, a total of 30,617 twins were included for further association analysis (**Figure 1**).

We used the method of 'Peas in the Pod Questionnaire (PPQ)' to determine zygosity, asking about the degree of similarity shared by twins when they were at school age. Two former studies, based on the data from the CNTR, have verified its accuracy from 85% to 89% (16, 17).

All participants provided informed consent, and the study protocol was approved by the Ethics Committee at Peking University Health Science Center (IRB00001052-11029/14021).

Data Collection

We collected information, including demographics (age, sex, marital status and educational attainment), lifestyles (smoking status, drinking status and physical activity), anthropometric measures (weight and height), twin zygosity and history of diseases (including diabetes and hypertension), from face-to-face questionnaire interview by trained interviewers.

Ascertainment of Overweight or Obesity

Self-reported questionnaire was used to attain the information of height (in centimeter) and weight (in kilograms) at baseline. BMI was calculated by weight in kilograms divided by squared height in meters (kg/m^2). According to the Chinese criteria of obesity

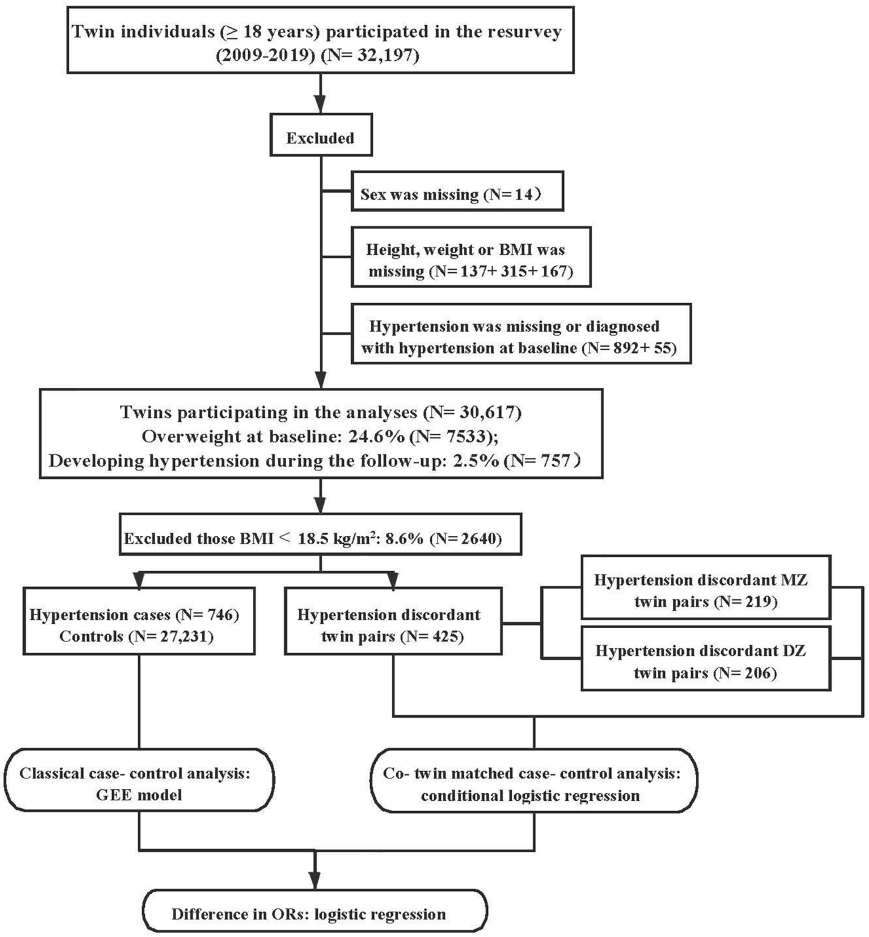


FIGURE 1 | Flow chart of the study population and data analysis. BMI, body mass index; MZ, monozygotic; DZ, dizygotic; GEE, generalized estimating equation; OR, odds ratio.

(18), we categorized BMI into four groups: underweight (<18.5 kg/m^2), normal weight (18.5 to 23.9 kg/m^2), overweight (24.0 to 27.9 kg/m^2) and obesity (≥ 28.0 kg/m^2). In the current study, overweight or obesity was defined as BMI ≥24 kg/m^2, that is, obesity was merged into overweight.

Assessment of Hypertension

We collected the information on hypertension during the follow-up, asking "Have you ever been diagnosed with hypertension by a county/district level or above hospital". Whether the participants are with hypertension or not depends on the doctor's definite diagnosis, not just on their self-reported symptoms.

Assessment of Covariates

Marital status was defined as married (or cohabitating) vs single (or divorced). Education attainment was categorized as primary, secondary and tertiary. Smoking status was grouped into never, former and current smoking. Drinking status was similarly divided into never, past and current drinking. Adequate physical activity was defined as exercising at least 30-minute moderate to high-intensity physical activity a day, and engaging in at least 5 days per week (19). Prevalence of diabetes was dichotomized into diabetes and diabetes-free.

Statistical Analyses

Descriptive Statistics

The characteristics were compared between hypertension and non-hypertension groups. $\chi 2$ tests were used for categorical variables, independent sample t tests for continuous variables with normal distribution, and Mann Whitney U tests for continuous variables with non-normal distribution.

Case-Control Analyses

Generalized estimating equation (GEE) models were applied to assess the overweight-hypertension association, which are conceptually equivalent to logistic regressions for the classic case-control analysis, but controlling for the clustering of twins within a pair. In the co-twin matched case-control design, in which co-twin (both MZ and DZ twins) without hypertension was treated as a control for hypertension twin, we used

conditional logistic regressions to explore the associations. Because cases and controls are matched for genetics and familial environments, discordant twin pairs were more informative than unrelated samples (20). Due to MZ twins share 100% genetic predisposition, while DZ twins share only 50%, we stratified the co-twin matched case-control analysis in MZ and DZ twins, respectively. Finally, logistic regressions were fitted to examine whether the ORs from the GEE model and conditional logistic regression are different, by comparing the distribution of overweight or obesity in the unmatched controls and co-twin controls (21). If a significant association between overweight or obesity and hypertension is only found in the GEE analysis, or OR in the co-twin analysis becomes significantly strengthened or attenuated, genetic and/or early-life environmental factors might play roles in their association (22, 23). In contrast, if difference in ORs between the GEE model and conditional logistic regression is not significant, then the genetic and shared familiar factors might not account for the observed association (14, 21, 24).

Age, sex, marital status, education attainment, smoking status, drinking status, physical activity and diabetes were considered as potential confounders. The basic-adjusted models were controlled for age and sex (twins within a pair have the same age, thus only sex was adjusted in the co-twin analysis). The multi-adjusted models were further adjusted for marital status, education, smoking, drinking, physical activity and diabetes.

Twin Modeling

The classical twin method decomposes the phenotype variation, based on the phenotypic correlations between twin pairs: MZ twins share 100% genetic materials, whereas DZ twins average share 50% of their segregating genes; all twins are correlated for environmental influences to the same extent. In this study, the liability threshold model, an extension of the classical twin modelling was used, assuming individual differences in a trait come from additive genetic (A), nonadditive genetic (D), shared environmental (C) and nonshared environmental (E) influences. The bivariate genetic model estimates the influence of A, C, D and E on each trait (BMI and hypertension), and also explores how much of the association (phenotypic correlation, r_{ph}) between them can be partitioned into addictive genetic (r_A), nonaddictive genetic (r_D), shared environmental (r_C), and unique environmental (r_E) correlation. Because of the effects of C and D are confounded in the classical twin model, including twin pairs reared together, they cannot be calculated simultaneously (25). For both BMI and hypertension, the ADE model was only fitted when the intraclass correlation coefficient (ICC) of MZ was more than double that of DZ twins. To investigate how much of the BMI-hypertension association is attributed to genetic or environmental correlations, we compared the difference of cross-trait, cross-twin correlations (CTCTs) between MZ and DZ twin pairs. A higher CTCT in MZ than in DZ twin pairs indicates that BMI and hypertension are associated because of correlated genetic influences. Based on the full ACE models, several nested models, including AE, CE and E, were fitted by dropping C, A and both components for the selection of best fitting model.

The likelihood ratio test was used to assess the fit of nested models, which approximately follows a $\chi 2$ distribution in that the degree of freedom is equal to the difference of the parameters number between the two models. Each nested model was compared with the full model to choose the best fitting model. The Akaike's information criterion (AIC) was applied for the model selection, in which lower values suggesting a better balance between explanatory power and parsimony (26).

Data cleaning and statistical analyses were performed using Stata/MP 14.0. Structural equation models were fitted in R 3.5.1 with the use of an open source software package named OpenMx (version 2.14.11) (27).

RESULTS

Characteristics of the Study Population

Overall, a total of 30,617 twin individuals, including 17,571 (57.4%) men and 13,570 (44.8%) DZ twins were included in the current study. The average age at baseline was 32.6 ± 11.4 years, the median follow-up time was 4.4 years. 7533 participants (24.6%) were overweight, and 757 (2.5%) participants were diagnosed with hypertension in the resurvey. And among the overweight or obesity individuals at baseline, 371 (4.9%) twins developed hypertension during the follow-up. Participants who had hypertension were more likely to be older, male, overweight, current smokers, current drinkers, to have adequate physical activity, higher education, and to be diagnosed with diabetes, compared with those who were hypertension-free (**Table 1**).

Case-Control Analyses

After adjustment of age, sex, marital status, education, smoking, drinking, physical activity and diabetes, overweight or obesity increased a 94% risk of hypertension in the GEE model (OR=1.94, 95% CI: 1.64~2.30) (**Table 2**). In the multi-adjusted conditional logistic regression, overweight or obesity was associated with an 80% higher hypertension risk (OR=1.80, 95% CI: 1.18~2.74). In the matched analysis of DZ twins, controlling 50% genetic factors, overweight or obesity increased the risk of hypertension under the control of confounders (OR=2.86, 95% CI: 1.57~5.21). However, in the co-twin case-control analysis of MZ twins, the association between overweight or obesity and hypertension was not significant, the multi-adjusted OR was 0.89 (95% CI: 0.46~1.72) (**Table 3**). When we adjusted for the potential confounding variables, the difference in ORs between unmatched and matched case-control analyses was significant in all twin pairs (P=0.016), suggesting genetic, early-life familial environmental factors or both of them may partially contribute to the overweight-hypertension association (**Table 4**).

Twin Model Fitting

For both BMI and hypertension, the ICCs of MZ twins were larger than DZ twins, which implied genetic influences on both traits. Compared with DZ twins (r=0.10, 95% CI: 0.04~0.16), the CTCT was higher for MZ twins (r=0.18, 95% CI: 0.13~0.23), suggesting a genetic correlation between BMI and hypertension

TABLE 1 | Characteristics of the study participants by hypertension diagnosis (N = 30617).

Characteristics	Hypertension-free (N = 29860)	Hypertension (N = 757)	Total (N = 30617)	*P* value
Age, years (mean, SD)	32.2 (11.2)	47.0 (11.3)	32.6 (11.4)	<0.001
Male, n (%)	17008 (57.0)	563 (74.4)	17571 (57.4)	<0.001
DZ, n (%)	13261 (44.9)	309 (41.1)	13570 (44.8)	0.039
Follow-up time, median (IQR)	4.4 (2.0, 5.9)	5.3 (4.4, 6.0)	4.4 (2.1, 5.9)	<0.001
BMI, kg/m^2 (%)				<0.001
Normal weight (BMI, 18.5–23.9)	20069 (67.2)	375 (49.5)	20444 (66.8)	
Underweight (BMI < 18.5)	2629 (8.8)	11 (1.5)	2640 (8.6)	
Overweight (BMI ≥ 24)	7162 (24.0)	371 (49.0)	7533 (24.6)	
Married, n (%)	16584 (76.7)	664 (91.1)	17248 (77.2)	<0.001
Educational attainment, n (%)				<0.001
Primary	2717 (12.6)	180 (24.7)	2897 (13.0)	
Secondary	12541 (58.0)	477 (65.3)	13018 (58.2)	
Tertiary	6366 (29.4)	73 (10.0)	6439 (28.8)	
Smoking status, n (%)				<0.001
Never	15384 (71.2)	444 (60.8)	15828 (70.8)	
Current	5957 (27.6)	268 (36.7)	6225 (27.9)	
Former	279 (1.3)	18 (2.5)	297 (1.3)	
Drinking status, n (%)				<0.001
Never	17092 (79.1)	492 (67.6)	17584 (78.7)	
Current	4362 (20.2)	226 (31.0)	4588 (20.5)	
Former	159 (0.7)	10 (1.4)	169 (0.8)	
Adequate physical activity, n (%)	8568 (42.6)	354 (53.7)	8922 (42.9)	<0.001
Diabetes, n (%)	178 (0.6)	17 (2.2)	195 (0.6)	<0.001

BMI, body mass index; DZ, dizygotic; SD, standard deviation; IQR, interquartile range.

TABLE 2 | ORs (95% CIs) of overweight or obesity-hypertension association (normal BMI as the reference) from the GEE models.

Models	No. of cases	OR (95% CI)
Model[a]	27977	2.03 (1.73,2.39)
Model[b]	20888	1.91 (1.62,2.25)
Model[c]	19394	1.94 (1.64,2.30)

BMI, body mass index; GEE, generalized estimating equation; OR, odds ratio; CI, confidence interval.
[a]Adjusted for age and sex.
[b]Adjusted for age, sex, marital status and education.
[c]Adjusted for age, sex, marital status, education, smoking status, alcohol consumption, physical activity and diabetes.

(**Table 5**). The analysis revealed that the full ACE model was the best-fitting one, which was therefore used for estimation of genetic and environmental influences (**Supplementary Table 3**). The additive genetic factors explained 45% (95% CI: 41%~49%) and 32% (95% CI: 8%~59%) variance of BMI and hypertension, respectively. The genetic correlation, which pointed to what degree genetic variance of BMI predicted the genetic influences on hypertension, was 0.59 (95% CI: 0.44~1.00). The shared and non-shared environmental correlations were not significant (**Table 6**).

Supplementary Analysis

Considering the different effects of overweight and obesity on the risk of incident hypertension, we also separated participants who

TABLE 3 | ORs (95% CIs) for the association between overweight or obesity and hypertension in co-twin control analyses using hypertension discordant twin pairs from the conditional logistic regressions.

Co-twin without hypertension	Twin with hypertension					
	MZ+DZ		DZ		MZ	
	Normal BMI	Overweight	Normal BMI	Overweight	Normal BMI	Overweight
Normal BMI	175	77	72	56	103	21
Overweight	45	128	22	56	23	72
OR (95% CI)[a]	1.63 (1.12,2.37)		2.39 (1.45,3.95)		0.91 (0.51,1.65)	
OR (95% CI)[b]	1.60 (1.09,2.33)		2.44 (1.46,4.09)		0.84 (0.45,1.54)	
OR (95% CI)[c]	1.80 (1.18,2.74)		2.86 (1.57,5.21)		0.89 (0.46,1.72)	

BMI, body mass index; MZ, monozygotic; DZ, dizygotic; OR, odds ratio; CI, confidence interval.
The 425 (206 DZ and 219 MZ) hypertension discordant pairs were divided into four groups with respect to exposure (overweight) status. In 175 (72 DZ and 103 MZ) twin pairs, both had normal BMI. In 128 (56 DZ and 72 MZ) twin pairs, both were overweight. In 77 (56 MZ and 21 MZ) twin pairs, the healthy (hypertension-free) co-twin had normal weight and the diseased twin was overweight. In 45 (22 DZ and 23 MZ) twin pairs, the diseased co-twin had normal BMI and the healthy twin was overweight.
[a]Adjusted for sex.
[b]Adjusted for sex, marital status, education.
[c]Adjusted for sex, marital status, education, smoking status, alcohol consumption, physical activity and diabetes.

TABLE 4 | Differences in ORs (95% CIs) for the unmatched GEE models and matched co-twin control analyses (the difference in overweight or obesity between unmatched and co-twin matched controls).

Models	MZ+DZ			DZ			MZ		
	No. of cases	OR (95% CI)	*P* value	No. of cases	OR (95% CI)	*P* value	No. of cases	OR (95% CI)	*P* value
Model[a]	27656	1.26 (1.03,1.55)	0.023	27437	1.22 (0.91,1.63)	0.190	27450	1.30 (0.99,1.72)	0.062
Model[b]	20574	1.32 (1.07,1.62)	0.009	20369	1.26 (0.94,1.69)	0.122	20375	1.36 (1.03,1.81)	0.032
Model[c]	19115	1.30 (1.05,1.62)	0.016	18924	1.26 (0.92,1.72)	0.146	18936	1.34 (1.00,1.79)	0.052

MZ, monozygotic; DZ, dizygotic; GEE, generalized estimating equation; OR, odds ratio; CI, confidence interval.
[a]*Adjusted for age and sex.*
[b]*Adjusted for age, sex, marital status and education.*
[c]*Adjusted for age, sex, marital status, education, smoking status, alcohol consumption, physical activity and diabetes.*

TABLE 5 | Correlations (95% CIs) for BMI, hypertension, and BMI-hypertension by zygosity.

Zygosity	Within-trait, cross-twin		Cross-trait, within-twin	Cross-trait, cross-twin
	BMI	Hypertension		
MZ	0.70 (0.68,0.72)	0.73 (0.67,0.79)	0.19 (0.09,0.24)	0.18 (0.13,0.23)
DZ	0.51 (0.48,0.53)	0.57 (0.45,0.67)	0.22 (0.16,0.28)	0.10 (0.04,0.16)

BMI, body mass index; MZ, monozygotic; DZ, dizygotic; CI, confidence interval.

TABLE 6 | Parameter estimates (95% CIs) from the best-fitting bivariate ACE full model of BMI and hypertension.

	Variance components		Correlation	
	BMI	Hypertension		
			r_{Ph}	0.21 (0.17,0.25)
A	0.45 (0.41,0.49)	0.32 (0.08,0.59)	r_A	0.59 (0.44,1.00)
C	0.36 (0.32,0.40)	0.41 (0.15,0.63)	r_C	-0.06 (-1.00,1.00)
E	0.19 (0.19,0.20)	0.27 (0.21,0.33)	r_E	0.05 (-0.05,0.14)

BMI, body mass index; CI, confidence interval; A, Additive genetic factors; C, shared environmental factors; E, non-shared environmental factors; r_{Ph}, phenotypic correlation; r_A, genetic correlation; r_C, shared environmental correlation; r_E, non-shared environmental correlation.

were overweight (BMI: 24- 28 kg/m2) and obesity (BMI ≥ 28 kg/m2) to explore their associations with hypertension. Under the control of potential influenced factors, both overweight and obesity increased hypertension risk, the ORs were 1.87 (95% CI: 1.57~2.24) and 2.64 (95% CI: 1.92~3.61), respectively (**Supplementary Table 1**). In the matched case-control study, overweight was associated with a 72% increased hypertension risk (OR=1.72, 95% CI: 1.12~2.67). However, due to the limited sample size, we found a borderline significant obesity-hypertension association in the matched analyses (OR=9.26, 95% CI: 1.00~85.50) (**Supplementary Table 2**). The different in ORs between unmatched and matched designs was significant for the obesity (*P*=0.009), but not significant for the overweight (*P*=0.075) (**Supplementary Table 3**).

DISCUSSION

In this large-scale, nationwide Chinese twin study, we found that both overweight and obesity were significantly associated with increased hypertension risk. And the associations were different between unmatched and matched analyses, and even non-significant in the co-twin analysis of MZ twins, indicating genetic or both genetic and environmental factors shared between co-twins are likely to contribute to the association. Furthermore, we found a positive correlation between BMI and hypertension, which was explained by a genetic correlation, providing evidence for the contribution of overlap genetic factors on their association.

Consistent with our findings, a growing number of studies have reported that overweight and obesity were independently associated with increased risk of hypertension. Using 5209 participants aged 30 to 62 years from the original Framingham cohort, Wilson et al. (4) found overweight and obese status were positively associated with hypertension: compared with those with normal weight, overweight increased a 48% (95% CI: 1.24~1.75) and 70% (95% CI: 1.48~1.94) risk of hypertension in the men and women, respectively; the age-adjusted RRs were 2.23 (95% CI: 1.75~2.84), and 2.63 (95% CI: 2.20~3.15) in male and female obese individuals, respectively. After 6.38-year follow-up, Qi et al. (28) reported a positive association between BMI and hypertension in the Chines: the RR was 3.13 (95% CI: 2.84~3.45) for the obesity. In the Framingham Study, Moore et al. (5) found weight loss ≥ 6.8 kg led to a 28% (RR=0.72, 95% CI: 0.49~1.05) and a 37% (RR=0.63, 95% CI: 0.42~0.95) reduction in hypertension risk for middle-aged and older adults, respectively; And sustained weight loss also reduced the hypertension risk: 22%

and 26% for middle-aged and older adults, respectively. These studies provided evidence that overweight and obesity were independently associated with hypertension risk.

The mechanisms underlying the overweight (obesity)-hypertension association are complex and not completely understood. Obesity can directly produce a variety of structural and functional changes of the cardiovascular system, including lower cardiac output, poorer left ventricular systolic function, higher peripheral resistance, increased left ventricular mass, left ventricular wall thickness and internal dimension (29). In addition, obesity is associated with mechanisms that could increase sympathetic nervous system (SNS) activity, which is believed to play an important role in the development of hypertension. Angiotensin II could increase SNS activity, while angiotensinogen is expressed in visceral adipocytes (30, 31). In the obesity, the inhibitory of arterial baroreflex on SNS activity is reduced, contributing to the increased SNS activity to muscle and kidney (32, 33). And the dysregulation of the hypothalamic-pituitary-adrenal axis, characterized by obesity, seems to be important to the activation of the SNS in obese humans (34).

The co-twin case-control analysis could explore associations, under the control of genetic and unmeasured early-life environmental factors. In the current study, we found the overweight-hypertension association was attenuated in the matched study, and even disappeared in the co-twin analysis of MZ twins, indicating that the observed association was fully explained by genetic and familiar factors shared within a twin pair. Our results indicate these with family history of obesity may have high hypertension risk, which showed the important to prevent hypertension in these population. However, not consistent with our findings, MR analysis has demonstrated the causal effect of obesity on hypertension. Including 119,859 participants from UK Biobank, Lee et al. (6) showed a positive association between genetically instrumented higher BMI and hypertension risk (OR= 1.64, 95% CI: 1.48~1.83). In a Korea cohort study, using genetic risk scores (GRS), created by 6 single-nucleotide polymorphisms associated with BMI, researchers found a causal effect of BMI on hypertension (OR: 1.13~1.26) (35). In addition, a large number of studies have investigated the biological mechanisms underlying obesity-hypertension association. Therefore, our findings based on statistics analysis should be interpreted with caution. More large studies from Chinese population are warranted to verify our results.

Furthermore, using bivariate twin model, we further found the positive BMI-hypertension association was explained by a genetic correlation between them, providing evidence for the contribution of overlap genes on their relationship. Including a total of 913 subjects from 179 families, Li et al. (36) found waist circumference (WC) was genetically correlated with systolic blood pressure (r_A=0.27), but found no significant genetic correlations between BMI and blood pressure. Although we found no study exploring the genetic link between overweight (obesity) and hypertension, numerous studies provide evidences for the pleiotropy between obesity and hypertension. *FTO* (fat mass and obesity-associated) gene, the first identified gene for obesity, is the strongest BMI related genetic factors (37, 38). Meta-analysis has demonstrated the associations between *FTO* SNPs and obesity risk (39, 40). It is reported that *FTO* gene is highly expressed in the hypothalamic nuclei (41, 42), involving in the control of energy homeostasis (43) and regulation of blood pressure (44). To date, several studies have investigated the association between *FTO* variants and risk of hypertension. Although the results are inconsistent, many large studies showed some of *FTO* genotypes significantly increased risks of hypertension (45). Many other genes, such as *MC4R* (46–48), TNF-α (49–51), *LEP* (*LEPR*) (52–56) and *β2AR* (57, 58), were all associated with both obesity and hypertension. However, because of the lack of comparable co-twin studies, the roles of shared genetic factors in the relationship between overweight or obesity and hypertension are still needed to be explored.

Although our co-twin analysis reported that early-life environment might contribute to the overweight-hypertension association, the common environmental correlation was not significant in the twin model. Not consistent with our results, as important parts of early-life environments, a growing body of studies described that poor fetal and early postnatal growth were associated with subsequent risk of obesity and hypertension. The majority of epidemiological studies utilize birth weight and gestational age as proxy markers for suboptimal utero growth. Abundant evidences suggested that high birth weight (HBW) and large for gestational age were associated with an increased risk of obesity later in life (59–61). In a meta-analysis, including 14 cohort studies, HBW (≥4000g) was associated with higher risk of obesity (OR=1.43, 95% CI: 1.25~1.64), but not low birth weight (LBW) (<2500 g) (60). Furthermore, low birth weight (LBW) was associated with higher hypertension risk (62–65). In the Shanghai Women's Health Study and the Shanghai Men's Health Study, an excess risk of hypertension was observed for LBW, hazard ratio (HR) was 1.20 (95% CI: 1.11~1.30) (63). In the Swedish twin study on the fetal origins of hypertension, Bergvall et al. (65) provided evidence that LBW was associated with increased hypertension risk. Although early-life environmental factors are associated with both obesity and hypertension, whether those factors confound the overweight (obesity)-hypertension association is unknown.

Some limitations of this study need to be mentioned. First, we calculated BMI from self-reported height and weight, which could have led to an underestimation of the overweight. Besides, we collected disease information from self-reported questionnaires without measuring the blood pressure. Because of the higher proportion of patients who were not aware of their hypertension (12.3%~24.7%) in the Chinese adults (66, 67), individuals with undiagnosed hypertension might have been misclassified as hypertension-free. Both of those could have led to biased estimation for the overweight-hypertension association. Finally, despite the large sample sizes, the number of cases for stratification analyses was small, especially for the co-twin control analysis. Specifically, we obtained a borderline significant association between obesity and hypertension, due to the limited sample sizes. Thus, further investigations are needed to assess whether the genetics and early-life environmental factors account for this association.

Nonetheless, the current study has several strengths. First, the large nationwide twin cohort allowed us to explore the effects of overweight and obesity on hypertension, and simultaneously estimate the potential influence of genetic and early-life environmental factors in their relationship with sufficient power. We used GEE models, controlling for the clustering of twins within a pair, to provide evidence for the overweight-hypertension association. And further assessing the roles of genetic and familiar factors on the given association, *via* co-twin case-control design. Second, we only included twins who were diagnosed with hypertension during the follow-up, making the temporality clear and thus minimize the possibility of reverse causality. Third, due to DZ twins only share 50% of their genetic predisposition, co-twin matched case-control analyses including both MZ and DZ twins do not completely control for genetic factors. Therefore, we further repeated co-twin design in MZ and DZ twins separately, which verified our results.

In conclusion, with the current study, we add evidence to the positive link between overweight (obesity) and hypertension, and show the importance of genetic and family environmental factors for their association. That is, due to the common genetic predisposition, individuals with higher BMI seem to be more likely to develop into hypertension. A next step would be to verify our findings in more prospective studies, and find more genes and environments responsible for the overweight (obesity)-hypertension association.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/**Supplementary Material**. Further inquiries can be directed to the corresponding authors.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Ethics Committee at Peking University Health Science Center (IRB00001052-11029/14021). The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

WC and WG contributed to the study design and supervise the whole project. ZP, MY, HW, XPW, ZD, FW, GJ, XJW, YL, JD, and LL contributed to conduct field study and collect the data. KZ, JL, CY, SW, TH, DS, CL, and YP contributed to the results interpretation and provided critical comments. LML helped to design and supervise the whole study and obtain funding. YX analyzed data and drafted the manuscript. All authors contributed to the article and approved the submitted version.

FUNDING

This study was funded by National Natural Science Foundation of China (82073633, 81973126, 81711530051, 81573223, 81473041), and Special Fund for Health Scientific Research in the Public Welfare (201502006, 201002007). The funders had no role in study design and conduct, data collection, analysis and interpretation, preparation of the manuscript, or the decision to publication.

ACKNOWLEDGMENTS

We thank all the participants and project staff who took part in the Chinese National Twin Registry for their contributions.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2021.743962/full#supplementary-material

REFERENCES

1. GBD 2019 Risk Factors Collaborators. Global Burden of 87 Risk Factors in 204 Countries and Territories, 1990-2019: A Systematic Analysis for the Global Burden of Disease Study 2019. *Lancet* (2020) 396(10258):1223–49. doi: 10.1016/S0140-6736(20)30752-2
2. WHO. *Hypertension* (2019). Available at: https://www.who.int/news-room/fact-sheets/detail/hypertension.
3. WHO. *Obesity and Overweight*. Available at: https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight.
4. Wilson PW, D'Agostino RB, Sullivan L, Parise H, Kannel WB. Overweight and Obesity as Determinants of Cardiovascular Risk: The Framingham Experience. *Arch Intern Med* (2002) 162(16):1867–72. doi: 10.1001/archinte.162.16.1867
5. Moore LL, Visioni AJ, Qureshi MM, Bradlee ML, Ellison RC, D'Agostino R, et al. Weight Loss in Overweight Adults and the Long-Term Risk of Hypertension: The Framingham Study. *Arch Intern Med* (2005) 165 (11):1298–303. doi: 10.1001/archinte.165.11.1298
6. Lyall DM, Celis-Morales C, Ward J, Iliodromiti S, Anderson JJ, Gill JMR, et al. Association of Body Mass Index With Cardiometabolic Disease in the UK Biobank: A Mendelian Randomization Study. *JAMA Cardiol* (2017) 2 (8):882–9. doi: 10.1001/jamacardio.2016.5804
7. Goodarzi MO. Genetics of Obesity: What Genetic Association Studies Have Taught Us About the Biology of Obesity and its Complications. *Lancet Diabetes Endocrinol* (2018) 6(3):223–36. doi: 10.1016/S2213-8587(17)30200-0
8. Nicolaidis S. Environment and Obesity. *Metabolism: Clinical and Experimental* (2019) 100S:153942. doi: 10.1016/j.metabol.2019.07.006
9. Pigeyre M, Yazdi FT, Kaur Y, Meyre D. Recent Progress in Genetics, Epigenetics and Metagenomics Unveils the Pathophysiology of Human Obesity. *Clin Sci (London Engl 1979)* (2016) 130(12):943–86. doi: 10.1042/CS20160136
10. Poulter NR, Prabhakaran D, Caulfield M. Hypertension. *Lancet* (2015) 386 (9995):801–12. doi: 10.1016/S0140-6736(14)61468-9
11. Carey RM, Muntner P, Bosworth HB, Whelton PK. Prevention and Control of Hypertension: JACC Health Promotion Series. *J Am Coll Cardiol* (2018) 72 (11):1278–93. doi: 10.1016/j.jacc.2018.07.008

12. Cowley AW. The Genetic Dissection of Essential Hypertension. *Nat Rev Genet* (2006) 7(11):829–40. doi: 10.1038/nrg1967
13. Lundqvist E, Kaprio J, Verkasalo PK, Pukkala E, Koskenvuo M, Söderberg KC, et al. Co-Twin Control and Cohort Analyses of Body Mass Index and Height in Relation to Breast, Prostate, Ovarian, Corpus Uteri, Colon and Rectal Cancer Among Swedish and Finnish Twins. *Int J Cancer* (2007) 121(4):810–8. doi: 10.1002/ijc.22746
14. Xu W, Qiu C, Gatz M, Pedersen NL, Johansson B, Fratiglioni L. Mid- and Late-Life Diabetes in Relation to the Risk of Dementia: A Population-Based Twin Study. *Diabetes* (2009) 58(1):71–7. doi: 10.2337/db08-0586
15. Gao W, Cao W, Lv J, Yu C, Wu T, Wang S, et al. The Chinese National Twin Registry: A 'Gold Mine' for Scientific Research. *J Intern Med* (2019) 286 (3):299–308. doi: 10.1111/joim.12926
16. Gao W, Li L, Cao W, Zhan S, Lv J, Qin Y, et al. Determination of Zygosity by Questionnaire and Physical Features Comparison in Chinese Adult Twins. *Twin Res Hum Genet* (2006) 9(2):266–71. doi: 10.1375/183242706776382446
17. Wang B, Gao W, Yu C, Cao W, Lv J, Wang S, et al. Determination of Zygosity in Adult Chinese Twins Using the 450K Methylation Array Versus Questionnaire Data. *PloS One* (2015) 10(4):e0123992. doi: 10.1371/journal.pone.0123992
18. Zhou B-F. Predictive Values of Body Mass Index and Waist Circumference for Risk Factors of Certain Related Diseases in Chinese Adults–Study on Optimal Cut-Off Points of Body Mass Index and Waist Circumference in Chinese Adults. *BioMed Environ Sci* (2002) 15(1):83–96
19. Wang S-S, Lay S, Yu H-N, Shen S-R. Dietary Guidelines for Chinese Residents (2016): Comments and Comparisons. *J Zhejiang Univ Sci B* (2016) 17(9):649–56. doi: 10.1631/jzus.B1600341
20. Gatz M, Svedberg P, Pedersen NL, Mortimer JA, Berg S, Johansson B. Education and the Risk of Alzheimer's Disease: Findings From the Study of Dementia in Swedish Twins. *journals gerontology Ser B psychol Sci Soc Sci* (2001) 56(5):P292–300. doi: 10.1093/geronb/56.5.p292
21. Xu WL, Atti AR, Gatz M, Pedersen NL, Johansson B, Fratiglioni L. Midlife Overweight and Obesity Increase Late-Life Dementia Risk: A Population-Based Twin Study. *Neurology* (2011) 76(18):1568–74. doi: 10.1212/WNL.0b013e3182190d09
22. Kato K, Sullivan PF, Evengård B, Pedersen NL. Premorbid Predictors of Chronic Fatigue. *Arch Gen Psychiatry* (2006) 63(11):1267–72. doi: 10.1002/ijc.31365
23. Bao C, Pedersen NL, Yang R, Marseglia A, Xu W, Wang Y, et al. Diabetes in Midlife and Risk of Cancer in Late Life: A Nationwide Swedish Twin Study. *Int J Cancer* (2018) 143(4):793–800. doi: 10.1002/ijc.31365
24. Bao C, Yang R, Pedersen NL, Xu W, Xu H, Song R, et al. Overweight in Midlife and Risk of Cancer in Late Life: A Nationwide Swedish Twin Study. *Int J Cancer* (2019) 144(9):2128–34. doi: 10.1002/ijc.32005
25. Rijsdijk FV, Sham PC. Analytic Approaches to Twin Data Using Structural Equation Models. *Brief Bioinform* (2002) 3(2):119–33. doi: 10.1093/bib/3.2.119
26. Akaike H. Factor Analysis and AIC. *Psychometrika* (1987) 52(3):317–32. doi: 10.1007/BF02294359
27. Boker S, Neale M, Maes H, Wilde M, Spiegel M, Brick T, et al. OpenMx: An Open Source Extended Structural Equation Modeling Framework. *Psychometrika* (2011) 76(2):306–17. doi: 10.1007/s11336-010-9200-6
28. Qi S-F, Zhang B, Wang H-J, Yan J, Du P, Zhang W, et al. Joint Effects of Age and Body Mass Index on the Incidence of Hypertension Subtypes in the China Health and Nutrition Survey: A Cohort Study Over 22years. *Prev Med* (2016) 89:23–30. doi: 10.1016/j.ypmed.2016.05.004
29. Bastien M, Poirier P, Lemieux I, Després J-P. Overview of Epidemiology and Contribution of Obesity to Cardiovascular Disease. *Prog Cardiovasc Dis* (2014) 56(4):369–81. doi: 10.1016/j.pcad.2013.10.016
30. Reid IA. Interactions Between ANG II, Sympathetic Nervous System, and Baroreceptor Reflexes in Regulation of Blood Pressure. *Am J Physiol* (1992) 262(6 Pt 1):E763–78. doi: 10.1152/ajpendo.1992.262.6.E763
31. Engeli S, Negrel R, Sharma AM. Physiology and Pathophysiology of the Adipose Tissue Renin-Angiotensin System. *Hypertension* (2000) 35(6):1270–7. doi: 10.1161/01.hyp.35.6.1270
32. Alvarez GE, Beske SD, Ballard TP, Davy KP. Sympathetic Neural Activation in Visceral Obesity. *Circulation* (2002) 106(20):2533–6. doi: 10.1161/01.cir.0000041244.79165.25
33. Grassi G, Seravalle G, Dell'Oro R, Turri C, Bolla GB, Mancia G. Adrenergic and Reflex Abnormalities in Obesity-Related Hypertension. *Hypertension* (2000) 36(4):538–42. doi: 10.1161/01.hyp.36.4.538
34. Dodt C, Wallin G, Fehm HL, Elam M. The Stress Hormone Adrenocorticotropin Enhances Sympathetic Outflow to the Muscle Vascular Bed in Humans. *J Hypertens* (1998) 16(2):195–201. doi: 10.1097/00004872-199816020-00010
35. Lee M-R, Lim Y-H, Hong Y-C. Causal Association of Body Mass Index With Hypertension Using a Mendelian Randomization Design. *Med (Baltimore)* (2018) 97(30):e11252. doi: 10.1097/MD.0000000000011252
36. Li JKY, Ng MCY, So WY, Chiu CKP, Ozaki R, Tong PCY, et al. Phenotypic and Genetic Clustering of Diabetes and Metabolic Syndrome in Chinese Families With Type 2 Diabetes Mellitus. *Diabetes Metab Res Rev* (2006) 22 (1):46–52. doi: 10.1002/dmrr.577
37. Frayling TM, Timpson NJ, Weedon MN, Zeggini E, Freathy RM, Lindgren CM, et al. A Common Variant in the FTO Gene Is Associated With Body Mass Index and Predisposes to Childhood and Adult Obesity. *Sci (New York NY)* (2007) 316(5826):889–94. doi: 10.1126/science.1141634
38. Xi B, Mi J. Genome-Wide Association Studies of Common Obesity: Now and Future. *BioMed Environ Sci* (2013) 26(10):787–91. doi: 10.3967/bes2013.001
39. Peng S, Zhu Y, Xu F, Ren X, Li X, Lai M. FTO Gene Polymorphisms and Obesity Risk: A Meta-Analysis. *BMC Med* (2011) 9:71. doi: 10.1186/1741-7015-9-71
40. Zhao N-N, Dong G-P, Wu W, Wang J-L, Ullah R, Fu J-F. FTO Gene Polymorphisms and Obesity Risk in Chinese Population: A Meta-Analysis. *World J Pediatr WJP* (2019) 15(4):382–9. doi: 10.1007/s12519-019-00254-2
41. Gerken T, Girard CA, Tung Y-CL, Webby CJ, Saudek V, Hewitson KS, et al. The Obesity-Associated FTO Gene Encodes a 2-Oxoglutarate-Dependent Nucleic Acid Demethylase. *Sci (New York NY)* (2007) 318(5855):1469–72. doi: 10.1126/science.1151710
42. Schmid PM, Heid I, Buechler C, Steege A, Resch M, Birner C, et al. Expression of Fourteen Novel Obesity-Related Genes in Zucker Diabetic Fatty Rats. *Cardiovasc Diabetol* (2012) 11:48. doi: 10.1186/1475-2840-11-48
43. Wardle J, Carnell S, Haworth CMA, Farooqi IS, O'Rahilly S, Plomin R. Obesity Associated Genetic Variation in FTO Is Associated With Diminished Satiety. *J Clin Endocrinol Metab* (2008) 93(9):3640–3. doi: 10.1210/jc.2008-0472
44. Guyenet PG. The Sympathetic Control of Blood Pressure. *Nat Rev Neurosci* (2006) 7(5):335–46. doi: 10.1038/nrn1902
45. He D, Fu M, Miao S, Hotta K, Chandak GR, Xi B. FTO Gene Variant and Risk of Hypertension: A Meta-Analysis of 57,464 Hypertensive Cases and 41,256 Controls. *Metabolism: Clin Exp* (2014) 63(5):633–9. doi: 10.1016/j.metabol.2014.02.008
46. Marcadenti A, Fuchs FD, Matte U, Sperb F, Moreira LB, Fuchs SC. Effects of FTO RS9939906 and MC4R RS17782313 on Obesity, Type 2 Diabetes Mellitus and Blood Pressure in Patients With Hypertension. *Cardiovasc Diabetol* (2013) 12:103. doi: 10.1186/1475-2840-12-103
47. da Silva AA, do Carmo JM, Wang Z, Hall JE. Melanocortin-4 Receptors and Sympathetic Nervous System Activation in Hypertension. *Curr Hypertens Rep* (2019) 21(6):46. doi: 10.1007/s11906-019-0951-x
48. Lotta LA, Mokrosiński J, Mendes de Oliveira E, Li C, Sharp SJ, Ja L, et al. Human Gain-of-Function MC4R Variants Show Signaling Bias and Protect Against Obesity. *Cell* (2019) 177(3). doi: 10.1016/j.cell.2019.03.044
49. Pausova Z, Deslauriers B, Gaudet D, Tremblay J, Kotchen TA, Larochelle P, et al. Role of Tumor Necrosis Factor-Alpha Gene Locus in Obesity and Obesity-Associated Hypertension in French Canadians. *Hypertension* (2000) 36(1):14–9. doi: 10.1161/01.hyp.36.1.14
50. Li Y-y. Tumor Necrosis Factor-Alpha G308α Gene Polymorphism and Essential Hypertension: A Meta-Analysis Involving 2244 Participants. *PloS One* (2012) 7(4):e35408. doi: 10.1371/journal.pone.0035408
51. Yao Y-S, Chang W-W, Jin Y-L. Association Between TNF-A Promoter -308G/A Polymorphism and Essential Hypertension in the Asian Population: A Meta-Analysis. *J Renin-Angiotensin-Aldosterone System JRAAS* (2017) 18 (4):1470320317741066. doi: 10.1177/1470320317741066
52. Li Y-X, Zhang Q, Shang X-M, Li Y-Q, Liu X-K, Liu C-Q, et al. Association of Two Well-Defined Polymorphisms in Leptin and Leptin Receptor Genes With Hypertension and Circulating Leptin: A Meta-Analysis. *Arch Med Res* (2015) 46(1):38–46. doi: 10.1016/j.arcmed.2014.11.012

53. Kilpeläinen TO, Carli JFM, Skowronski AA, Sun Q, Kriebel J, Feitosa MF, et al. Genome-Wide Meta-Analysis Uncovers Novel Loci Influencing Circulating Leptin Levels. *Nat Commun* (2016) 7:10494. doi: 10.1038/ncomms10494
54. de Luis DA, Perez Castrillón JL, Dueñas A. Leptin and Obesity. *Minerva Med* (2009) 100(3):229–36.
55. Pan WW, Myers MG. Leptin and the Maintenance of Elevated Body Weight. *Nat Rev Neurosci* (2018) 19(2):95–105. doi: 10.1038/nrn.2017.168
56. Gu P, Jiang W, Chen M, Lu B, Shao J, Du H, et al. Association of Leptin Receptor Gene Polymorphisms and Essential Hypertension in a Chinese Population. *J Endocrinological Invest* (2012) 35(9):859–65. doi: 10.3275/8238
57. Daghestani MH, Warsy A, Daghestani MH, Al-Odaib AN, Eldali A, Al-Eisa NA, et al. Arginine 16 Glycine Polymorphism in β2-Adrenergic Receptor Gene is Associated With Obesity, Hyperlipidemia, Hyperleptinemia, and Insulin Resistance in Saudis. *Int J Endocrinol* (2012) 2012:945608. doi: 10.1155/2012/945608
58. Gjesing AP, Andersen G, Burgdorf KS, Borch-Johnsen K, Jørgensen T, Hansen T, et al. Studies of the Associations Between Functional Beta2-Adrenergic Receptor Variants and Obesity, Hypertension and Type 2 Diabetes in 7,808 White Subjects. *Diabetologia* (2007) 50(3):563–8. doi: 10.1007/s00125-006-0578-8
59. Johnsson IW, Haglund B, Ahlsson F, Gustafsson J. A High Birth Weight is Associated With Increased Risk of Type 2 Diabetes and Obesity. *Pediatr Obes* (2015) 10(2):77–83. doi: 10.1111/ijpo.230
60. Zhao Y, Wang S-F, Mu M, Sheng J. Birth Weight and Overweight/Obesity in Adults: A Meta-Analysis. *Eur J Pediatr* (2012) 171(12):1737–46. doi: 10.1007/s00431-012-1701-0
61. Derraik JGB, Maessen SE, Gibbins JD, Cutfield WS, Lundgren M, Ahlsson F. Large-for-Gestational-Age Phenotypes and Obesity Risk in Adulthood: A Study of 195,936 Women. *Sci Rep* (2020) 10(1):2157. doi: 10.1038/s41598-020-58827-5
62. Knop MR, Geng T-T, Gorny AW, Ding R, Li C, Ley SH, et al. Birth Weight and Risk of Type 2 Diabetes Mellitus, Cardiovascular Disease, and Hypertension in Adults: A Meta-Analysis of 7 646 267 Participants From 135 Studies. *J Am Heart Assoc* (2018) 7(23):e008870. doi: 10.1161/JAHA.118.008870
63. Xia Q, Cai H, Xiang Y-B, Zhou P, Li H, Yang G, et al. Prospective Cohort Studies of Birth Weight and Risk of Obesity, Diabetes, and Hypertension in Adulthood Among the Chinese Population. *J Diabetes* (2019) 11(1):55–64. doi: 10.1111/1753-0407.12800
64. Tian J-Y, Cheng Q, Song X-M, Li G, Jiang G-X, Gu Y-Y, et al. Birth Weight and Risk of Type 2 Diabetes, Abdominal Obesity and Hypertension Among Chinese Adults. *Eur J Endocrinol* (2006) 155(4):601–7. doi: 10.1530/eje.1.02265
65. Bergvall N, Iliadou A, Johansson S, de Faire U, Kramer MS, Pawitan Y, et al. Genetic and Shared Environmental Factors do Not Confound the Association Between Birth Weight and Hypertension: A Study Among Swedish Twins. *Circulation* (2007) 115(23):2931–8. doi: 10.1161/CIRCULATIONAHA.106.674812
66. Wang Z, Chen Z, Zhang L, Wang X, Hao G, Zhang Z, et al. Status of Hypertension in China: Results From the China Hypertension Survey, 2012-2015. *Circulation* (2018) 137(22):2344–56. doi: 10.1161/CIRCULATIONAHA.117.032380
67. Lu J, Lu Y, Wang X, Li X, Linderman GC, Wu C, et al. Prevalence, Awareness, Treatment, and Control of Hypertension in China: Data From 1·7 Million Adults in a Population-Based Screening Study (China PEACE Million Persons Project). *Lancet* (2017) 390(10112):2549–58. doi: 10.1016/S0140-6736(17)32478-9

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

The reviewer XT declared a shared affiliation, with no collaboration, with ZP to the handling editor at the time of review.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 05 October 2021
doi: 10.3389/fendo.2021.736077

Association of Anthropometric Indices With the Development of Diabetes Among Hypertensive Patients in China: A Cohort Study

Yingshan Liu[1,2†], Xiaocong Liu[3†], Shuting Zhang[2†], Qibo Zhu[2], Xiaoying Fu[2], Hongmei Chen[1,2], Haixia Guan[1,2], Yinghua Xia[4], Qun He[4] and Jian Kuang[1,2]*

[1] The Second School of Clinical Medicine, Southern Medical University, Guangzhou, China, [2] Department of Endocrinology, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou, China, [3] Department of Cardiology, Guangdong Cardiovascular Institute, Guangdong Provincial People's Hospital, Guangdong Academy of Medical Sciences, Guangzhou, China, [4] Guangdong Provincial Institute of Public Health, Guangdong Provincial Center for Disease Control and Prevention, Guangzhou, China

OPEN ACCESS

Edited by:
Ozra Tabatabaei-Malazy, Tehran University of Medical Sciences, Iran

Reviewed by:
Tim Welborn, Sir Charles Gairdner Hospital, Australia
Francesca Battista, University of Padua, Italy

****Correspondence:***
Jian Kuang
kuangjian@gdph.org.cn

[†]These authors have contributed equally to this work

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *04 July 2021*
Accepted: *25 August 2021*
Published: *05 October 2021*

Citation:
Liu Y, Liu X, Zhang S, Zhu Q, Fu X, Chen H, Guan H, Xia Y, He Q and Kuang J (2021) Association of Anthropometric Indices With the Development of Diabetes Among Hypertensive Patients in China: A Cohort Study. Front. Endocrinol. 12:736077. doi: 10.3389/fendo.2021.736077

Background: Patients with comorbidity of hypertension and diabetes are associated with higher morbidity and mortality of cardiovascular disease than those with hypertension or diabetes alone. The present study aimed to identify anthropometric risk factors for diabetes among hypertensive patients who were included in a retrospective cohort study.

Methods: Hypertensive adults without diabetes were recruited in China. Demographic, clinical, biochemical, and anthropometric indices were collected at baseline and during the follow-up. Anthropometric measures included BMI, waist circumference, waist-to-height ratio (WHtR), and waist-to-hip ratio, and several novel indices. To estimate the effect of baseline and dynamic changes of each anthropometric index on risk of new-onset diabetes (defined as self-reported physician-diagnosed diabetes and/or use of hypoglycemic medication, or new-onset FPG≥7.0 mmol/L during follow-up), Cox regression models were used.

Results: A total of 3852 hypertensive patients were studied, of whom 1167 developed diabetes during follow-up. Multivariate Cox regression analyses showed that there was a graded increased risk of incident diabetes with successively increasing anthropometric indices mentioned above (all $P<0.05$). Regardless of the baseline general obesity status, elevated WHtR was both related to higher risk of diabetes; the HRs (95%CI) of baseline BMI<24 kg/m^2 & WHtR≥0.5 group and BMI≥24 kg/m^2 & WHtR≥0.5 group were 1.34 (1.05, 1.72), 1.85 (1.48, 2.31), respectively. Moreover, the dynamic changes of WHtR could sensitively reflect diabetes risk. Diabetes risk significantly increased when patients with baseline WHtR<0.5 progressed to WHtR≥0.5 during the follow-up (HR=1.63; 95% CI, 1.11, 2.40). There was also a decreasing trend towards the risk of incident diabetes when baseline abnormal WHtR reversed to normal at follow-up (HR=1.93; 95%CI, 1.36, 2.72) compared with those whose WHtR remained abnormal at follow-up (HR=2.04; 95% CI, 1.54, 2.71).

Conclusions: Central obesity is an independent and modifiable risk factor for the development of diabetes among hypertensive patients. Measuring indices of central obesity in addition to BMI in clinics could provide incremental benefits in the discrimination of diabetes among Chinese hypertensive patients. Dynamic changes of WHtR could sensitively reflect changes in the risk of diabetes. Therefore, long-term monitoring of hypertensive patients using non-invasive anthropometric measures and timely lifestyle intervention could effectively reduce the development of diabetes.

Keywords: diabetes, hypertension, anthropometric indices, central obesity, waist to height ratio (WHtR), cohort study

INTRODUCTION

Both hypertension and diabetes represent recognized overall global public health burden (1). Globally, the prevalence of hypertension and diabetes presents a persistently increasing trend; Approximately 1.13 billion and 0.42 billion have hypertension and diabetes, respectively. These two diseases frequently coexist and are closely related, both existing as major risk factors for cardiovascular and cerebrovascular diseases (2). Evidence has revealed that hypertensive patients with diabetes had a two-fold increased risk for developing cardiovascular diseases (CVD) compared with those without diabetes (3). As a result, early recognition of hypertensive patients with a high risk of diabetes is crucial to preventing further progress to cardiovascular and cerebrovascular diseases and improving prognosis.

Obesity is a well-recognized major modifiable risk factor for both diabetes and hypertension. Monitoring the changes in obesity has crucial medical implications for preventing the development of diabetes. Anthropometry is an extensively used, non-invasive, and cost-saving public health tool. Thus, it is of important clinical and public health significance to dig out more effective anthropometric indices related to the onset risk of diabetes among hypertensive patients. BMI is still the most widely used index of obesity for current. Yet, its reliability for determining obesity has been questioned (4–6) since it could not be used to differentiate body composition (fat mass and fat-free mass). Moreover, central obesity has recently received increasing attention because it is more closely correlated with metabolic complications, such as insulin resistance, diabetes, and CVD, than general obesity (7). Importantly, it is associated with an increased risk of diabetes among adults within a healthy BMI range (less than 25kg/m^2) (8).

Central obesity includes several established and novel parameters. Waist circumferences (WC) and waist-to-hip ratios (WHR) have been frequently used indices in clinical settings. Since WC considers abdominal obesity but ignores the height, another index, waist-to-height ratios (WHtR), has also been used as the alternative anthropometric index for predicting diabetes (9). Nevertheless, comparisons between BMI, WC, and WHtR do not seem to provide sufficient information for anyone of them to have an absolute advantage in predicting diabetes with great sensitivity or specificity (10, 11). In addition, several novel anthropometric indices have been proposed and used: abdominal volume index (AVI) (12), body adiposity index (BAI) (13), body roundness index (BRI) (14), conicity index (CI) (15), and weight-adjusted-waist-index (WWI) (16). Although the mentioned anthropometric indices have been used in various studies, their usefulness has not been systematically evaluated. Furthermore, previous studies mostly are cross-sectional designed and focus on the diabetes risk among general populations (17, 18), lacking concern for the hypertensive patients, especially those with normal-weight central obesity.

Therefore, the present study aimed to compare associations between baseline and changing trends of anthropometric indices with the development of diabetes among hypertensive patients in China. In addition, we examined associations of different combinations of BMI and established measures of central obesity (WC, WHR, and WHtR) with occurrences of diabetes and assessed the magnitude of risk for onset of diabetes from normal BMI with central obesity.

METHODS

Study Design and Study Population

The retrospective cohort study was conducted in Liaobu Town, Dongguan City, Guangdong Province, China, from 2011 to 2013. The Liaobu Town is a suburb of Dongguan, a well-developed city with a population of 0.42 million, next to the megacity Guangzhou. The subjects were recruited *via* advertisements from the general population in Liaobu community health center hospital using cluster sampling based on the following inclusion criteria: over 18 years old, with hypertension, no history of cancer, willingness to do at least 1-year follow-up. Exclusion criteria were: with self-reported diabetes, unable to respond to interviews, and without valid anthropometric indices or serum biochemical examination at baseline or at least one follow-up. Subsequently, potentially eligible subjects were further assessed based on clinical interviews, health screening questionnaires, physical examination, and fasting-blood analyses, and appropriate subjects were then included for our investigation.

Our study was approved by the Medical Research Ethics Committee of the Guangdong Provincial People's Hospital (Guangzhou, China). All participants provided written informed consents before their voluntary participation.

Health Screening Measurements

A structured health screening questionnaire was administered by healthcare staff to each qualified subject and to acquire information on demographics (age, sex, and ethnicities), lifestyle (smoking, alcohol drinking), past medical history (hypertension, diabetes, and dyslipidemia), and current medication uses, if any, for these conditions, and family history of diabetes.

Health Screening Measurements

All health screening measurements were performed by trained healthcare staff using standard anthropometric techniques. Participants were asked to wear thin clothing with no footwear when taking anthropometric measurements. Body weight, height, WC, and hip circumferences (HC) were each measured twice with the mean recorded. Body weight and height were measured by a standard digital weighing scale and stadiometer, respectively. WC and HC were each measured with the subject standing and during slight expiration using a calibrated tape measure. Waist circumference was measured at the midpoint between the iliac crest and last rib, and hip circumference was measured at the widest part of the hip at the level of the greater trochanter.

Based on the above information, other anthropometric measurements were also collected. BMIs were calculated as weight (kg)/height^2 (m). WHRs were calculated as WC (m)/HC (m), and WHtRs were calculated as WC (m)/height (m). AVIs were calculated as (2*WC^2 (cm) + 0.7*(WC (cm)–HC (cm))^2)/1000 (12). BAIs were calculated as (HC (cm)/height$^{1.5}$ (m))–18 (13). BRIs were calculated as 364.2–365.5*sqrt (1-(WC (m)/(2π))^2/(0.5*height (m))^2) (14). CIs were calculated as WC (m)/(0.109*sqrt (weight (kg)/height (m)) (15). WWIs were calculated as WC (cm)/sqrt (weight (kg)) (16) (**Table S1**).

Blood pressure was measured using mercury sphygmomanometers, and participants were required to sit quietly for 5 minutes before the measurement. Hypertension was defined as having systolic blood pressure (SBP)≥140 mmHg, or diastolic blood pressure (DBP)≥90mmHg, or with a self-reported history of hypertension, or use of antihypertensive medications.

The health screening measurements as above-mentioned were conducted at baseline and each annual follow-up.

Laboratory Examinations

Blood samples were collected in the morning after overnight fasting for at least 8 hours. Serum levels of fasting plasma glucose (FPG), triglycerides (TG), total cholesterol (TC), high-density lipoprotein (HDL), low-density lipoprotein (LDL), uric acid (UA), creatinine (Cr), and urinary albumin excretion rate (UAER) were measured *via* a biochemical autonomic analyzer (OLYMPUS, Tokyo, Japan) in the central laboratory, Liaobu community health center hospital. The estimated glomerular filtration rates (eGFR) were calculated using the CKD-EPI creatinine equation (19).

The laboratory measurements as above-mentioned were conducted at baseline and each annual follow-up.

Obesity Definition

Overweight was defined as having BMI≥24 kg/m^2 and <28 kg/m2 and obesity as ≥28 kg/m^2 according to the Working Group on Obesity in China (WGOC) (20). Central obesity was defined as WC≥90 cm for males and WC≥80 cm for females according to the International Diabetes Federation (21), or WHR≥0.90 for males and WHR≥0.85 for females according to WHO guidelines (22). Elevated WHtR was defined as ≥0.5 (9).

Lacking unifying classification standards, cut-off points for novel anthropometric indices (AVI, BAI, BRI, CI, and WWI) were selected at the level of 75% according to the distribution characteristics of BMI in the studied populations (**Table S2**).

Clinical Outcome

The outcome of the present study was new-onset diabetes, defined as self-reported physician-diagnosed diabetes and/or use of hypoglycemic medication during follow-up, or new-onset FPG≥7.0mmol/L examined at the follow-up examination. All participants were followed until the date of incident diabetes or otherwise until the last follow-up date.

Statistical Analyses

As estimated in PASS software version 15.0, 390 events would be needed in a Cox regression of the log hazard ratio (HR) to provide 90% power at a 0.05 significance level to detect a regression coefficient equal to 0.20 under an overall event rate of 0.30. For continuous variables, data in line with normal distribution were presented as mean ± standard deviation (SD), while data in line with non-normal distribution were presented as median (1st quartile, 3rd quartile). Categorical data were presented as frequencies (percentages). Differences among the groups were evaluated by the student's T-test for normally distributed continuous data, by the Kruskal-Wallis rank-sum test for non-normally distributed continuous data, and by the chi-square tests for categorical variables. Univariate Cox regression models were performed to evaluate associations of demographic, biochemical, and clinical characteristics, and anthropometric indices with diabetes. Independent effects of baseline and dynamic changes of each anthropometric index and different combinations among BMI, WC, WHR, and WHtR on the risk of diabetes were estimated using multivariate Cox regression models. Two models with different sets of covariates were fitted. Stratified and interaction analyses were also conducted to evaluate the potential interactions between WHtR and demographic, biochemical and clinical characteristics, and other anthropometric indices. P<0.05 (two-tailed) was considered statistically significant. All statistical analyses were conducted using the statistical software packages R 4.0.3 (http://www.R-project.org, The R Foundation).

RESULTS

Characteristics of the Participants

From our initial recruitment effort, 43001 subjects aged at least 18 years old underwent the clinical assessment based on clinical interviews, health screening questionnaires, physical

examination, and a fasting blood sampling. Among these subjects, 35714 valid questionnaires were returned (83.05% response rate). Based on clinical assessments, 6190 patients with hypertension were included. Finally, after excluding participants with self-reported diabetics or newly diagnosed diabetics at baseline and those who were lost to follow-ups, a total of 3852 participants were included in this analysis (**Figure 1**).

Baseline characteristics of the participants are outlined in **Table 1**. An overall 3852 subjects were studied, out of which 57.7% were females and 42.3% were males. During the median follow-up of 2 years, 1167 participants developed diabetes, 734 were females and 433 were males. Their baseline characteristics as stratified by sex are presented in **Table S3**. The anthropometric indices weight, BMI, WC, WHtR, WHR, AVI, BAI, BRI, CI, and WWI levels were statistically higher in subjects who developed diabetes (P<0.001). In addition, compared with subjects who didn't develop diabetes, those who developed diabetes during follow-up were older, had a higher proportion of females, had higher values of FPG, TG, TC, and SBP, had a higher prevalence of family history of diabetes, while with lower eGFR values and smoking rates (P<0.05).

Correlations Between Baseline Anthropometric Measures and the Development of Diabetes in Follow-Ups

Correlations between the baseline clinical variables and new-onset diabetes are displayed in **Table S4**. The univariate Cox regression analyses revealed that the development of diabetes was positively correlated with age, sex, TG, TC, and family history of diabetes, and negatively correlated with smoking status and eGFR (P<0.05).

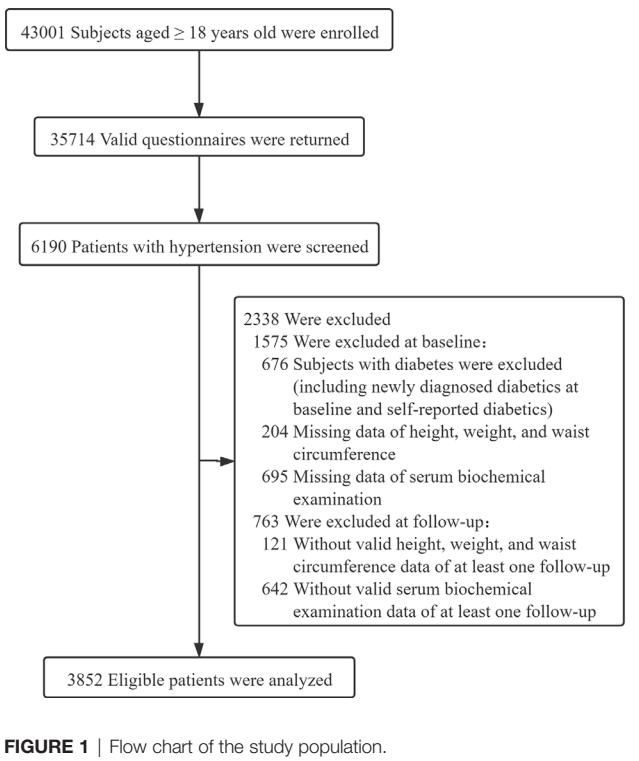


FIGURE 1 | Flow chart of the study population.

After fully adjustment for sex, age, smoking status, drinking status, and family history of diabetes at baseline, and differences of FPG, TG, TC, HDL, LDL, SBP, DBP between the baseline and follow-up, the elevation of all indices analyzed in this study were each independently associated with an increase in risk of incident diabetes (**Tables 2** and **S5**): elevated weight (HR=1.41; 95%CI: 1.31, 1.51; P<0.001), BMI (HR=1.32; 95%CI: 1.25, 1.40; P<0.001), WC (HR=1.32; 95%CI: 1.25, 1.40; P<0.001), WHtR (HR=1.27; 95%CI: 1.20, 1.36; P<0.001), WHR (HR=1.21; 95%CI: 1.15, 1.28; P<0.001), AVI (HR=1.31; 95%CI: 1.23, 1.38; P <0.001), BAI (HR=1.15; 95%CI: 1.07, 1.23; P<0.001), BRI (HR=1.26; 95%CI: 1.19, 1.33; P <0.001), CI (HR=1.14; 95%CI: 1.07, 1.22; P<0.001), WWI (HR=1.11; 95%CI: 1.04, 1.19; P=0.003).

Adopting internationally recognized diagnostic for central obesity, there was a graded increased risk of incident diabetes with successively increasing WC (HR=1.51; 95%CI: 1.30, 1.74; P<0.001), WHtR (HR=1.61; 95%CI: 1.32, 1.98; P<0.001) and WHR (HR=1.47; 95%CI: 1.24, 1.75; P<0.001). Moreover, selecting the level of 75% for the novel anthropometric indices as the cut-off point, the new-onset diabetes risk increased when any of the following conditions was met: AVI≥18 (HR=1.53; 95% CI: 1.34, 1.73 P<0.001), BAI≥34 (HR=1.25; 95%CI: 1.08, 1.44; P=0.002), BRI≥5.5 (HR=1.38; 95%CI: 1.21, 1.58; P<0.001), CI≥1.35 (HR=1.21; 95%CI: 1.04, 1.40; P<0.001), WWI≥11.5 (HR=1.25; 95%CI: 1.08, 1.44; P=0.002) (**Figure 2**).

New-onset diabetes risk increased significantly with the baseline WHtR levels above 0.5 whether the BMI, WC, and WHR were within the normal range (BMI<24kg/m^2, WC<90cm in males or <80cm in females, WHR<0.90 in males or <0.85 in females) or not at baseline. Elevated WHtR (WHtR≥0.5) at baseline was significantly associated with increased diabetes risk with baseline BMI<24kg/m^2 (HR=1.37; 95%CI: 1.08, 1.75; P =0.011), WC<90cm in males or <80cm in females (HR=1.34; 95%CI: 1.06, 1.71; P =0.015), WHR<0.90 in males or <0.85 in females (HR=1.62; 95%CI: 1.18, 2.22; P=0.003). The risk increased further when BMI>24kg/m^2 (HR=1.85; 95%CI: 1.48, 2.31; P<0.001), WC≥90cm in males or ≥80cm in females (HR=1.77; 95%CI: 1.43, 2.18; P<0.001), and WHR≥0.90 in males or ≥0.85 in females (HR=1.84; 95%CI: 1.42, 2.37; P<0.001) at baseline. The highest risk for incident diabetes (HR=2.28; 95%CI: 1.72, 3.03; P<0.001) was observed when BMI, WC, WHR, WHtR were all greater than the critical value (**Table 3** and **Figure 3**). Interaction and stratified analyses revealed that there were no significant interactions between WHtR and other clinical variables (**Table S7**).

Correlations Between Dynamic Changes of Anthropometric Measures and the Development of Diabetes During Follow-Ups

As shown in **Tables 4**, **S6**, and **Figure 4**, in the fully adjusted model, compared with the subjects whose WHtR was less than 0.5 at baseline and follow-up, elevated WHtR (WHtR≥0.5) at baseline or follow-up was associated with a higher risk of developing diabetes (P<0.05). When WHtR≥0.5 was detected at

TABLE 1 | Baseline characteristics of subjects who did and didn't develop new-onset diabetes during follow-up.

	Total	Diabetes (n = 1167)	Non-diabetes (n = 2685)	*P*-value
Age (years)	61.8 ± 13.6	62.8 ± 12.3	61.3 ± 14.1	0.002**
Sex (male (n (%))	1632 (42.37%)	433 (37.10%)	1199 (44.66%)	<0.001***
FPG (mmol/L)	5.44 ± 1.68	6.53 ± 2.61	4.97 ± 0.60	<0.001***
TG (mmol/L)	2.12 ± 1.71	1.90 (1.32-2.81)	1.61 (1.14-2.27)	<0.001***
TC (mmol/L)	5.06 ± 1.26	5.13 ± 1.19	5.03 ± 1.29	0.025*
HDL (mmol/L)	1.32 ± 0.36	1.32 ± 0.40	1.32 ± 0.33	0.744
LDL (mmol/L)	2.88 ± 0.80	2.88 ± 0.83	2.88 ± 0.79	0.924
UA (μmol/L)	381.71 ± 104.11	382.70 ± 103.89	381.28 ± 104.22	0.702
Scr (μmol/L)	73.00 (62.00-88.00)	72.00 (61.00-88.00)	73.00 (62.00-88.00)	0.587
eGFR (mL/(min·1.73 m^2))	83.62 ± 23.77	81.84 ± 22.43	84.39 ± 24.29	0.002**
Weight (kg)	62.63 ± 12.46	64.36 ± 12.04	61.88 ± 12.57	<0.001***
BMI (kg/m^2)	25.37 ± 3.94	26.19 ± 3.92	25.02 ± 3.90	<0.001***
WC (cm)	87.93 ± 9.83	90.22 ± 9.71	86.93 ± 9.71	<0.001***
WHtR	0.56 ± 0.07	0.58 ± 0.07	0.56 ± 0.06	<0.001***
WHR	0.92 ± 0.06	0.93 ± 0.06	0.92 ± 0.06	<0.001***
AVI	15.72 ± 3.48	16.53 ± 3.55	15.37 ± 3.40	<0.001***
BAI	30.91 ± 5.34	31.76 ± 5.46	30.54 ± 5.25	<0.001***
BRI	4.68 ± 1.43	5.02 ± 1.48	4.54 ± 1.39	<0.001***
CI	1.28 ± 0.09	1.30 ± 0.09	1.28 ± 0.09	<0.001***
WWI	11.19 ± 0.94	11.31 ± 0.90	11.14 ± 0.95	<0.001***
SBP	160.40 ± 22.64	161.62 ± 23.27	159.87 ± 22.35	0.027*
DBP	94.54 ± 11.56	94.10 ± 11.72	94.74 ± 11.48	0.119
Smoking (n (%))	743 (19.29%)	190 (16.28%)	553 (20.60%)	0.002**
Drinking (n (%))	199 (5.17%)	49 (4.20%)	150 (5.59%)	0.074
Family history of diabetes (n (%))	91 (2.39%)	55 (4.74%)	36 (1.36%)	<0.001***

Continuous data are shown as the mean ± SD or median (Q1-Q3), and categorical data as n (%).

FPG, fasting plasma glucose; TG, triglycerides; TC, total cholesterol; HDL, high-density lipoprotein; LDL, low-density lipoprotein; UA, urid acid; Scr, serum creatinine; eGFR, estimated glomerular filtration rate; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio; WHR, waist-to-hip ratio; AVI, abdominal volume index; BAI, body adiposity index; BRI, body roundness index; CI, conicity index; WWI, weight-adjusted-waist index; SBP, systolic blood pressure; DBP, diastolic blood pressure.

**P-value < 0.05; **P-value < 0.01; ***P-value < 0.001.*

TABLE 2 | Multivariate cox regression models evaluating the associations of baseline established anthropometric indices with the development of diabetes.

	Unadjusted model		Model 1		Model 2	
	HR (95%CI)	*P*-value	HR (95%CI)	*P*-value	HR (95%CI)	*P*-value
Weight (kg)						
As continuous variables (per SD increment)	1.18 (1.12, 1.25)	<0.001***	1.44 (1.35, 1.54)	<0.001***	1.41 (1.31, 1.51)	<0.001***
<75 in males or <65 in females	1.0		1.0		1.0	
≥75 in males or ≥65 in females	1.40 (1.24, 1.59)	<0.001***	1.56 (1.37, 1.77)	<0.001***	1.52 (1.33, 1.74)	<0.001***
BMI (kg/m^2)						
As continuous variables (per SD increment)	1.27 (1.21, 1.34)	<0.001***	1.33 (1.26, 1.40)	<0.001***	1.32 (1.25, 1.40)	<0.001***
<24.0	1.0		1.0		1.0	
24.0-28.0	1.40 (1.22, 1.61)	<0.001***	1.46 (1.27, 1.69)	<0.001***	1.37 (1.19, 1.59)	<0.001***
≥28.0	1.80 (1.54, 2.09)	<0.001***	1.92 (1.64, 2.24)	<0.001***	1.79 (1.53, 2.11)	<0.001***
WC (cm)						
As continuous variables (per SD increment)	1.34 (1.26, 1.41)	<0.001***	1.36 (1.28, 1.44)	<0.001***	1.32 (1.25, 1.40)	<0.001***
<90 in males or <80 in females	1.0		1.0		1.0	
≥90 in males or ≥80 in females	1.64 (1.44, 1.88)	<0.001***	1.62 (1.41, 1.87)	<0.001***	1.51 (1.30, 1.74)	<0.001***
WHtR						
As continuous variables (per SD increment)	1.33 (1.26, 1.41)	<0.001***	1.31 (1.24, 1.39)	<0.001***	1.27 (1.20, 1.36)	<0.001***
<0.5	1.0		1.0		1.0	
≥0.5	1.80 (1.48, 2.19)	<0.001***	1.72 (1.41, 2.10)	<0.001***	1.61 (1.32, 1.98)	<0.001***
WHR						
As continuous variables (per SD increment)	1.22 (1.16, 1.28)	<0.001***	1.23 (1.16, 1.29)	<0.001***	1.21 (1.15, 1.28)	<0.001***
<0.90 in males or <0.85 in females	1.0		1.0		1.0	
≥0.90 in males or ≥0.85 in females	1.61 (1.37, 1.90)	<0.001***	1.54 (1.30, 1.82)	<0.001***	1.47 (1.24, 1.75)	<0.001***

BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio; WHR, waist-to-hip ratio.

Model 1: adjusted by sex, age, smoking status, drinking status, and family history of diabetes at baseline.

Model 2: adjusted by model 1 plus differences of FPG, TG, TC, HDL, LDL, SBP, DBP between the baseline and follow-up.

****P -value < 0.001.*

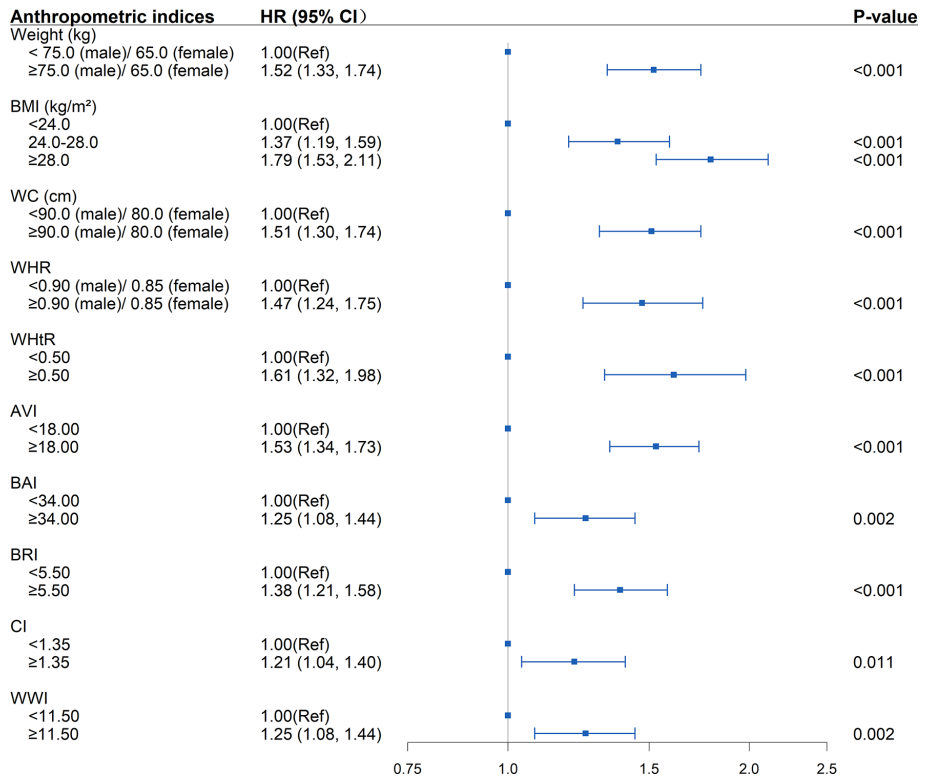


FIGURE 2 | Association between separate anthropometric indices with the development of diabetes (weight, body mass index [BMI], waist circumference [WC], waist-to-hip ratio [WHR], waist-to-height ratio [WHtR], abdominal volume index [AVI], body adiposity index [BAI], body roundness index [BRI], conicity index [CI], weight-adjusted-waist index [WWI]). The correlation was assessed by multivariate cox regression analysis, adjusted by sex, age, smoking status, drinking status, and family history of diabetes at baseline and differences of FPG, TG, TC, HDL, LDL, SBP, DBP between the baseline and follow-up. Hazard ratios (HRs) of the anthropometric indices were represented as the squares and 95% confidence intervals (CIs) by the lines through the squares.

baseline, the risk of diabetes tended to be higher in subjects with WHtR≥0.5 at follow-up (HR=2.04; 95%CI: 1.54, 2.71; P<0.001) than those whose WHtR returned to less than 0.5 (HR=1.93; 95% CI: 1.36, 2.72; P<0.001). The highest risk of diabetes onset was observed when WHtR≥0.5 both at baseline and follow-up. Among the subjects with BMI within the normal range at the baseline, compared with the subjects who remained BMI<24kg/m^2 during the follow-up, subjects whose BMI became overweight or obese at follow-up were not at a significant increase in risk for diabetes. The same was observed in WC and WHR.

Other significant associations between the changing trends in anthropometric indices and the development of diabetes included elevated AVI (AVI≥18) at baseline and/or follow-up, elevated BAI, BRI, CI, WWI at baseline (BAI≥34, BRI≥5.5, CI≥1.35, WWI≥11.5) regardless of returning to less than the critical value during follow-up or not, weight loss ≥5 kg, and weight gain ≥5 kg (P<0.05).

DISCUSSION

Diabetes and hypertension frequently coexist in patients, and both are established risk factors for CVD (2, 23). Individuals with diabetes and hypertension have higher morbidity and mortality of CVD compared with those with either disease alone (3). Therefore, it is crucial to explore and intervene in the risk factors for developing diabetes in the hypertension population. Hypertension could be easily identified by non-invasive BP measurements, yet diabetes often goes undetected until patients present with diabetic complications. Anthropometric measurements have been widely used in clinical screening for CVD and metabolic syndrome (MetS), owing to their simple, low-cost, quick, and non-invasive characteristics. The present study was conducted to compare the strength of associations between different anthropometric indices with the development of diabetes in the hypertension population.

In the present cohort of patients with hypertension, our data indicate that anthropometric measurements analyzed in this study, BMI, WC, WHtR, WHR, AVI, BAI, BRI, CI, and WWI, were each independently associated with increased risk for the development of diabetes. Among these indices, WHtR had the strongest association with the new-onset diabetic risk, with dynamic changes showing stronger associations than BMI, WC, and WHR. Additionally, regardless of whether the BMI, WC, and WHR were within the normal range or not, elevated WHtR at baseline was associated with an increased risk of diabetes. Our

TABLE 3 | Multivariate cox regression models evaluating the associations of different combinations of BMI and established anthropometric indices of central obesity (WC, WHR and WHtR) with the development of diabetes.

	Unadjusted model		Model 1		Model 2	
	HR (95%CI)	*P*-value	HR (95%CI)	*P*-value	HR (95%CI)	*P*-value
BMI (kg/m^2) & WC (cm)						
BMI<24 & WC<90 (male)/80 (female)	1.0		1.0		1.0	
BMI≥24 & WC<90 (male)/80 (female)	1.26 (1.00, 1.60)	0.053	1.34 (1.05, 1.71)	0.017*	1.32 (1.04, 1.69)	0.025*
BMI<24 & WC≥90 (male)/80 (female)	1.42 (1.14, 1.76)	0.002**	1.31 (1.05, 1.64)	0.019*	1.27 (1.01, 1.60)	0.042*
BMI≥24 & WC≥90 (male)/80 (female)	1.87 (1.59, 2.19)	<0.001***	1.88 (1.60, 2.22)	<0.001***	1.73 (1.46, 2.05)	<0.001***
BMI (kg/m^2) & WHtR						
BMI<24 & WHtR<0.5	1.0		1.0		1.0	
BMI≥24 & WHtR<0.5	1.39 (0.82, 2.37)	0.219	1.61 (0.94, 2.74)	0.081	1.55 (0.89, 2.68)	0.119
BMI<24 & WHtR≥0.5	1.50 (1.18, 1.90)	<0.001***	1.37 (1.08, 1.75)	0.011*	1.34 (1.05, 1.72)	0.020*
BMI≥24 & WHtR≥0.5	2.03 (1.64, 2.52)	<0.001***	2.00 (1.61, 2.48)	<0.001***	1.85 (1.48, 2.31)	<0.001***
BMI (kg/m^2) & WHR						
BMI<28 & WHR<0.90 (male)/0.85 (female)	1.0		1.0		1.0	
BMI≥28 & WHR<0.90 (male)/0.85 (female)	1.62 (1.19, 2.19)	0.002**	1.72 (1.26, 2.34)	<0.001***	1.67 (1.21, 2.30)	0.002**
BMI<28 & WHR≥0.90 (male)/0.85 (female)	1.56 (1.22, 2.00)	<0.001***	1.43 (1.11, 1.84)	0.005**	1.43 (1.11, 1.85)	0.006**
BMI≥28 & WHR≥0.90 (male)/0.85 (female)	2.16 (1.73, 2.70)	<0.001***	2.10 (1.68, 2.64)	<0.001***	1.97 (1.56, 2.48)	<0.001***
WC (cm) & WHtR						
WC<90 (male)/80 (female) & WHtR<0.5	1.0		1.0		1.0	
WC≥90 (male)/80 (female) & WHtR<0.5	2.90 (0.92, 9.13)	0.069	2.40 (0.76, 7.62)	0.137	1.95 (0.48, 7.99)	0.351
WC<90 (male)/80 (female) & WHtR≥0.5	1.38 (1.09, 1.74)	0.008**	1.34 (1.06, 1.71)	0.015*	1.31 (1.03, 1.67)	0.030*
WC≥90 (male)/80 (female) & WHtR≥0.5	1.99 (1.63, 2.43)	<0.001***	1.93 (1.57, 2.37)	<0.001***	1.77 (1.43, 2.18)	<0.001***
WC (cm) & WHR						
WC<90 (male)/80 (female) & WHR<0.90 (male)/0.85 (female)	1.0		1.0		1.0	
WC≥90 (male)/80 (female) & WHR<0.90 (male)/0.85 (female)	1.62 (1.16, 2.28)	0.005**	1.66 (1.17, 2.37)	0.005**	1.57 (1.09, 2.26)	0.016*
WC<90 (male)/80 (female) & WHR≥0.90 (male)/0.85 (female)	1.34 (1.06, 1.69)	0.013*	1.31 (1.04, 1.65)	0.023*	1.31 (1.03, 1.66)	0.026*
WC≥90 (male)/80 (female) & WHR≥0.90 (male)/0.85 (female)	1.96 (1.62, 2.37)	<0.001***	1.90 (1.56, 2.32)	<0.001***	1.77 (1.44, 2.17)	<0.001***
WHR & WHtR						
WHR<0.90 (male)/0.85 (female) & WHtR<0.5	1.0		1.0		1.0	
WHR≥0.90 (male)/0.85 (female) & WHtR<0.5	1.53 (1.05, 2.24)	0.028*	1.47 (1.00, 2.16)	0.049*	1.37 (0.92, 2.05)	0.119
WHR<0.90 (male)/0.85 (female) & WHtR≥0.5	1.65 (1.21, 2.25)	0.002**	1.62 (1.18, 2.22)	0.003***	1.48 (1.07, 2.04)	0.017*
WHR≥0.90 (male)/0.85 (female) & WHtR≥0.5	2.15 (1.68, 2.76)	<0.001***	2.02 (1.57, 2.60)	<0.001	1.84 (1.42, 2.37)	<0.001***
BMI, WC, WHtR & WHR						
All indicators were normal	1.0		1.0		1.0	
Any one indicator was abnormal	1.61 (1.14, 2.28)	0.007**	1.60 (1.13, 2.27)	0.008**	1.54 (1.08, 2.19)	0.018*
Any two indicators were abnormal	1.71 (1.23, 2.36)	0.001**	1.65 (1.19, 2.29)	0.003**	1.60 (1.15, 2.23)	0.006**
Any three indicators were abnormal	1.91 (1.42, 2.57)	<0.001***	1.81 (1.35, 2.45)	<0.001***	1.71 (1.26, 2.31)	<0.001***
All indicators were abnormal	2.61 (1.98, 3.45)	<0.001***	2.54 (1.92, 3.36)	<0.001***	2.28 (1.72, 3.03)	<0.001***

BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio; WHR, waist-to-hip ratio.
Model 1: adjusted by sex, age, smoking status, drinking status, and family history of diabetes at baseline.
Model 2: adjusted by model 1 plus differences of FPG, TG, TC, HDL, LDL, SBP, DBP between the baseline and follow-up.
**P-value < 0.05; **P -value < 0.01; ***P -value < 0.001.*

results were consistent with a previous robust meta-analysis which shows that measures of abdominal obesity were better indicators for obesity-related cardiometabolic risk than BMI, and WHtR was a better screening tool than WC and BMI for diabetes, hypertension, and CVD (9). Previous studies have considered mechanisms to explain why measures of central obesity are better than BMI in predicting diabetes. Yet, there are few related studies on why should WHtR be superior to WC. We speculate that on the one hand, for the adults whose height is generally stable, the change of WHtR is essentially the change in WC, whereas WHtR partially precludes the influence of age and sex. On the other hand, adverse early life exposures lead to short stature and are also closely associated with predisposition to abdominal obesity and insulin resistance in adults (9), which is biologically plausible.

For identification of central obesity, WC, WHR, and WHtR measurements have often been used. Our data indicated that for the population with hypertension, normal weight but central obesity was also associated with elevated risk for diabetes. Our finding is consistent with a previous report that Asian populations are susceptible to develop diabetes despite having relatively lower BMI than other ethnicities (24). This indicates that abdominal obesity may be a more useful indicator than BMI for diabetes, especially for hypertensive patients. Several potential mechanisms could be used to explain our findings. To begin with, ectopic fat accumulation, whose marker is abdominal fat, has been confirmed to increase the risk of metabolic abnormality and future development of diabetes (25, 26). Additionally, compared with subcutaneous fat, visceral fat with abdominal cavities is related to higher metabolic and inflammatory activities, thus prompting the development of diabetes (27). Further, normal weight with central obesity indicates that such individuals have excessive visceral fat, and

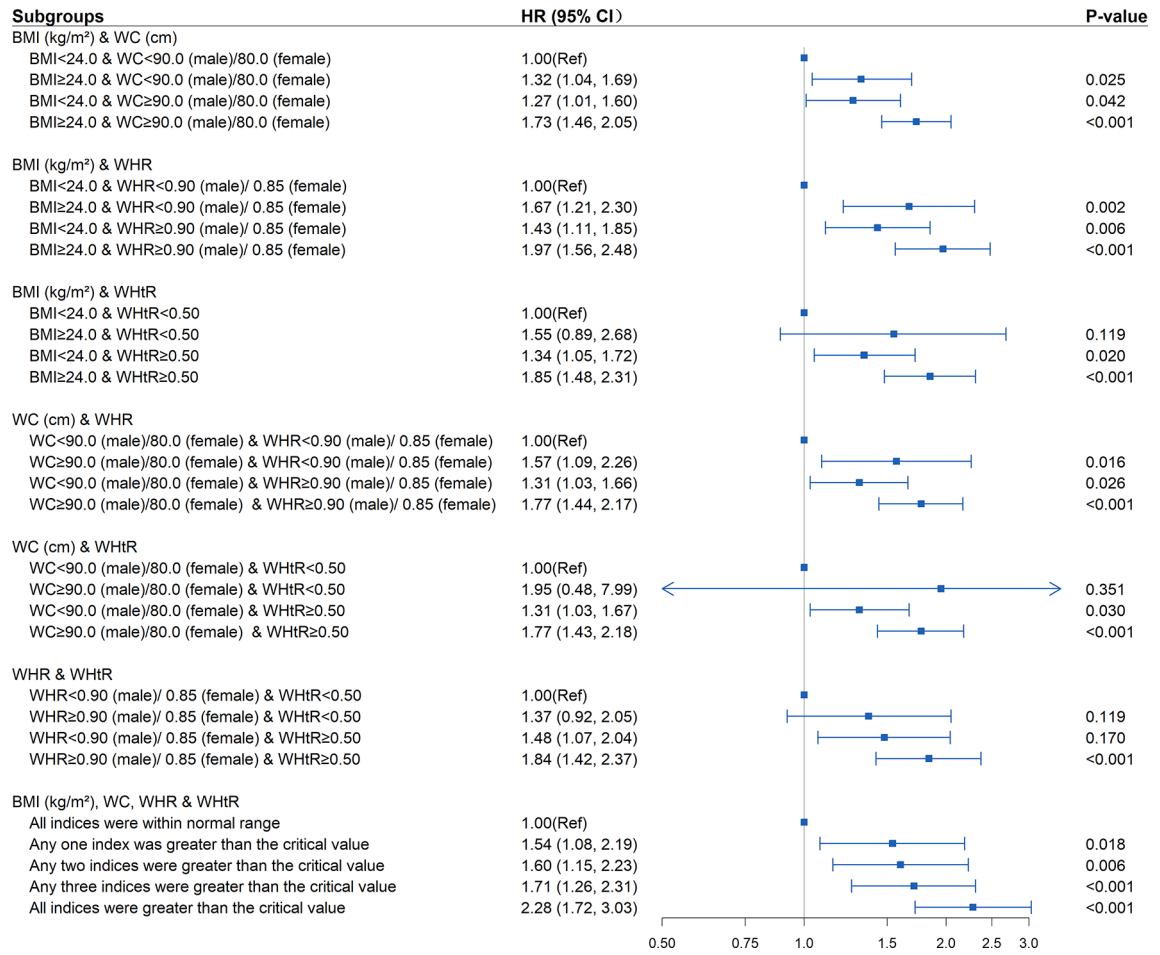


FIGURE 3 | Association between different combinations of body mass index (BMI) and established anthropometric indices of central obesity (waist circumference [WC], waist-to-hip ratio [WHR], waist-to-height ratio [WHtR]) with the development of diabetes. The correlation was assessed by multivariate cox regression analysis, adjusted by sex, age, smoking status, drinking status, and family history of diabetes at baseline and differences of FPG, TG, TC, HDL, LDL, SBP, DBP between the baseline and follow-up. Hazard ratios (HRs) of the combined anthropometric indices were represented as the squares and 95% confidence intervals (CIs) by the lines through the squares.

their normal BMI usually means they are at higher risk of less muscle compared with the same BMI but no central obesity. And the lack of muscle mass has been confirmed to be associated with adverse metabolic profiles (28). At the same time, it's interesting to note that the increased risk of diabetes among hypertensive patients with overweight/obesity but with WHtR<0.5 was not statistically significant, further revealing that the development of diabetes is more closely related to the distribution rather than the absolute degree of adiposity per se. Therefore, the indices of central obesity could be measured in addition to BMI to identify the patients with normal-weight central obesity who are also at high risk of diabetes, and thus provide incremental benefit in the pre-screening of hypertensive patients with diabetes.

Our study revealed a trend to reduce the onset risk of diabetes when WC, WHR, and WHtR were reversed towards normal from abnormal levels. In addition, the diabetes risk could also be observed to be increased with increasing WHtR during follow-up. Of note, the changing trends of AVI performed similarly in reflecting the risk of T2DM as WHtR, showing superior sensitivity of both its baseline value and dynamic changes on reflecting the development than other indices, which suggests that AVI could also be effective predictive indicators of diabetes. It's well documented that the development of diabetes could be delayed or prevented through lifestyle intervention, including dietary modification, weight loss, and exercise training. Therefore, through long-term monitoring of these non-invasive and straightforward measures and applying timely lifestyle intervention, it's expected to promote the switch from abnormal towards normal levels of these established or novel indices of central obesity, which is essential for preventing or delaying the development of diabetes.

Our study has important implications for public health and clinical practice. First, according to the current established guidelines, individuals with normal weight based on BMI, regardless of central obesity status, were generally regarded as normal in clinical practice. This could lead to a missed

TABLE 4 | Multivariate cox regression models evaluating the associations of dynamic changes of established anthropometric indices with the development of diabetes.

	Unadjusted model		Model 1		Model 2	
	HR (95%CI)	***P*-value**	**HR (95%CI)**	***P*-value**	**HR (95%CI)**	***P*-value**
Weight change (kg)						
<5	1.0		1.0		1.0	
Loss ≥5	1.32 (1.12, 1.55)	<0.001***	1.33 (1.13, 1.56)	<0.001***	1.40 (1.13, 1.74)	0.002**
Gain ≥5	1.47 (1.23, 1.76)	<0.001***	1.45 (1.21, 1.74)	<0.001***	1.35 (1.05, 1.73)	0.019*
Dynamic changes of BMI (kg/m^2)						
<24 at baseline & <24 at follow-up	1.0		1.0		1.0	
<24 at baseline & ≥24 at follow-up	1.38 (1.08, 1.76)	0.010***	1.36 (1.07, 1.74)	0.014*	1.30 (1.00, 1.69)	0.053
≥24 at baseline & <24 at follow-up	1.32 (1.04, 1.68)	0.023*	1.35 (1.06, 1.72)	0.014*	1.32 (1.03, 1.69)	0.031*
≥24 at baseline & ≥24 at follow-up	1.68 (1.45, 1.95)	<0.001***	1.76 (1.51, 2.05)	<0.001***	1.70 (1.45, 1.98)	<0.001***
Dynamic changes of WC (cm)						
<90 (male)/80 (female) at baseline & <90 (male)/80 (female) at follow-up	1.0		1.0		1.0	
<90 (male)/80 (female) at baseline & ≥90 (male)/80 (female) at follow-up	1.27 (0.98, 1.65)	0.068	1.35 (1.03, 1.76)	0.029*	1.19 (0.89, 1.58)	0.245
≥90 (male)/80 (female) at baseline & <90 (male)/80 (female) at follow-up	1.59 (1.18, 2.14)	0.002**	1.60 (1.19, 2.16)	0.002**	1.45 (1.05, 2.00)	0.023*
≥90 (male)/80 (female) at baseline & ≥90 (male)/80 (female) at follow-up	1.99 (1.58, 2.51)	<0.001***	2.00 (1.58, 2.53)	<0.001***	1.74 (1.36, 2.24)	<0.001***
Dynamic changes of WHtR						
<0.5 at baseline & <0.5 at follow-up	1.0		1.0		1.0	
<0.5 at baseline & ≥0.5 at follow-up	1.80 (1.23, 2.64)	0.003**	1.71 (1.16, 2.51)	0.006**	1.63 (1.11, 2.40)	0.014*
≥0.5 at baseline & <0.5 at follow-up	2.02 (1.44, 2.84)	<0.001***	1.98 (1.41, 2.78)	<0.001***	1.93 (1.36, 2.72)	<0.001***
≥0.5 at baseline & ≥0.5 at follow-up	2.33 (1.77, 3.07)	<0.001***	2.18 (1.65, 2.88)	<0.001***	2.04 (1.54, 2.71)	<0.001***
Dynamic changes of WHR						
<0.90 (male)/0.85 (female) at baseline & <0.90 (male)/0.85 (female) at follow-up	1.0		1.0		1.0	
<0.90 (male)/0.85 (female) at baseline & ≥0.90 (male)/0.85 (female) at follow-up	1.33 (0.75, 2.34)	0.330	1.30 (0.72, 2.33)	0.390	1.44 (0.80, 2.59)	0.229
≥0.90 (male)/0.85 (female) at baseline & <0.9 (male)/0.85 (female) at follow-up	1.22 (0.93, 1.60)	0.158	1.21 (0.92, 1.60)	0.164	1.22 (0.92, 1.61)	0.162
≥0.90 (male)/0.85 (female) at baseline & ≥0.90 (male)/0.85 (female) at follow-up	1.69 (1.42, 2.01)	<0.001***	1.60 (1.34, 1.92)	<0.001***	1.56 (1.30, 1.87)	<0.001***

BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio; WHR, waist-to-hip ratio.

Model 1: adjusted by sex, age, smoking status, drinking status, and family history of diabetes at baseline.

Model 2: adjusted by model 1 plus differences of FPG, TG, TC, HDL, LDL, SBP, DBP between the baseline and follow-up.

**P-value < 0.05; **P-value < 0.01; ***P-value < 0.001.*

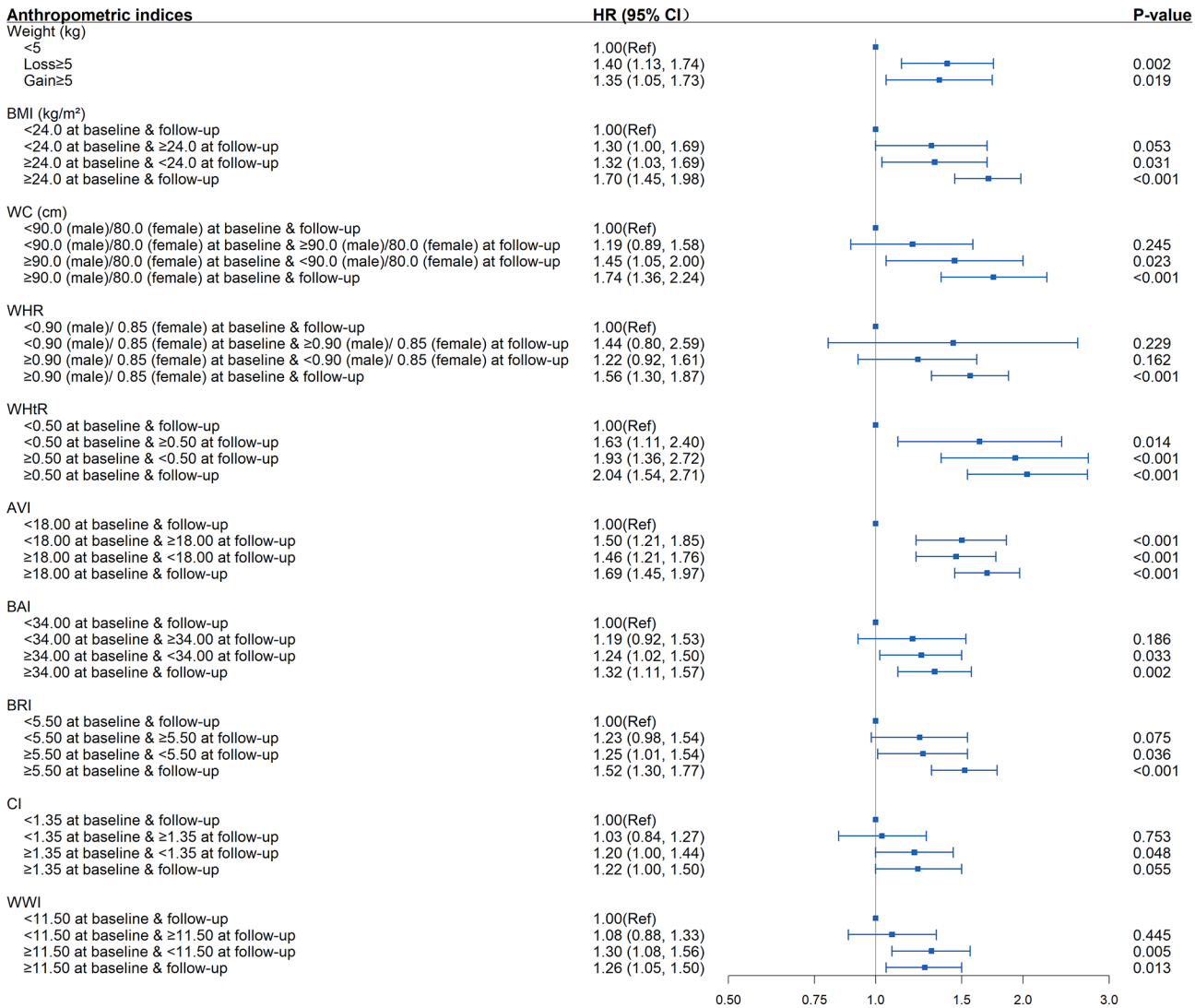


FIGURE 4 | Association between dynamic changes of separate anthropometric indices with the development of diabetes (weight, body mass index [BMI], waist circumference [WC], waist-to-hip ratio [WHR], waist-to-height ratio [WHtR], abdominal volume index [AVI], body adiposity index [BAI], body roundness index [BRI], conicity index [CI], weight-adjusted-waist index [WWI]). The correlation was assessed by multivariate cox regression analysis, adjusted by sex, age, smoking status, drinking status, and family history of diabetes at baseline and differences of FPG, TG, TC, HDL, LDL, SBP, DBP between the baseline and follow-up. Hazard ratios (HRs) of the anthropometric indices were represented as the squares and 95% confidence intervals (CIs) by the lines through the squares.

opportunity for timely evaluation and intervention for a subgroup that is at high risk but easily neglected (i.e., those with normal-weight central obesity). Second, since all the anthropometric indices of central obesity were calculated based on WC, which could be easily implemented in different levels of hospitals with only a tape used and simple standardized training of the healthcare personnel, WC is recommended to be routinely obtained in daily clinical practice. Third, our findings suggested that the dynamic changes of WC, WHR, and WHtR could sensitively reflect the variation of diabetes onset risk. Thus, the decrease in WC should be the vital focus for public health preventive interventions for diabetes since the height remains nearly unchanged.

The following limitations should be taken into account when interpreting our findings. First, this was a monocentric study. Although the study was conducted in a representative population in China, our findings might not be extrapolated to other populations in China or Asia. Second, some known risk factors for diabetes, such as dietary habits and physical activity status, were not collected, and their impact on our study could not be adjusted. Third, due to the lack of uniform criteria for the novel anthropometric indices in the Chinese population, the 75% value was selected as the cut-off point to explore the association with diabetes risk in the present study. Fourth, the retrospective nature of the study is a limitation since we could not account for those with missing data, and those lost to follow-up

(including deaths). Last, the diagnosis of new diabetes is based on suboptimal criteria. Therefore, further studies with larger sample size and multicenter design are needed to confirm our findings. Despite these limitations, our study still has some important strengths, including the cohort study design, which could establish the temporal sequencing of a causal association. Additionally, except for the baseline values of anthropometric indices, we also examined the associations of different combinations of BMI and these indices and changing trends of these indices with diabetes risk, providing deeper insights into the role of central obesity on diabetes. Last, the anthropometric measures were collected directly by the trained healthcare personnel instead of self-reported by the participants.

In conclusion, our study has demonstrated that central obesity is a significant, independent, and modifiable risk factor for diabetes among the population with hypertension. Measuring indices of central obesity, especially WHtR, in addition to BMI in clinics could provide incremental benefits in the discrimination of diabetes in Chinese hypertensive patients. Moreover, we suggest that AVI might be a promising predictor for diabetes screening.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, upon reasonable request.

ETHICS STATEMENT

Our study was approved by the Medical Research Ethics Committee of the Guangdong Provincial People's Hospital (Guangzhou, China). All participants provided written informed consents before their voluntary participation.

AUTHOR CONTRIBUTIONS

All authors have contributed to the creation of this manuscript for important intellectual content. Conceptualization, JK and HG. Methodology, JK, HC, and HG. Validation, YX and QH. Formal Analysis, YL and XL. Investigation, XF. Resources, QZ. Data Curation, SZ. Writing — original draft preparation, YL. Writing – review and editing, JK and HC. Visualization, XL. Supervision, JK. Project administration, JK. Funding acquisition, JK and HC. All authors contributed to the article and approved the submitted version.

FUNDING

This work was supported by the National Key R&D Program of China under Grant No. 2018YFC1314100, the Key-Area Research and Development Program of Guangdong Province under Grant No. 2019B020230001, and the Science and Technology Plan of Guangzhou under Grant No. 201707010330.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2021.736077/full#supplementary-material

REFERENCES

1. Sarwar N, Gao P, Seshasai SR, Gobin R, Kaptoge S, Di Angelantonio E, et al. Diabetes Mellitus, Fasting Blood Glucose Concentration, and Risk of Vascular Disease: A Collaborative Meta-Analysis of 102 Prospective Studies. *Lancet* (2010) 375:2215–22. doi: 10.1016/s0140-6736(10)60484-9
2. Bress AP, King JB, Kreider KE, Beddhu S, Simmons DL, Cheung AK, et al. Effect of Intensive Versus Standard Blood Pressure Treatment According to Baseline Prediabetes Status: A *Post Hoc* Analysis of a Randomized Trial. *Diabetes Care* (2017) 40:1401–8. doi: 10.2337/dc17-0885
3. Ferrannini E, Cushman WC. Diabetes and Hypertension: The Bad Companions. *Lancet* (2012) 380:601–10. doi: 10.1016/s0140-6736(12)60987-8
4. Nuttall FQ. Body Mass Index: Obesity, BMI, and Health: A Critical Review. *Nutr Today* (2015) 50:117–28. doi: 10.1097/nt.0000000000000092
5. Kopelman PG. Obesity as a Medical Problem. *Nature* (2000) 404:635–43. doi: 10.1038/35007508
6. Wellens RI, Roche AF, Khamis HJ, Jackson AS, Pollock ML, Siervogel RM. Relationships Between the Body Mass Index and Body Composition. *Obes Res* (1996) 4:35–44. doi: 10.1002/j.1550-8528.1996.tb00510.x
7. American Diabetes Association. Standards of Medical Care in Diabetes–2014. *Diabetes Care* (2014) 37(Suppl 1):S14–80. doi: 10.2337/dc14-S014
8. Centers for Disease Control and Prevention (CDC). Prevalence of Diabetes and Impaired Fasting Glucose in Adults–United States, 1999-2000. *JAMA* (2003) 290(13):1702–3. doi: 10.1001/jama.290.13.1702
9. Ashwell M, Gunn P, Gibson S. Waist-To-Height Ratio Is a Better Screening Tool Than Waist Circumference and BMI for Adult Cardiometabolic Risk Factors: Systematic Review and Meta-Analysis. *Obes Rev* (2012) 13:275–86. doi: 10.1111/j.1467-789X.2011.00952.x
10. Sommer I, Teufer B, Szelag M, Nussbaumer-Streit B, Titscher V, Klerings I, et al. The Performance of Anthropometric Tools to Determine Obesity: A Systematic Review and Meta-Analysis. *Sci Rep* (2020) 10:12699. doi: 10.1038/s41598-020-69498-7
11. Kodama S, Horikawa C, Fujihara K, Heianza Y, Hirasawa R, Yachi Y, et al. Comparisons of the Strength of Associations With Future Type 2 Diabetes Risk Among Anthropometric Obesity Indicators, Including Waist-to-Height Ratio: A Meta-Analysis. *Am J Epidemiol* (2012) 176:959–69. doi: 10.1093/aje/kws172
12. Perona JS, Schmidt Rio-Valle J, Ramírez-Vélez R, Correa-Rodríguez M, Fernández-Aparicio Á, González-Jiménez E. Waist Circumference and Abdominal Volume Index Are the Strongest Anthropometric Discriminators of Metabolic Syndrome in Spanish Adolescents. *Eur J Clin Invest* (2019) 49:e13060. doi: 10.1111/eci.13060
13. Bergman RN, Stefanovski D, Buchanan TA, Sumner AE, Reynolds JC, Sebring NG, et al. A Better Index of Body Adiposity. *Obes (Silver Spring)* (2011) 19:1083–9. doi: 10.1038/oby.2011.38
14. Thomas DM, Bredlau C, Bosy-Westphal A, Mueller M, Shen W, Gallagher D, et al. Relationships Between Body Roundness With Body Fat and Visceral Adipose Tissue Emerging From a New Geometrical Model. *Obes (Silver Spring)* (2013) 21:2264–71. doi: 10.1002/oby.20408
15. Valdez R. A Simple Model-Based Index of Abdominal Adiposity. *J Clin Epidemiol* (1991) 44:955–6. doi: 10.1016/0895-4356(91)90059-i
16. Park Y, Kim NH, Kwon TY, Kim SG. A Novel Adiposity Index as an Integrated Predictor of Cardiometabolic Disease Morbidity and Mortality. *Sci Rep* (2018) 8:16753. doi: 10.1038/s41598-018-35073-4

17. Zhang FL, Ren JX, Zhang P, Jin H, Qu Y, Yu Y, et al. Strong Association of Waist Circumference (WC), Body Mass Index (BMI), Waist-To-Height Ratio (WHtR), and Waist-To-Hip Ratio (WHR) With Diabetes: A Population-Based Cross-Sectional Study in Jilin Province, China. *J Diabetes Res* (2021) 2021:8812431. doi: 10.1155/2021/8812431
18. Skogberg N, Laatikainen T, Lundqvist A, Lilja E, Härkänen T, Koponen P. Which Anthropometric Measures Best Indicate Type 2 Diabetes Among Russian, Somali and Kurdish Origin Migrants in Finland? *A Cross-sectional Study BMJ Open* (2018) 8:e019166. doi: 10.1136/bmjopen-2017-019166
19. Inker LA, Schmid CH, Tighiouart H, Eckfeldt JH, Feldman HI, Greene T, et al. Estimating Glomerular Filtration Rate From Serum Creatinine and Cystatin C. *N Engl J Med* (2012) 367:20–9. doi: 10.1056/NEJMoa1114248
20. Coorperative Meta-Analysis Group Of China Obesity Task Force. Predictive Values of Body Mass Index and Waist Circumference to Risk Factors of Related Diseases in Chinese Adult Population. *Chinese J Epidemiol* (2002) 23(1):5–10.
21. Xu Y, Wang L, He J, Bi Y, Li M, Wang T, et al. Prevalence and Control of Diabetes in Chinese Adults. *JAMA* (2013) 310:948–59. doi: 10.1001/jama.2013.168118
22. *World Health Organization Waist Circumference and Waist-Hip Ratio: Report of a WHO Expert Consultation*. Geneva, Switzerland: WHO (2008).
23. Strain WD, Paldánius PM. Diabetes, Cardiovascular Disease and the Microcirculation. *Cardiovasc Diabetol* (2018) 17:57. doi: 10.1186/s12933-018-0703-2
24. Koch E, Romero T, Romero CX, Aguilera H, Paredes M, Vargas M, et al. Early Life and Adult Socioeconomic Influences on Mortality Risk: Preliminary Report of a 'Pauper Rich' Paradox in a Chilean Adult Cohort. *Ann Epidemiol* (2010) 20:487–92. doi: 10.1016/j.annepidem.2010.03.009
25. McQuaid SE, Hodson L, Neville MJ, Dennis AL, Cheeseman J, Humphreys SM, et al. Downregulation of Adipose Tissue Fatty Acid Trafficking in Obesity: A Driver for Ectopic Fat Deposition? *Diabetes* (2011) 60:47–55. doi: 10.2337/db10-0867
26. Neeland IJ, Turer AT, Ayers CR, Powell-Wiley TM, Vega GL, Farzaneh-Far R, et al. Dysfunctional Adiposity and the Risk of Prediabetes and Type 2 Diabetes in Obese Adults. *JAMA* (2012) 308:1150–9. doi: 10.1001/2012.jama.11132
27. Hermsdorff HH, Monteiro JB. [Visceral, Subcutaneous or Intramuscular Fat: Where Is the Problem]?. *Arq Bras Endocrinol Metabol* (2004) 48:803–11. doi: 10.1590/s0004-27302004000600005
28. Srikanthan P, Karlamangla AS. Muscle Mass Index as a Predictor of Longevity in Older Adults. *Am J Med* (2014) 127:547–53. doi: 10.1016/j.amjmed.2014.02.007

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers in Endocrinology

SYSTEMATIC REVIEW
published: 21 October 2021
doi: 10.3389/fendo.2021.694681

Tri-Ponderal Mass Index as a Screening Tool for Identifying Body Fat and Cardiovascular Risk Factors in Children and Adolescents: A Systematic Review

Jiahong Sun[1], *Rong Yang*[1], *Min Zhao*[2], *Pascal Bovet*[3] *and Bo Xi*[1]*

[1] Department of Epidemiology, School of Public Health, Cheeloo College of Medicine, Shandong University, Jinan, China, [2] Department of Nutrition and Food Hygiene, School of Public Health, Cheeloo College of Medicine, Shandong University, Jinan, China, [3] Center for Primary Care and Public Health (Unisanté), University of Lausanne, Lausanne, Switzerland

Because of the limitation of body mass index (BMI) in distinguishing adipose mass from muscle, the tri-ponderal mass index (TMI) has been proposed as a new indicator for better assessing adiposity in children and adolescents. However, it remains unclear whether TMI performs better than BMI or other adiposity indices in predicting obesity status in childhood and obesity-related cardiovascular risk factors (CVRFs) in childhood or adulthood. We searched PubMed, Cochrane Library, and Web of Science for eligible publications until June 15, 2021. A total of 32 eligible studies were included in this systematic review. We found that TMI had a similar or better ability to predict body fat among children and adolescents than BMI. However, most of the included studies suggested that TMI was similar to BMI in identifying metabolic syndrome although TMI was suggested to be a useful tool when used in combination with other indicators (e.g., BMI and waist circumference). In addition, limited evidence showed that TMI did not perform better than BMI for identifying specific CVRFs, including insulin resistance, high blood pressure, dyslipidemia, and inflammation in children and adolescents, as well as CVRFs in adults.

Systematic Review Registration: https://www.crd.york.ac.uk/prospero, CRD42021260356.

Keywords: children, tri-ponderal mass index, obesity, body fat, cardiovascular risk factors

OPEN ACCESS

Edited by:
Mostafa Qorbani, Alborz University of Medical Sciences, Iran

Reviewed by:
Motahar Heidari-Beni, Isfahan University of Medical Sciences, Iran
Shirin Djalalinia, Ministry of Health and Medical Education, Iran

***Correspondence:**
Bo Xi
xibo2007@126.com

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *13 April 2021*
Accepted: *28 September 2021*
Published: *21 October 2021*

Citation:
Sun J, Yang R, Zhao M, Bovet P and Xi B (2021) Tri-Ponderal Mass Index as a Screening Tool for Identifying Body Fat and Cardiovascular Risk Factors in Children and Adolescents: A Systematic Review. Front. Endocrinol. 12:694681. doi: 10.3389/fendo.2021.694681

INTRODUCTION

The age-standardized prevalence of obesity among children and adolescents aged 5 to 19 years has globally increased from 0.7% in 1975 to 5.6% in 2016 among girls and 0.9% to 7.8% among boys (1). Obesity in children is a cause of several detrimental health outcomes in childhood and later in adulthood, such as left ventricular hypertrophy (2), increased carotid intima-media thickness (3), kidney disease (4), and liver disease (5), cancer, cardiovascular diseases, and death (6–8). Thus, an early and accurate diagnosis of obesity in children and adolescents is urgently needed, in order to reduce the short-term and long-term burden of pediatric obesity-related health outcomes.

Body mass index (BMI, kg/m^2) is the most widely used physical indicator of adiposity in both children (with overweight/obesity cutoffs based on age and sex percentiles) and among adults (overweight: BMI 25–29; obesity BMI ≥ 30). Although BMI is strongly correlated with adiposity, the indicator cannot distinguish well between excess weight due to increased fat mass or increased muscle mass (9), especially for changes in body composition during adolescence, leading to weight increase being out of proportion of the change in height squared (10, 11). Although the percentage of body fat is suggested as an accurate method for identifying obesity in children and adolescents, it is less applicable for routine health care, as well as in school-based settings (12).

Tri-ponderal mass index (TMI), calculated as weight (kg)/height (m^3), is an emerging indicator, which has been suggested to predict percent body fat (10) and metabolic syndrome (MetS) (13) at least as well as, or better than BMI. However, findings in other previous studies were inconsistent (14–17). For instance, the prevalence of overweight and obesity was higher when identified with BMI (based on standard deviation score, SDS) than with TMI in children and adolescents aged 6–17 years (14). It was also found that BMI (or BMI z-score or BMI-SDS) predicted MetS better than TMI among adolescents aged 10–17 years (15, 16).

It is however unclear whether the emerging TMI can better identify adiposity in childhood or adolescence than the commonly used BMI (10, 14, 18–26) and better predict obesity-related cardiovascular risk factors (CVRFs) such as high blood pressure, dyslipidemia, insulin resistance, and the MetS in childhood (13–16, 18, 21, 26–34) or adulthood (35–37). The misclassification of obesity may lead to either omissions of children who are at high risk of obesity-related diseases or excessive anxiety due to overdiagnosis and then unnecessary waste of medical resources (38). Identifying potential adiposity-related indicators that can accurately predict body fat or related risks has significant implications for prevention, treatment, and management of pediatric obesity.

Therefore, in order to assess whether TMI can be a substitute for BMI in routine pediatric clinical practice to estimate obesity and related CVRFs in children and adolescents or adults, we reviewed articles on the ability of TMI to identify increased body fat, in children and adolescents, and to predict CVRFs in both childhood and adulthood.

METHODS

Search Strategy

This review was performed according to the recommendation from the Preferred Reporting Items for Systematic Reviews and Meta-Analyses statement (PRISMA) (http://www.prisma-statement.org/). We searched relevant articles in PubMed, Cochrane Library, and Web of Science until June 15, 2021 using the following search strategy: ("Triponderal mass index" OR "Tri-ponderal mass index" OR "Tri-ponderal index") AND ("children" OR "childhood" OR "adolescents" OR "adolescence" OR "teenagers" OR "youth" OR "students") AND ("body mass index" OR "obesity" OR "body fat" OR "cardiovascular disease risk" OR "hypertension" OR "dyslipidemia" OR "insulin resistance" OR "impaired glucose" OR "metabolic syndrome" OR "MetS" OR "inflammation"). We also identified eligible papers from the lists of references in the identified papers. We have registered on PROSPERO (available at: https://www.crd.york.ac.uk/prospero/#aboutpage), and the ID is CRD42021260356.

Inclusion Criteria and Exclusion Criteria

Inclusion criteria were as follows: 1) original article; 2) body fat or adiposity assessed using TMI and BMI in childhood or adolescence; 3) the paper described the association of TMI and BMI measured in childhood and adult with selected CVRFs [i.e., hypertension; dyslipidemia; insulin resistance (IR) or impaired glucose; MetS; and inflammation] measured either in childhood (e.g., at the same time of measurement of the BMI/TMI, e.g., in cross-sectional surveys) or in adulthood (e.g., cohort studies) or both; and 4) cross-sectional, cohort, or retrospective studies. Exclusion criteria were as follows: 1) obviously irrelevant articles; 2) TMI measured in adulthood; 3) other languages rather than English; 4) letter or comment; and 5) studies without data of interest.

Identification of Relevant Studies and Data Extraction

Two independent authors (JS and RY) performed the literature search and extracted the data. In case of disagreement between the two authors, a third expert (BX) was consulted to reach an agreement. The information on the first author, publication year, country of origin, study design, sample size, age and sex distribution of the study population, exposures, outcome definition, adjusted covariates, and results was extracted from each eligible study.

Study Quality Assessment

An 11-item checklist of the cross-sectional study evaluation scale recommended by the Agency for Healthcare Research and Quality (AHRQ) was used to evaluate the quality of cross-sectional studies (39), with answers coded as "Yes" (1) or "No or not clear" (0). The total score of the scale is 11 points. A score of 8 to 11 points is rated as high quality, 4 to 7 points as moderate quality, and less than 4 points as low quality. The Newcastle–Ottawa Scale (NOS) star system (range 0 to 9 stars) was used to evaluate the quality of cohort and case–control studies (40). In brief, four items related to the selection of participants, two items to the comparability of participants, and three items to the exposures or outcomes. A score of "0–3" was regarded as low quality, "4–6" as moderate quality, and "7–9" as high quality.

RESULTS

Study Selection

A total of 76 articles were initially identified. After excluding 28 duplicate articles, 48 remained for screening. After excluding 11 irrelevant articles, 2 letters/editorials, 1 in adults, 1 with

overlapping data, 1 in Spanish, and 3 with no data of interest, 28 relevant studies were included. In addition, 4 additional articles were identified from the lists of references, resulting in a total of 32 articles eligible for the final systematic review. The detailed PRISMA flowchart of inclusion/exclusion of potential publications is presented in **Figure 1**.

Study Characteristics

Table 1 describes the characteristics of the included studies including 14 for the association between TMI and body fat in childhood and adolescence (10, 14, 18–26, 41–43), 20 for TMI and CVRFs in childhood and adolescence (13–18, 21, 26–34, 42–45), and 4 for the association in adulthood (35–37, 46). Twenty-five of the included studies used a cross-sectional design (10, 13, 15–28, 30–34, 41–43, 45), 2 a retrospective design (14, 44), 1 a case-control design (29), and 4 a cohort design (35–37, 46).

Results

As shown in **Table 2**, All of the 32 included studies were of moderate to high quality except for one article rated as having a low quality (quality score = 3) (14).

TMI for Screening Body Fat in Children and Adolescents

A total of 14 articles evaluated the ability of TMI to identify body fat mass in children and adolescents compared with BMI (**Table 2**) (10, 14, 18–26, 41–43).

It has been shown that percent of body fat (BF%) as a gold standard was better predicted by TMI than by BMI (10, 25), although one study reported that both relative fat mass pediatric (RFMp) based on height and waist circumference [WC], and waist-to-height ratio (WHtR) performed better than both TMI and BMI (25). When WHtR was used to define central obesity as the gold standard, three articles showed that TMI was better than BMI correlated with central fat accumulation in both preschool-aged children aged 2–5 years (24) and children and adolescents aged 5–17 years (19, 43).

When BMI and TMI were used as continuous variables, TMI correlated similarly or better than BMI with BF% in children and adolescents (18, 19, 22, 23, 41, 42). Although TMI and BMI among children and adolescents aged 5–18 years explained a similar proportion of the variability for BF%, TMI was recommended to replace BMI z-score in children and adolescents due to its lower false-positive rate of obesity than the BMI z-score (boys: 2.9% *vs.* 21.8%; girls: 17.5% *vs.* 28.5%) (18, 19, 22). TMI presented a higher area under the curve (AUC) value than BMI for predicting high BF% (0.96 *vs.* 0.93, $p < 0.001$) measured by dual-energy X-ray absorptiometry (DEXA) among children and adolescents aged 8–17 years (23) or more strongly correlated with BF% compared to BMI in adolescents (41, 42).

The remaining four studies could not conclude about a possible advantage of either TMI or BMI to identify overweight or obesity status because of the lack of a gold standard (to objectively assess adiposity) and inconsistent cutoffs (14, 20, 21, 26). Akcan et al. found that TMI identified a lower prevalence of overweight and obesity among children aged 6–17 years compared to BMI-SDS (14), which was contrary to the finding among children aged 9–13 years, independent of sex (20). When considering overweight and

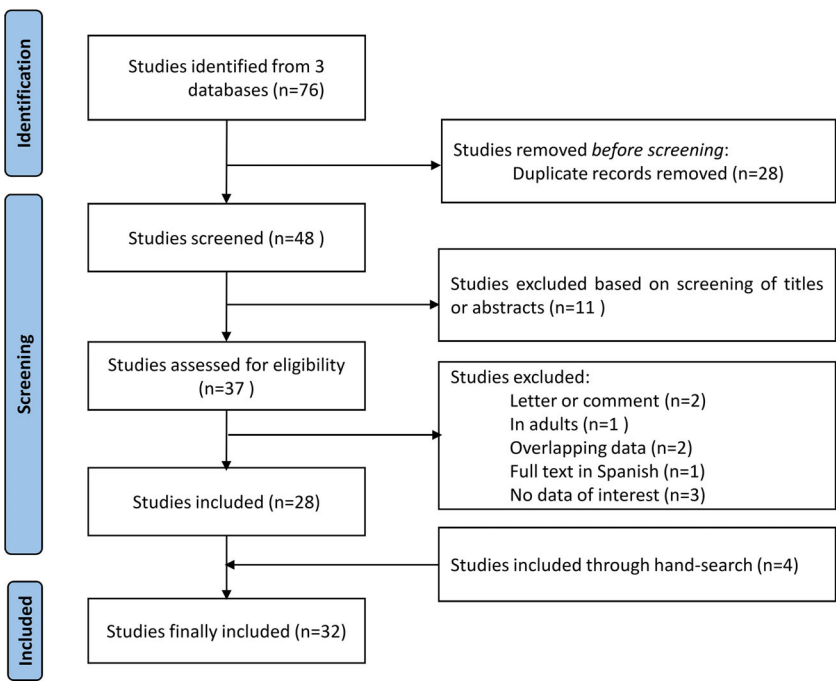


FIGURE 1 | PRISMA flowchart of study selection.

TABLE 1 | Study characteristics of the included studies.

Outcome	First author,year	Study name	Country of origin and design	Study design	Age, years	Sample size	Sex: (male, %)	Exposures	Outcome definition
Body fat									
	Peterson, 2017 (10)	The 1990–2006 US National Health and Nutrition Examination Survey	USA	Cross-sectional	8–17	2,285	55.1	TMI *vs*. BMI	Continuous: BF% by DXA Categorical: Overweight was based on the 85th–95th percentiles of TMI, BMI, and BF%
	Jiang, 2018 (18)	A multicentre cross-sectional study in east and southwest China	China	Cross-sectional	7–18	1,860	49.7	TMI *vs*. WHtR, BMI SDS, WC SDS	Continuous: BF% by DXA
	Sims, 2018 (19)	The Canadian Study of Determinants of Endometabolic Health in China	Canada	Cross-sectional	5–17	181: 44 SCBT and 137 non-cancer control children	53.6	TMI *vs*. BMI	Continuous BF% by bioelectrical impedance, WHtR, WHpR
	Akcan, 2019 (14)	Study from the Pediatric Endocrinology Outpatient Clinics of the Near East University	Cyprus	Retrospective	6.3–17.6	143	42	TMI *vs*. BMI	Overweight: BMI-SDS +1.0 to +2.0; TMI 16.8 kg/m^3 for girls and 16.0 kg/m^3 for boys obesity: BMI-SDS ≥+2.0; TMI 19.7 kg/m^3 for girls and 18.8 kg/m^3 for boys
	Moselakgomo,2019 (20)	Study from the Limpopo and Mpumalanga province of South Africa	South African	Cross-sectional	9–13	1,361	49.8	TMI *vs*. BMI	Overweight and obesity were based on age- and sex-specific TMI and BMI percentages of the study population
	Ashley-Martin, 2019 (21)	Canadian Health Measures Survey	Canada	Cross-sectional	6–19	5,814	50.7	TMI *vs*. BMI	Overweight and obesity: based on BMI z-score of the International Obesity Task Force and age- and sex-specific 85th and 95th TMI percentiles of the National Health and Nutrition Examination Survey.
	Zaniqueli, 2019 (22)	Study from the municipality of Serra and Vitória, State of Espírito Santo, Brazil	Brazil	Cross-sectional	6–18	1,149	53.2	TMI *vs*. BMI	BF% was by bioelectrical impedance. Obesity: respectively based on the 95th percentile of TMI, BMI, and BF%
	De Lorenzo, 2019 (23)	Study from the University of Rome Tor Vergata, Human Nutrition Unit, Italy	Italy	Cross-sectional	8–17	485	42.7	TMI *vs*. BMI	BF% by DXA High adiposity: ≥75^{th} percentile of BF%
	Nascimento, 2019 (24)	Study from Taubaté, São Paulo, Brazil	Brazil	Cross-sectional	2–5	919	50.1	TMI *vs*. BMI	WHtR was used to define central fat accumulation: the upper tertile of the study population
	Woolcott, 2019 (25)	The National Health and Nutrition Examination Survey from 1999 to 2006	USA	Cross-sectional	8–19	10,390	56.8	TMI *vs*. RFMp (RFM), BMI, WHtR	BF% by DXA Overweight and obesity diagnoses using BMI, TMI, RFMp, RFM, and WHtR were defined based on 85th and 95th percentiles, respectively (BMI

(Continued)

TABLE 1 | Continued

Outcome	First author,year	Study name	Country of origin and design	Study design	Age, years	Sample size	Sex: (male, %)	Exposures	Outcome definition
									specific for sex and age, the others specific for sex).
	Park, 2020 (26)	Korea National Health and Nutrition Examination Survey, 2007–2016	Korea	Cross-sectional	10–20	9,749	51.5	TMI *vs*. BMI	Overweight: BMI or TMI was ≥85th percentile and <95th percentile Obesity: BMI or TMI was ≥95th percentile
	Ye, 2020 (41)	Data from the Qibao Community in Minhang District of Shanghai	China	Cross-sectional	6–17	14,042	54.3	TMI *vs*. BMI, WHtR, WHR, WC, body adiposity index	BF% measured using bioelectrical impedance analysis (boys aged 6–18 years: ≥20%; girls aged 6–14 years: ≥25%; girls aged 15–18 years: ≥30%)
	Alfaraidi 2021 (42)	The Improving Renal Complications in Adolescents with Type 2 Diabetes Through Research cohort Study	Canada	Cross-sectional	10.2–17.9	116	31.0	TMI *vs*. BMI z-score	FM% and WHtR
	Malavazos 2021 (43)	The Italian "Educazione A limentare Teenagers" project survey	Italy	Cross-sectional	12–13	3479	54.3	TMI *vs*. BMI or BMI z-score	Central obesity was defined as WHtR ≥0.5
Obesity-related cardiovascular risk factors									
	Ramírez-Vélez, 2018 (27)	The Fuprecol Study in Bogotá, Colombia	Columbia	Cross-sectional	9–25	4673	42.9	TMI *vs*. FMI	MetS was defined as 3 or more of following criteria (1): abdominal obesity: WC ≥90 cm for men and 80 cm for women; (2) hypertriglyceridemia: ≥150 g/dl; (3) low HDL-C: <40 mg/dl for men and <50 mg/dl for women; (4) high BP: ≥130/85 mmHg; (5) high fasting glucose: ≥100 mg/dl.
	Gomes, 2018 (15)	Study from the North and Central regions of mainland Portugal	Portugal	Cross-sectional	10–17	1,324	47.1	TMI *vs*. BMI, BMI z-score, WC, WC/H, and WC/Hadj.	A standardized metabolic risk score was computed by summing of standardized values for fasting glucose, triglycerides, high-density lipoprotein cholesterol, and mean arterial BP.
	Jiang, 2018 (18)	A multicentre cross-sectional study in east and southwest China	China	Cross-sectional	7–18	1,860	49.7	TMI *vs*. WHtR, BMI SDS, WC SDS, and BF%	CMR1 and CMR2 were defined as 3 or more and 2 or more following abnormalities: (1) Hypertension: based on age- and sex-specific reference of Chinese children and adolescents, (2) Dyslipidemia: TG ≥ 1.76 mmol/l or TC ≥ 5.2 mmol/l or LDL-C ≥ 3.38 mmol/l, or HDL-C ≤ 1.04 mmol/l, (3) Elevated fasting blood glucose ≥ 5.6 mmol/l, (4) Central obesity:

(Continued)

TABLE 1 | Continued

Outcome	First author,year	Study name	Country of origin and design	Study design	Age, years	Sample size	Sex: (male, %)	Exposures	Outcome definition
									recommended by the China children's obesity working group.
	Ashley-Martin, 2019 (21)	The Canadian Health Measures Survey	Canada	Cross-sectional	6–19	5,814	50.7	TMI *vs*. BMI	High TC: ≥ 200 mg/dl, low HDL-C: <40 mg/dl, TG ≥ 100 mg/dl for 0–9 years and ≥130 mg/dl for 10–19 years, C-reactive protein: >3.0 mg/l, HOMA-IR: ≥90th percentile, and high BP: SBP and/or DBP ≥90th percentile.
	Shim, 2019 (28)	Korea National Health and Nutrition Examination Survey, 2007–2016.	Korea	Cross-sectional	10–20	8,464	51.6	TMI	MetS was defined as 1 or more of the following criteria: (1) elevated WC: ≥90th percentile, (2) elevated BP: ≥90th percentile, (3) elevated glucose: ≥110 mg/dl, (4) elevated TGs:≥110 mg/dl, and (5) reduced HDL-C: <40 mg/dl.
	Akcan, 2019 (14)	Study from the Pediatric Endocrinology Outpatient Clinics of the Near East University	Cyprus	Retrospective	6.3–17.6	143	42.0	TMI *vs*. BMI	IR: Prepubertal girls: 2.22; prepubertal boys: 2.67; pubertal girls: 3.82; and pubertal boys: 5.22 High liver enzymes: The threshold for serum glutamic oxaloacetic transaminase: 5–34 U/l, serum glutamic pyruvic transaminase: 0–55 U/l Dyslipidemia: TC ≥ 200 mg/dl; TG ≥ 150 mg/dl; HDL < 40 mg/dl; and LDL ≥ 100 mg/dl
	Arsang-Jang, 2019 (30)	The Adolescence Surveillance and Prevention of Adult Non-communicable disease survey	Iran	Cross-sectional study	7–18	24,409	50.1	TMI *vs*. BMI, TBSI, WC, WH.5R, WHtR	MetS: abdominal obesity plus at least 2 of the following risk factors: (1) high TG ≥ 150 mg/dl; low HDL-C: males, <40 mg/dl and females, <50 mg/dl; high BP, SBP/DBP ≥ 130/85 mm Hg; high FPG: ≥100 mg/dl or previously diagnosed as T2DM
	Radetti, 2019 (16)	Study from the obesity inpatient clinic of the Istituto Auxologico Italiano, Piancavallo, Verbania, Italy	Italy	Cross-sectional	10–17	1,332	41.6	TMI *vs*. BMI, BMI SDS, FFMI, FMI, WHtR, BMFI	Mets: abdominal obesity plus at least 2 of the following risk factors: (1) high TG ≥ 150 mg/dl; low HDL-C: males, <40 mg/dl and females, <50 mg/dl; high BP, SBP/DBP ≥ 130/85 mm Hg; high FPG: ≥100 mg/dl or previously diagnosed as T2DM
	Umano, 2019 (31)	Obesity outpatient clinic in Italy	Italy	Cross-sectional	4–18	1,387	51.4	TMI *vs*. BMI z-score, WC z-score and WHtR	BP, glucose, insulin, and lipid profile
	Wang, 2020 (13)	A Chinese National School-based Health Survey and United States National Health and Nutrition	China and the USA	Cross-sectional	7–18 for Chinese; 12–18 for American	57,201 Chinese children and 10,441 American children	51.6 for Chinese; 50.9 for American	TMI *vs*. BMI, BMI z-score, weight/height$^{2.5}$	Impaired FPG:≥5.6 mmol/l; dyslipidemia: TC ≥ 170 mg/dl; high LDL-C: ≥110 mg/dl; low HDL-C: <120 mg/dl; TG ≥ 75 mg/dl for children under 9 years and ≥90 mg/dl for children more than 10 years; HBP: BP ≥ 90th percentile

(Continued)

TABLE 1 | Continued

Outcome	First author,year	Study name	Country of origin and design	Study design	Age, years	Sample size	Sex: (male, %)	Exposures	Outcome definition
		Examination Survey							
	Park, 2020 (26)	Korea National Health and Nutrition Examination Survey, 2007–2016.	Korea	Cross-sectional	10–20	9,749	51.5	TMI *vs*. BMI	DBP, SBP, HDL-C, LDL-C, TC, TG, WC
	Akcan, 2020 (29)	Study from the Pediatric Endocrinology Outpatient Clinics of the Near East University	Cyprus	Case–control study	5.3–17.4	80	42.5	TMI *vs*. BMI	IR: prepubertal girls: 2.22; prepubertal boys: 2.67; pubertal girls: 3.82; and pubertal boys: 5.22; Low HDL-C: <40 mg/dl; High TG: >150 mg/dl
	Matsuo, 2020 (32)	Study on the effectiveness of multidisciplinary obesity treatment program in Brazil	Brazil	Cross-sectional	12–18	217	38.7	TMI *vs*. BMI, WC, WHtR	HOMA-IR: cutoff point of ≤3.16
	Khoshhali, 2020 (33)	The fifth survey of "Childhood and Adolescence Surveillance and Prevention of Adult Non-communicable Disease"	Iran	Cross-sectional	7–18	3731	52.6	TMI *vs*. BMI	MetS was defined as 3 or more of following criteria: (1) abdominal obesity: WHtR ≥0.5, (2) elevated FBG: ≥100 mg/dl, (4) high TG: ≥100 mg/dl, (5) low HDL-C: <40 mg/dl, (6) elevated BP: ≥ age-, sex-, and height-specific 90th percentile
	Neves, 2020 (34)	Study from the Vitória, Espírito Santo, Brazil	Brazil	Cross-sectional	8–14	296	45.6	TMI *vs*. BMI z-score	HOMA-IR: based on β-cell function (%) = 20*insulin/(glucose-3.5); resistance = insulin/($22.5e^{-\ln glucose}$)
	Leone, 2020 (17)	International Center for the Assessment of Nutritional Status	Italy	Cross-sectional	7–20	403	44.4	TMI *vs*. BMI z-score, WHtR, body shape index z-score, and conicity index	MetS: 7–10 years (three or more of the following criteria: WC ≥ 90th percentile; systolic or diastolic BP ≥ 90th percentile; TG ≥ 90th percentile or HDL ≤ 10th percentile; HOMA-IR ≥ 90th percentile or FPG ≥ 90th percentile; 10–20 years: IDF criteria
	Umano, 2020 (44)	A study from an obesity outpatient clinic of the Department of Pediatrics of the University of Campania Luigi Vanvitelli of Naples	Italy	Retrospective study	10.5 ± 2.89	1,900	50.2	TMI *vs*. BMI z-score and WHR	Non-alcoholic fatty liver disease was assessed based on high-level and abnormally intense echoes from the liver kidney and hepatic parenchyma in echo amplitude
	Alfaraidi, 2021 (42)	Improving Renal Complications in Adolescents with Type 2	Canada	Cross-sectional	10.2–17.9	116	31.0	TMI *vs*. BMI z-score	HDL

(Continued)

TABLE 1 | Continued

Outcome	First author,year	Study name	Country of origin and design	Study design	Age, years	Sample size	Sex: (male, %)	Exposures	Outcome definition
		diabetes Through Research cohort study							
	Calcaterra 2021 (45)	Outpatient clinics in Milan	Italy	Cross-sectional	6–18	585	47.7	TMI *vs*. BMI or BMI z-score	HOMA-IR; HOMA-β; quantitative insulin sensitivity check index; triglyceride and glucose index
	Malavazos 2021 (43)	The Italian "Educazione A limentare Teenagers" project survey	Italy	Cross-sectional	12–13	3,479	54.3	TMI *vs*. BMI or BMI z-score	BP ≥ age-, sex-, and height-specific 90th percentile of the NHBPEP Working Group
Adult health conditions									
	Wu, 2018 (1) (36)	The Childhood Determinants of Adult Health Study	Australia	Cohort	7–15 at baseline	2,345	49.1	TMI *vs*. WC, WC adjusted for height, weight adjusted for height, HC, waist–hip ratio, WHtR, BMI, conicity index, AVI, body adiposity index, and a body shape index.	HOMA2-β: beta-cell function and fasting insulin ≥75th percentile; HOMA-IR: HOMA index ≥75th percentile; High fasting insulin:≥ 5.6 mmol/l
	Wu, 2018(2) (35)	The Cardiovascular Risk in Young Finns Study	Finland	Cohort	3–18 at baseline	2,626	–	TMI and its combination with BMI or SST *vs*. BMI	T2D: FPG ≥ 126 mg/dl or hemoglobin A1c ≥6.5%, or used glucose-lowing medication; obesity: BMI ≥ 30 kg/m^2; Hypertension: SBP and/or DBP ≥ 140/90 mmHg, abnormal LDL-C: ≥160 mg/dl, HDL-C:<40 mg/dl, and high carotid intima-media thickness: ≥90th percentiles
	Wu, 2020 (37)	The ongoing Special Turku Coronary Risk Factor Intervention Project	Finland	Cohort	2–20	432	48.1	TMI *vs*. BMI	Aortic intima-media thickness, IFG, elevated insulin levels, HOMA-IR, serum lipids, and hypertension
	Wu, 2021 (46)	Taipei City Hospital Radiation Building Database	Taiwan (China)	Cohort	13–18	1,387	49.7	TMI *vs*. BMI-z score	Diabetes: FPG ≥ 126 mg/dl or diagnosed by physicians or current use of diabetes medicine

AUC, area under the curve; AVI, abdominal volume index; BMI, body mass index; BP, blood pressure; BMFI, body mass fat index; CMR, cardiometabolic risk; FPG, raised fasting plasma glucose; FMI, fat mass index; FFMI, fat-free mass index; FMI, fat mass index; HC, hip circumference; HOMA-IR, homeostasis model assessment-insulin resistance; HOMA2-β, homeostasis model assessment of beta-cell function; HOMA2-IR, homeostasis model assessment of insulin resistance; HDL-C, high-density lipoprotein cholesterol; IFG, impaired fasting glucose; IR insulin resistance; LDL-C, low-density lipoprotein cholesterol; MetS, metabolic syndrome; RFMp: relative fat mass pediatric; SCBT, survivors of childhood brain tumors; SD, standard deviation; TMI, tri-ponderal mass index; TC, total cholesterol; T2D, type 2 diabetes; TBSI, tri-ponderal body shape index; TG, triglycerides; WC, waist circumference; WC/H, WC/height ratio; WC/Hadj, WC/H adjusted ratio; WH.5R, WC to height 5; WHtR, waist-to-height ratio; WHR, waist to hip ratio; FM%, percent of fat mass; SGOT, serum glutamic oxaloacetic transaminase; SGPT, serum glutamic-pyruvic transaminase; SST, subscapular skinfold thickness.

obesity separately, Ashley-Martin et al. found that BMI defined more overweight than TMI, whereas TMI defined more obesity than BMI among children and adolescents aged 6–19 years (21), inversely to the findings among children and adolescents aged 10–20 years reported by Park et al. (26).

Overall, studies using a gold standard for comparison and using BMI and TMI as continuous variables suggested that TMI performed equally or better than the widely used BMI to predict BF% and central fat among children and adolescents. TMI was preferred in adolescence due to its stability.

TABLE 2 | Results of the included studies.

Outcome	First author, year	Results	Adjusted covariates	Study quality
Body fat				
	Peterson, 2017 (10)	(1) For children and adolescents aged 8 to 17 years, TMI was better to estimate BF% than BMI, especially in boys (boys: R^2 = 0.64 *vs*. 0.38; girls: R^2 = 0.72 *vs*. 0.66). (2) The misclassification of overweight was less than BMI z-score (8.4% *vs*. 19.4%, p < 0.001) but equal to updated BMI percentiles based on the same data set (8.4% *vs*. 8.0%, p = 0.62). However, TMI was preferred due to its simplicity with no complicated percentiles. (3) The results were similar when stratified by sex.	None	6
	Jiang, 2018 (18)	(1) WHtR was most strongly correlated with BF% (*rho* coefficient =0.73, $p < 0.001$), followed by WC SDS, TMI, and BMI SDS (*rho* =0.71 *vs*. 0.68 *vs*. 0.68, $p < 0.001$). (2) TMI and WHtR were more applicable for public health use than BMI, WC, and BF% due to their simplicity in calculating and identifying obesity. The AUCs of these indicators remained similar when stratified by sex.	None	6
	Sims, 2018 (19)	After adjusting for potential variables, the correlation between TMI and BF% was equal to BMI z-score (*r*= 0.85 *vs*. 0.85), whereas the correlation between TMI and WHpR (*r* = 0.46 *vs*. 0.41) or WHtR (*r*= 0.86 *vs*. 0.78) was stronger than BMI z-score.	Age, sex, treatment, and puberty	6
	Akcan, 2019 (14)	TMI revealed less overweight and obesity than BMI. About 22 overweight children and 8 obese children identified by BMI-SDS were regarded as normal-weight children identified by TMI. 44 obese children (based on BMI) were overweight according to TMI.	None	3*
	Moselakgomo, 2019 (20)	TMI revealed more overweight and obesity than BMI (overweight: 5.66% *vs*. 1.84%; obesity: 1.98% *vs*. 0.66%). The classification of overweight and obesity by TMI and BMI were as follows: overweight: boys: 7.3% *vs*. 2.6%; 2.2% *vs*. 0.7%; girls: 4.0% *vs*. 1.0%; obesity: boys: 2.2% *vs*. 0.7%; girls: 1.8% *vs*. 0.6%.	None	5
	Ashley-Martin, 2019 (21)	The prevalence of overweight defined by TMI was lower than that defined by BMI (15% *vs*. 18%), but the prevalence of obesity defined by TMI was higher than that defined by BMI (9.7% *vs*. 8.9%)	None	4
	Zaniqueli, 2019 (22)	Although TMI (R^2 = 0.73 for boys and R^2 = 0.75 for girls) and BMI (R^2 = 0.74 for boys and R^2 = 0.75 for girls) performed similar in the portion of the variability for BF%, TMI was recommended to replace the BMI z-score in children and adolescents due to a lower false-positive rate of obesity (boys: 21.8% *vs*. 3.9%; girls: 28.5% *vs*. 17.5%).	None	6
	De Lorenzo, 2019 (23)	TMI was a better predictor for BF% in both sexes than BMI (boys: R^2 = 0.67 *vs*. 0.44; girls: R^2 = 0.79 *vs*. 0.74). TMI presents a higher AUC value than BMI for predicting high adiposity in children and adolescents (0.96 *vs*. 0.93).	None	5
	Nascimento, 2019 (24)	The AUC of TMI was higher than BMI for screening central fat accumulation (0.92 *vs*. 0.87), regardless of sex.	None	6
	Woolcott, 2019 (25)	(1) RFMp and WHtR showed similar linear association with BF%, followed by TMI and BMI in children and adolescents 8 to 14 years (R^2 = 0.77, 0.76, 0.69, 0.55 for boys, R^2 = 0.74, 0.74, 0.71, 0.65 for girls). (2) Similar results in boys aged 15 to 19 years. WHtR (R^2 = 0.80 for boys and 0.70 for girls) showed higher predicting ability than RFM (0.79 for boys and 0.72 for girls) among boys, followed by BMI and TMI in children and adolescents aged 15-19 years (R^2 = 0.70 and 0.69 for boys and 0.73 and 0.72 for girls. However, the predicting ability was similar among girls. (3) RFMp for children and adolescents 8 to 14 years of age and RFM for adolescents 15 to 19 years of age were useful to estimate whole-body fat percentage and diagnose body fat-defined overweight or obesity.	None	6
	Park, 2020 (26)	The prevalence of overweight defined by TMI was slightly higher than that defined by BMI (10.6% *vs*. 10.2%), but the prevalence of obesity defined by TMI was lower than that defined by BMI (5.3% *vs*. 10.6%), similar in both sex.	None	5
	Ye, 2020 (41)	The correlation between BMI and BF% (r = 0.919) was higher than TMI (r = 0.896), WC (r = 0.842), WHtR (r = 0.830), and WHR (r = 0.522). For children aged 6–11 years, the AUC values of BMI (0.980 for boys and 0.981 for girls) was significantly higher than TMI (0.957 and 0.948), WC (0.940 and 0.945), and WHtR (0.939 and 0.921) whereas for adolescents aged 12–17 years, TMI (0.976 for males and 0.945 for females) performed better than BMI (0.967 and 0.943), WHtR (0.960 and 0.878), and WC (0.945 and 0.864) to identify obesity	Age and sex	8
	Alfaraidi, 2021 (42)	TMI was associated with FM% (r = 0.74, $p < 0.0001$) and WHtR (r = 0.85, $p < 0.0001$), among adolescents with type 2 diabetes, whereas BMI was not.	Age and sex	8
	Malavazos, 2021 (43)	TMI was better than BMI and BMI z-score to discriminate central fat among adolescents. (AUC in boys: TMI 0.96, BMI, 0.95, $p < 0.001$, BMI z-score 0.95, $p = 0.002$; AUC in girls: TMI 0.97, BMI 0.96, $p < 0.0001$, BMI z-score 0.96, $p < 0.0001$)) The prevalence of central obesity based on TMI (96.6% in boys and 97.3% in girls) was higher	None	7

(Continued)

TABLE 2 | Continued

Outcome	First author, year	Results	Adjusted covariates	Study quality
		than BMI (50.7% in boys and 34.6% in girls) and BMI z-score (52.4% in boys and 38.7% in girls) among adolescents with overweight		
Obesity-related cardiovascular risk factors				
	Ramírez-Vélez, 2018 (27)	The power of TMI to detect MetS was comparable to FMI 9–12 years AUCs for girls: TMI: 0.674; FMI: 0.698. AUCs for boys: TMI: 0.755; FMI: 0.752. 13–17 years AUCs for girls: TMI: 0.684; FMI: 0.699. AUCs for boys: TMI 0.729; FMI: 0.745	None	5
	Gomes, 2018 (15)	BMI z-score (AUC 0.678), BMI (0.683), and WC (0.676) were a stronger predictor for metabolic risk score than TMI (0.655)	None	4
	Jiang, 2018 (18)	TMI showed similar good performance in identifying CMR (AUC of CMR1 and CMR2: 0.88, 95% CI 0.84–0.92; 0.82, 0.79–0.85) to WHtR (0.88, 0.83–0.92; 0.82, 0.79–0.86), BMI SDS (0.89, 0.85–0.93; 0.84, 0.81–0.87), and WC SDS (0.89, 0.85–0.93; 0.84, 0.81–0.87), but higher performance than BF% (0.83, 0.78–0.88; 0.77, 0.74–0.80).	None	6
	Ashley-Martin, 2019 (21)	Similar to BMI, TMI was a good predictor for HOMA-IR or having more than 3 abnormal tests (AUC 0.83 and 0.81), but poor for CRP (0.73 and 0.74), high TG (0.67 and 0.68), low-HDL-C (0.67 and 0.66), high TC (0.60 and 0.62), and high BP (SBP: 0.66 and 0.66; DBP: 0.55 and 0.56).	None	4
	Shim, 2019 (28)	Compared with normal weight, overweight defined by TMI was associated with MetS (OR 25.57) and its components, including low HDL-C (2.31), elevated TG (2.55), elevated BP (1.33), and elevated WC (29.18). The association was stronger for obesity defined by TMI, suggesting TMI might be used as a screening tool for overweight and obesity in a clinical setting.	Age, sex, alcohol consumption, smoking, household income, physical activity, rural residence, hypertension, diabetes mellitus, and dyslipidemia	7
	Akcan, 2019 (14)	IR: Compared to BMI, TMI was more likely to overlook IR. Of 22 overweight children defined by BMI with normal TMI, 22.7% had IR. 2 of 8 obese children (25%) defined by BMI with normal TMI had IR. Among 44 obese children based on BMI but overweight based on TMI and 40.9% were detected as IR. High Liver enzymes: Compared to BMI, TMI was better to predict visceral adiposity High liver enzymes were not found in any of the children with normal TMI. Dyslipidemia: Among overweight children based on BMI but normal with TMI, 9.1% had high TC, 4.5% high TG and low HDL-C, and 50% high LDL	None	3*
	Arsang-Jang, 2019 (30)	Among adolescents, compared with BMI, TMI, WC, WHtR, and WH.5R, the TBSI (WC z-score/($TMI^{2/3}$*$Height^{1/2}$) was considered the best predictor of MetS. The TBSI was significantly more accurate than the BMI and TMI (Youden index: 0.85 *vs*. 0.73 *vs*. 0.70) for classifying individuals with MetS and in healthy groups.	None	5
	Radetti, 2019 (16)	BMFI (BMI*FM% *WC; AUC female, 0.69; male 0.59) performed marginally better than BMI (0.68 and 0.58), TMI (0.66 and 0.55), FMI (0.67 and 0.58), FFMI (0.61 and 0.55), WHtR (0.68 and 0.56), and BMI SDS (0.68 and 0.58) in predicting MetS	None	6
	Umano, 2019 (31)	WHtR performed best in predicting lipid metabolism markers and glucose, followed by the TMI, WC z-score, and BMI z-score among children and adolescents with obesity.	Age, gender, and pubertal stage	7
	Wang, 2020 (13)	(1) TMI was significantly associated with metabolic variables, the ranges of ORs were 1.09 (95% CI 1.04, 1.14) for impaired FPG, 1.13 for dyslipidemia (95% CI 1.11, 1.15), and 1.23 (95% CI 1.22, 1.25) for high BP. Similar results were found among Americans. (2) TMI showed similar values to BMI percentiles but were more precise than BMI z-score to predict cardiovascular risks. However, for specific cardiovascular risks, TMI was similar to BMI to identify IR, better than BMI to identify high BP, and poor as BMI to identify dyslipidemia. (3) The ranges of misclassification rates were 19.1% to 34.7% for TMI and 26.3% to 36.8% for BMI z-score in Chinese, similar for American subjects.	Age and sex	8
	Park, 2020 (26)	(1)Among those with normal BMI, boys with overweight TMI had higher TC (174.4 mg/dl *vs*. 153.6 mg/dl, $p = 0.002$) and TG (101.9 mg/dl *vs*. 77.4 mg/dl, $p = 0.028$), compared with boys with normal TMI; girls with overweight TMI had lower HDL-C (50.1 mg/dl *vs*. 53.5 mg/dl, $p = 0.045$) and higher TG (102.8 mg/dl *vs*. 81.4 mg/dl, $p = 0.029$), compared with girls with normal TMI. (2) Among those with overweight BMI, boys with overweight TMI had higher TC (169.8 mg/dl *vs*. 157.5 mg/dl, $p < 0.001$) and LDL-C (101.7 mg/dl *vs*. 90.8 mg/dl, $p < 0.001$), girls had lower HDL-C (49.5 mg/dl *vs*. 51.9 mg/dl, $p = 0.013$) and TG (96.5 mg/dl *vs*. 82.6 mg/dl, $p = 0.004$), compared with those with normal TMI. (3) The obesity-related comorbidities (except for DBP) of the overweight group (based on TMI) were worse under the same BMI category (normal or overweight).	None	6

(Continued)

TABLE 2 | Continued

Outcome	First author, year	Results	Adjusted covariates	Study quality
	Akcan, 2020 (29)	TMI was associated with a similar amount of metabolic markers to BMI. BMI as a continuous variable seemed to be more strongly associated with TC (R^2: 0.32 *vs*. 0.27), HDL (-0.52 *vs*. -0.46), and TG (0.32 *vs*. 0.27) and TMI was more strongly associated with low-density lipoprotein-cholesterol (LDL-C) (0.38 *vs*. 0.33). Leptin, IL-6, and fetuin-A were more closely correlated with BMI than TMI	None	4*
	Matsuo, 2020 (32)	(1) In overweight adolescents, WC presented the most predictive capacity to explain IR and BMI had a slightly better predictive capacity than TMI, regardless of sex. (2) In boys, TMI and BMI showed similar values of sensibility (88.4% *vs*. 88.2%) and specificity (42.4% *vs*. 45.5%). Nevertheless, BMI had a better sensibility (57.1% *vs*. 49.0%) while TMI had a better specificity (88.1% *vs*. 81.0%) for girls. WC demonstrated a strong sensibility (boys: 82.4%; girls: 79.6%) for both sexes.	None	6
	Khoshhali, 2020 (33)	Among boys, the AUC in identifying MetS of TMI was similar to BMI for both 7–10 years (0.72 *vs*. 0.69), 15–18 years (0.70 *vs*. 0.67), 11–14 years (0.74 *vs*. 0.74), and 7–18 years (0.72 *vs*. 0.69), as well as among girls at age 7–18 years (AUC = 0.68 *vs*. 0.67)	None	5
	Neves, 2020 (34)	TMI showed a similar performance in identifying HOMA-IR to BMI z-score for both sex (boys: TMI = 0.843, BMI z-scores = 0.831; girls: TMI = 0.763, BMI z-scores = 0.756).	None	4
	Leone, 2020 (17)	MetS Children aged <10 years: only BMI z-score was associated with MetS ($\beta = 2.21$, $p < 0.05$) Children aged ≥10 years: BMI z-score ($\beta = 2.67$, $p < 0.001$), TMI ($\beta = 0.19$, $p < 0.01$), conicity index ($\beta = 9.02$, $p < 0.001$) and WHR ($\beta = 12.32$, $p < 0.001$) were associated with MetS. Similar results were found among males, whereas only conicity index ($\beta = 7.37$, $p < 0.05$) and WHR ($\beta = 7.94$, $p < 0.05$) were associated with MetS among females. High BP: BMI z-score was the best predictor of high BP in both children and adolescents, whereas TMI performed better among males. High TG: conicity index was the best predictor for high TG in females and WHR was best in males. Low HDL-C: BMI z-score was the best indicator for low HDL-C.	Age and sex	8
	Umano, 2020 (44)	The AUC of WHR (0.62) was higher than TMI (0.58) and BMI (0.58).	None	7
	Alfaraidi, 2021 (42)	TMI was associated with HDL (r = -0.26, $p < 0.005$) among adolescents with type 2 diabetes, whereas BMI was not.	Age and sex	8
	Calcaterra, 2021 (45)	Among children and adolescents with obesity, TMI was associated with IR indicators only in females while BMI correlated with all IR indicators except for triglyceride and glucose index in females and BMI z score correlated with all IR indicators except for HOMA-β in males.	None	7
	Malavazos 2021 (43)	TMI was better than BMI and BMI z-score to discriminate hypertension. (AUC in boys: TMI 0.73, BMI, 0.70, $p = 0.002$, BMI z-score, 0.70, $p = 0.020$; AUC in girls: TMI 0.76, BMI 0.73, $p = 0.002$, BMI z-score, 0.74, $p = 0.020$)	None	7
Adult health conditions				
	Wu, 2018a (36)	TMI of children was significantly correlated with adult HOMA2-IR (RR 1.15, 95% CI 1.02, 1.29), high HOMA2-β (RR 1.25, 95% CI 1.11, 1.40), and high fasting insulin (RR 1.17, 95% CI 1.04, 1.31). However, the predictive ability was low with AUCs of 0.53, 0.56, and 0.54, respectively, which was lower than other indicators such as abdominal volume index, BMI, and WC.	None	7*
	Wu, 2018b (35)	(1) Youth TMI, BMI, and subscapular skinfold thickness were significantly associated with adult T2D, obesity, high carotid intima-media thickness, and high LDL-C level. (2) Youth TMI was not associated with adult hypertension and low HDL-C (3) Youth BMI was superior or comparable to TMI and SST in predicting adult T2D (AUC 0.688 *vs*. 0.682 *vs*. 0.683), obesity (0.726 *vs*. 0.673 *vs*. 0.683), hypertension (0.660 *vs*. 0.656 *vs*. 0.660), high carotid intima-media thickness (0.568 *vs*. 0.554 *vs*. 0.557), and high LDL-C level (0.609 *vs*. 0.608 *vs*. 0.614).	None	7*
	Wu, 2020 (37)	(1) BMI had stronger associations with insulin (at age 16 years), SBP (age 5–20 years), and TG (age 18 years) than TMI. (2) Between the ages of 14 and 16, BMI outperformed TMI for elevated insulin levels (difference in AUC = 0.018 and 0.025) and IR (difference in AUC = 0.018–0.024). At age 16–20 years, BMI outperformed TMI for hypertension (difference in AUC = 0.017–0.022). For other outcomes of impaired FPG, high aortic intima-media thickness, high LDL-C, low HDL-C, and high TG, the predictive utilities were similar.	None	7*
	Wu, 2021 (46)	Persistent increase of TMI during 13–18 years was associated with increased risk of diabetes in adulthood (hazard ratio: 2.85, 95% confidence interval: 1.01–8.09). No association was found for BMI z score (2.79, 0.35–22.00)	Age, sex, baseline weight status, height, family history of diabetes, smoking, systolic and diastolic BP, TG, and fasting glucose cholesterol	8

**The study quality was assessed by Newcastle-Ottawa Scale and others were assessed by Agency for Healthcare Research and Quality.*

TMI and Obesity-Related Cardiovascular Risk Factors in Children and Adolescents

Twenty articles on the association between TMI and MetS and its components were included in this systematic review (**Table 2**) (13–18, 21, 26–34, 42–45).

MetS

Ten articles have evaluated the association of TMI and other anthropometric indicators with MetS, metabolic risk score, or cardio-metabolic risk (13, 15–18, 27–30, 33). Three of the 10 articles showed that TMI was not better than other indicators such as BMI (or BMI z-score or BMI-SDS) among children and adolescents aged 10–17 years to predict MetS and a metabolic risk score (15–17). However, the other seven articles suggested that TMI could be a useful screening tool or similar to BMI in predicting MetS or cardio-metabolic risks in children and adolescents aged 5.3 to 25 years (13, 18, 27–30, 33).

TMI was found to be associated with obesity-related CVRFs, including MetS and its components [elevated blood pressure (BP), elevated WC, low high-density lipoprotein cholesterol (HDL-C), and elevated triglycerides (TG)] in late adolescence (28). It was reported that TMI performed similarly to FMI (27) or BMI, or was an auxiliary indicator in addition to BMI, to identify MetS, a metabolic risk score, or CVRFs among children and adolescents aged 5–18 years (13, 18, 29, 33). However, the tri-ponderal body shape index [WC z-score/($TMI^{2/3}$*$height^{1/2}$)] including TMI and WC z-score components performed more accurately in predicting MetS than BMI and TMI (Youden index: 0.85 *vs*. 0.73 *vs*. 0.70) among children and adolescents aged 7–18 years, suggesting that the combination of TMI and a WC z-score could be considered as a useful predictor for MetS in children and adolescents (30).

Overall, TMI performed similarly as compared to BMI and other indicators in predicting MetS in most of the included studies, and TMI was also suggested to be a useful tool when used in combination with other adiposity indicators (e.g., BMI and WC) for identifying MetS.

Insulin Resistance

Eight articles compared TMI and BMI for identifying insulin resistance (IR) or impaired glucose in children and adolescents (**Table 2**) (13, 14, 21, 29, 31, 32, 34, 45). Among these eight articles, seven reported that BMI (used as a continuous variable) performed similarly or marginally better than TMI for identifying IR (13, 21, 29, 31, 32, 34, 45). In addition, compared to BMI, TMI was more likely to underestimate IR (14). The inconsistent cutoffs of TMI and BMI for identifying overweight might lead to different identification of IR. When restricted to children and adolescents aged 4–18 years with overweight or obesity, WHtR or WC, used as continuous variables, seemed to perform best among the four obesity-related indicators (TMI, WC z-score, BMI z-score, and BMI) to predict IR (31, 32).

Overall, TMI did not seem to be superior to BMI for predicting IR in children and adolescents. However, WHtR or WC could be a useful indicator for identifying IR among children and adolescents with overweight and obesity.

Blood Pressure

Only five studies compared the correlation of TMI and BMI with BP, with inconsistent results (**Table 2**) (13, 17, 21, 33, 43). Although BMI correlated with BP levels stronger than TMI (17, 33), one study based on 5,814 children and adolescents aged 6–19 years showed that, similar to BMI using a continuous variable, TMI (used as a continuous variable) had a low ability to identify high BP, with an AUC of only 0.66 to predict systolic BP and 0.60 to predict diastolic BP (21); similar findings were found among 57,201 Chinese children and adolescents aged 7–18 years, among 10,441 American adolescents aged 12–18 years (13) and among Italian adolescents (43).

Overall, only a few studies examined the question and they tended to suggest that either TMI or BMI performed poorly in identifying high BP in children and adolescents, and the ability varied in different populations.

Dyslipidemia

As shown in **Table 2**, three articles showed that both TMI and BMI poorly predicted dyslipidemia (13, 14, 21). Although using the same BMI classification, total cholesterol (TC) in boys and HDL-C and TG in girls were worse among children with overweight defined by TMI than among those with normal TMI (26), BMI (as a continuous variable) seemed to be more strongly associated with TC (R^2: 0.32 *vs*. 0.27), HDL (-0.52 *vs*. -0.46), and TG (0.32 *vs*. 0.27) compared to TMI, while TMI (as a continuous variable) was more strongly associated with low-density lipoprotein-cholesterol (LDL-C) than BMI (0.38 *vs*. 0.33) (29), similar to findings on low HDL-C reported by Leone et al. (17), but inversely to findings by Alfaraidi et al. (42).

Overall, there are limited studies on the association of TMI and BMI with dyslipidemia components, and findings suggest that BMI performs better than TMI to identify high TC and TG, whereas TMI is superior to BMI to identify high LDL-C. This will need further evaluation.

Inflammatory and Liver Function Markers

As shown in **Table 2**, for C-reactive protein (CRP), the prediction accuracy of TMI and BMI z-score was similar (AUC: 0.74 *vs*. 0.73) (21), whereas other inflammatory markers including leptin, IL-6, and fetuin-A were more closely correlated with BMI than TMI (29). For liver enzymes, overweight and obese status based on TMI could significantly predict elevated serum glutamic oxaloacetic transaminase or elevated serum glutamic pyruvic transaminase, compared with overweight and obesity status based on BMI. However, different cutoffs were defined for BMI *vs*. TMI, which limits direct comparison (14). For non-alcoholic fatty liver, the discriminating ability of TMI was similarly poor as BMI, with AUC values of only 0.58 (44).

Overall, there is only limited evidence about the performance of TMI and BMI to predict inflammatory markers, which needs further research.

TMI in Childhood or Adolescence for Prediction of Specific CVRFs in Adulthood

Only four articles focused on the association of TMI *vs*. other obesity-related indicators in childhood or adolescence with

CVRFs in adulthood (35–37, 46) (**Table 2**). BMI at ages 2 to 20 years predicted the presence of CVRFs in young adults aged 20 years as well or better than TMI. For example, the ability to predict adult IR, elevated insulin levels, and hypertension seemed to be stronger for BMI *vs*. TMI (as assessed in childhood), but similar for the prediction in adults of impaired fasting plasma glucose (FPG), low HDL-C, high LDL-C, high TG, and high aortic intima-media thickness (37). Similarly, another study showed that the AUC values for TMI, or for combination of TMI and BMI, did not outperform BMI alone in predicting adult obesity, diabetes, high carotid intima-media thickness, high LDL-C, and hypertension (35). The AUCs were low for TMI (0.53, 0.56, and 0.54), as well as for other adiposity indicators such as abdominal volume index (0.61, 0.62, and 0.61), BMI (0.59, 0.60, and 0.59), and WC (0.61, 0.61, and 0.61) in childhood to predict adult homeostasis model assessment 2-insulin resistance (HOMA2-IR), HOMA2-β, and high fasting insulin (36). However, when considering growth trajectory instead of a single measurement in childhood, a persistently high TMI during adolescence had predicted diabetes quite well in adults (AUC value as high as 0.81) (46).

Overall, TMI in childhood or adolescence seems to have a lower ability than BMI and other adiposity indicators to predict specific CVRFs in adulthood, whereas TMI trajectory has a higher ability than BMI trajectory in predicting diabetes in adulthood.

DISCUSSION

Main findings

To the best of our knowledge, this is the first review to summarize the evidence regarding TMI as a screening tool for body fat and CVRFs in childhood and adulthood. TMI seemed to perform similarly or better than BMI for predicting body fat and central fat and performed similarly well as BMI in identifying MetS. However, the available evidence on the comparison of TMI and BMI (measured in childhood) for identifying specific CVRFs (in childhood or later in adulthood) including IR, high BP, dyslipidemia, and inflammation was limited and not compelling.

TMI Performed Better Than BMI to Estimate Body Fat in Children and Adolescents

Unlike for adults, no standard BF% cutoff was established to define excess adiposity among children and adolescents until now (47), and objective measurements of fat mass [e.g., DEXA, doubly-labeled water (48), and isotope dilution technique (49)] were much complex and expensive. The components of TMI or BMI (weight and height) can be simply measured using the weight scale and the stadiometer that are widely used for routine pediatric clinical practice. Therefore, in this review, we compared the performance of TMI and BMI and our study suggested that TMI performed better than BMI to estimate body fat in children and adolescents at clinical practice.

The disadvantage of BMI and the advantage of TMI to estimate body fat are as follows. First, although BMI z-score seemed to predict well total fat mass, it predicted BF% weakly with altered body composition among adolescents (50). Second, the definition of overweight and obesity using BMI should be based on sex- and age-specific percentile values in childhood, but this requires using complex tables (10), which may overestimate the actual prevalence of adiposity in children, excessively worrying families and patients (50–53), particularly for adolescents who may be more prone to fat-shaming and weight bias (54). Third, TMI (which is defined independently of age and sex) could be simpler to use compared to age- and sex-stratified BMI cutoffs and a specific cutoff of TMI has been proposed (10). A better relation of TMI with body fat mass across age may be consistent with the fact that BF% may change largely during adolescence (possibly more among girls) due to the height spurt in this age range (55). Fourth, compared to BMI, TMI was more correlated with WHtR, which is a reliable clinical measure of abdominal obesity and is consistently associated with CVRFs (56). TMI could therefore help identify children and adolescents who are overweight or obese based on BMI but also have central obesity and increased risk of CVRFs.

TMI Was More Simple and Accurate Than Other Indicators to Estimate Body Fat in Children and Adolescents

Although the RFMp calculated based on WC and height performed better to estimate BF% than TMI (25), the inter-operator variability between WC measurements is significant, which may cause more misclassification of MetS (57). Furthermore, for tall and thin people, WHtR may be unusually high, causing RFMp and RFM to tend to be 0 or negative (25). Therefore, considering the accuracy and simplicity of the use of TMI in primary health care services and its constancy in predicting adiposity at adolescence, TMI may be useful to evaluate body fat in adolescents. Yet, definite answers about the performance of BMI *vs*. TMI to predict adiposity in chidden and adolescents needs further studies using objective measurement of body fat mass (e.g., DEXA, isotope dilution) as the gold standard for comparisons, and do so in several populations, and within different ethnic, age, and sex groups.

TMI Performed Similarly as Compared to BMI and Other Indicators in Predicting MetS and Its Components

Although TMI was superior to BMI to screen central fat (19, 24), in this review, it was similar to or not better than BMI to identify MetS and specific CVRFs. One possible reason might be that adiposity defined according to TMI or BMI only accounts for one of the MetS criteria. Another reason might be the inconsistent performances of three indicators (TMI, BMI, and WC) in identifying specific CVRFs including IR, high BP, dyslipidemia, and inflammation (13, 14, 21, 29, 31, 32), which are the main components of MetS.

Age and Trajectory Influence the Association Between TMI *vs*. BMI in Childhood or Adolescence and Obesity-Related Morbidity in Adulthood

Although BMI in childhood or adolescence seemed to perform marginally better than TMI to predict obesity-related morbidity

in adulthood (35, 36), the difference disappeared after adjusting for age (35), suggesting that age might be an important confounding factor that influences the association between BMI in childhood and obesity-related morbidity in adulthood. BMI was better than TMI only in late adolescence to predict adult IR and hypertension, suggesting that the variation of BMI during adolescence influences the strength of the association (10, 37). When considering trajectories, persistently high TMI during 13 and 18 years performed better than the BMI trajectory to predict adult diabetes (46), suggesting that, during adolescence, TMI trajectory (i.e., repeated measurements) may better reflect growth and predict adult CVRD outcomes. Therefore, further prospective studies with large sample sizes, multiethnic populations, and repeated measurements of anthropometric indicators are needed to confirm these findings.

Strengths and Limitations

To the best of our knowledge, this is the first comprehensive review that compared TMI with BMI or other indicators in children and adolescents to predict obesity-related morbidity in both childhood and adulthood. Several limitations should be noted in this review. First, there was high heterogeneity between studies in the considered variables and how the adiposity cutoffs were defined, which limits direct comparisons. Second, most studies on the identification of CVRFs in childhood and adolescence were cross-sectional, which cannot prove causality (55). It must be however mentioned that a marker does not necessarily need to be causally related to an outcome to enable a good prediction. Third, a majority of the included studies came from Western countries, which limits the extrapolation of the results to other populations. Further studies with various ethnic/race groups are needed to confirm the predictive ability of TMI to predict adiposity in children and adolescents. Fourth, although TMI seems better than BMI to predict concomitant fat mass in children and adolescents, neither TMI nor BMI can distinguish fat mass from non-fat mass, and these indicators cannot replace objective measurement of fat mass (e.g., DEXA, isotope dilution). Again, an ultimate fully valid method to compare how BMI or TMI predicts adiposity should rely on objectively measured adiposity as the gold standard (e.g., DEXA, isotope dilution methods) and use a similar dichotomization of categories of elevated BMI or elevated TMI (e.g., using the same percentile cutoffs, e.g., p80 or p90) to enable valid comparisons; this was only rarely performed in the considered studies.

CONCLUSIONS

In conclusion, TMI only requires a single threshold according to sex (i.e., no need for sex- and age-specific thresholds) and TMI seems to predict adiposity similarly or better in children and adolescents than BMI. In addition, TMI seemed to perform similarly as BMI for identifying MetS. However, the clinical use of TMI *vs*. BMI in childhood, in order to predict specific elevated CVRFs in childhood or later in adulthood, is still not definitive and needs further studies, particularly those with a longitudinal design.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding author.

AUTHOR CONTRIBUTIONS

BX and PB designed the research. JS and RY conducted the literature search and performed the statistical analysis of the data. JS, BX, and PB wrote the manuscript draft. JS, BX, MZ, and PB contributed to the critical revision of the manuscript for important intellectual content. All authors contributed to the article and approved the submitted version.

FUNDING

This work was supported by National Natural Science Foundation of China, Grant/Award Number: 81673195; Youth Team of Humanistic and Social Science of Shandong University.

REFERENCES

1. N. C. D. Risk Factor Collaboration. Worldwide Trends in Body-Mass Index, Underweight, Overweight, and Obesity From 1975 to 2016: A Pooled Analysis of 2416 Population-Based Measurement Studies in 128.9 Million Children, Adolescents, and Adults. *Lancet* (2017) 390(10113):2627–42. doi: 10.1016/S0140-6736(17)32129-3
2. Jing L, Nevius CD, Friday CM, Suever JD, Pulenthiran A, Mejia-Spiegeler A, et al. Ambulatory Systolic Blood Pressure and Obesity Are Independently Associated With Left Ventricular Hypertrophic Remodeling in Children. *J Cardiovasc Magn Reson* (2017) 19(1):86. doi: 10.1186/s12968-017-0401-3
3. Park MH, Skow A, De Matteis S, Kessel AS, Saxena S, Viner RM, et al. Adiposity and Carotid-Intima Media Thickness in Children and Adolescents: A Systematic Review. *BMC Pediatr* (2015) 15:161. doi: 10.1186/s12887-015-0478-5
4. Cho H, Kim JH. Prevalence of Microalbuminuria and its Associated Cardiometabolic Risk Factors in Korean Youth: Data From the Korea National Health and Nutrition Examination Survey. *PloS One* (2017) 12(6):e0178716. doi: 10.1371/journal.pone.0178716
5. Anderson EL, Howe LD, Jones HE, Higgins JP, Lawlor DA, Fraser A. The Prevalence of Non-Alcoholic Fatty Liver Disease in Children and Adolescents: A Systematic Review and Meta-Analysis. *PloS One* (2015) 10(10):e0140908. doi: 10.1371/journal.pone.0140908
6. Weihrauch-Bluher S, Schwarz P, Klusmann JH. Childhood Obesity: Increased Risk for Cardiometabolic Disease and Cancer in Adulthood. *Metabolism* (2019) 92:147–52. doi: 10.1016/j.metabol.2018.12.001
7. Park MH, Falconer C, Viner RM, Kinra S. The Impact of Childhood Obesity on Morbidity and Mortality in Adulthood: A Systematic Review. *Obes Rev* (2012) 13(11):985–1000. doi: 10.1111/j.1467-789X.2012.01015.x
8. Simmonds M, Burch J, Llewellyn A, Griffiths C, Yang H, Owen C, et al. The Use of Measures of Obesity in Childhood for Predicting Obesity and the Development of Obesity-Related Diseases in Adulthood: A Systematic Review and Meta-Analysis. *Health Technol Assess* (2015) 19(43):1–336. doi: 10.3310/hta19430

9. Rothman KJ. BMI-Related Errors in the Measurement of Obesity. *Int J Obes (Lond)* (2008) 32(Suppl 3):S56–9. doi: 10.1038/ijo.2008.87
10. Peterson CM, Su H, Thomas DM, Heo M, Golnabi AH, Pietrobelli A, et al. Tri-Ponderal Mass Index *vs* Body Mass Index in Estimating Body Fat During Adolescence. *JAMA Pediatr* (2017) 171(7):629–36. doi: 10.1001/jamapediatrics.2017.0460
11. Schmidt SC, Bosy-Westphal A, Niessner C, Woll A. Representative Body Composition Percentiles From Bioelectrical Impedance Analyses Among Children and Adolescents. The MoMo Study. *Clin Nutr* (2019) 38(6):2712–20. doi: 10.1016/j.clnu.2018.11.026
12. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a Standard Definition for Child Overweight and Obesity Worldwide: International Survey. *BMJ* (2000) 320(7244):1240–3. doi: 10.1136/bmj.320.7244.1240
13. Wang X, Dong B, Ma J, Song Y, Zou Z, Arnold L. Role of Tri-Ponderal Mass Index in Cardio-Metabolic Risk Assessment in Children and Adolescents: Compared With Body Mass Index. *Int J Obes (Lond)* (2020) 44(4):886–94. doi: 10.1038/s41366-019-0416-y
14. Akcan N, Bundak R. Accuracy of Tri-Ponderal Mass Index and Body Mass Index in Estimating Insulin Resistance, Hyperlipidemia, Impaired Liver Enzymes or Thyroid Hormone Function and Vitamin D Levels in Children and Adolescents. *J Clin Res Pediatr Endocrinol* (2019) 11(4):366–73. doi: 10.4274/jcrpe.galenos.2019.2018.0279
15. Gomes TN, Nevill A, Katzmarzyk PT, Pereira S, Dos Santos MM, Buranarugsa R, et al. Identifying the Best Body-Weight-Status Index Associated With Metabolic Risk in Youth. *Scand J Med Sci Sports* (2018) 28(11):2375–83. doi: 10.1111/sms.13249
16. Radetti G, Fanolla A, Grugni G, Lupi F, Sartorio A. Indexes of Adiposity and Body Composition in the Prediction of Metabolic Syndrome in Obese Children and Adolescents: Which Is the Best? *Nutr Metab Cardiovasc Dis* (2019) 29(11):1189–96. doi: 10.1016/j.numecd.2019.06.011
17. Leone A, Vizzuso S, Brambilla P, Mameli C, Ravella S, De Amicis R, et al. Evaluation of Different Adiposity Indices and Association With Metabolic Syndrome Risk in Obese Children: Is There a Winner? *Int J Mol Sci* (2020) 21 (11):4083. doi: 10.3390/ijms21114083
18. Jiang Y, Dou YL, Xiong F, Zhang L, Zhu GH, Wu T, et al. Waist-To-Height Ratio Remains an Accurate and Practical Way of Identifying Cardiometabolic Risks in Children and Adolescents. *Acta Paediatr* (2018) 107:1629–34. doi: 10.1111/apa.14323
19. Sims ED, Wang KW, Fleming A, Johnston DL, Zelcer SM, Rassekh SR, et al. Tri-Ponderal Mass Index in Survivors of Childhood Brain Tumors: A Cross-Sectional Study. *Sci Rep* (2018) 8(1):16336. doi: 10.1038/s41598-018-34602-5
20. Moselakgomo VK, Van Staden M. Diagnostic Accuracy of Tri-Ponderal Mass Index and Body Mass Index in Estimating Overweight and Obesity in South African Children. *Afr J Prim Health Care Fam Med* (2019) 11(1):e1–7. doi: 10.4102/phcfm.v11i1.1949
21. Ashley-Martin J, Ensenauer R, Maguire B, Kuhle S. Predicting Cardiometabolic Markers in Children Using Tri-Ponderal Mass Index: A Cross-Sectional Study. *Arch Dis Child* (2019) 104(6):577–82. doi: 10.1136/archdischild-2018-316028
22. Zaniqueli D, Oliosa PR, Neves FS, Pani VO, Martins CR, de Souza Pecanha MA, et al. Ponderal Index Classifies Obesity in Children and Adolescents More Accurately Than Body Mass Index Z-Scores. *Pediatr Res* (2019) 86 (1):128–33. doi: 10.1038/s41390-019-0395-7
23. De Lorenzo A, Romano L, Di Renzo L, Gualtieri P, Salimei C, Carrano E, et al. Triponderal Mass Index Rather Than Body Mass Index: An Indicator of High Adiposity in Italian Children and Adolescents. *Nutrition* (2019) 60:41–7. doi: 10.1016/j.nut.2018.09.007
24. Nascimento VG, Bertoli CJ, Gallo PR, Abreu LC, Leone C. Tri-Ponderal Mass Index: A Screening Tool for Risk of Central Fat Accumulation in Brazilian Preschool Children. *Medicina (Kaunas)* (2019) 55(9):557. doi: 10.3390/medicina55090577
25. Woolcott OO, Bergman RN. Relative Fat Mass as an Estimator of Whole-Body Fat Percentage Among Children and Adolescents: A Cross-Sectional Study Using NHANES. *Sci Rep* (2019) 9(1):15279. doi: 10.1038/s41598-019-51701-z
26. Park HK, Shim YS. Distribution of Tri-Ponderal Mass Index and its Relation to Body Mass Index in Children and Adolescents Aged 10 to 20 Years. *J Clin Endocrinol Metab* (2020) 105(3):e826–e34. doi: 10.1210/clinem/dgaa030
27. Ramírez-Vélez R, Correa-Bautista J, Carrillo H, González-Jiménez E, Schmidt-RioValle J, Correa-Rodríguez M, et al. Tri-Ponderal Mass Index *vs*. Fat Mass/Height^3 as a Screening Tool for Metabolic Syndrome Prediction in Colombian Children and Young People. *Nutrients* (2018) 10(4):412. doi: 10.3390/nu10040412
28. Shim YS. The Relationship Between Tri-Ponderal Mass Index and Metabolic Syndrome and Its Components in Youth Aged 10-20 Years. *Sci Rep* (2019) 9 (1):14462. doi: 10.1038/s41598-019-50987-3
29. Akcan N, Obaid M, Salem J, Bundak R. Evidence in Obese Children: Contribution of Tri-Ponderal Mass Index or Body Mass Index to Dyslipidemia, Obesity-Inflammation, and Insulin Sensitivity. *J Pediatr Endocrinol Metab* (2020) 33(2):223–31. doi: 10.1515/jpem-2019-0106
30. Arsang-Jang S, Kelishadi R, Esmail Motlagh M, Heshmat R, Mansourian M. Temporal Trend of Non-Invasive Method Capacity for Early Detection of Metabolic Syndrome in Children and Adolescents: A Bayesian Multilevel Analysis of Pseudo-Panel Data. *Ann Nutr Metab* (2019) 75(1):55–65. doi: 10.1159/000500274
31. Umano GR, Di Sessa A, Cirillo G, Ursi D, Marzuillo P, Miraglia Del Giudice E. Waist-To-Height Ratio Is More Strongly Associated Than Other Weight-Related Anthropometric Measures With Metabolic Variables. *Acta Paediatr* (2019) 108(12):2296–97. doi: 10.1111/apa.14992
32. Matsuo AR, Lopes WA, Locatelli JC, Simoes CF, de Oliveira GH, Nardo NJr. Tri-Ponderal Mass Index as a Tool for Insulin Resistance Prediction in Overweight Adolescents: A Cross-Sectional Study. *Nutrition* (2020) 74:110744. doi: 10.1016/j.nut.2020.110744
33. Khoshhali M, Heidari-Beni M, Qorbani M, Motlagh ME, Ziaodini H, Heshmat R, et al. Tri-Ponderal Mass Index and Body Mass Index in Prediction of Pediatric Metabolic Syndrome: The CASPIAN-V Study. *Arch Endocrinol Metab* (2020) 64(2):171–78. doi: 10.20945/2359-3997000000206
34. Neves FS, Alvim RO, Zaniqueli D, Pani VO, Martins CR, Pecanha MAS, et al. Tri-Ponderal Mass Index Is Useful for Screening Children and Adolescents With Insulin Resistance. *Rev Paul Pediatr* (2020) 38:e2019066. doi: 10.1590/1984-0462/2020/38/2019066
35. Wu F, Buscot MJ, Juonala M, Hutri-Kahonen N, Viikari JSA, Raitakari OT, et al. Association of Youth Triponderal Mass Index *vs* Body Mass Index With Obesity-Related Outcomes in Adulthood. *JAMA Pediatr* (2018) 172 (12):1192–95. doi: 10.1001/jamapediatrics.2018.3034
36. Wu F, Ho V, Fraser BJ, Schmidt MD, Dwyer T, Venn AJ, et al. Predictive Utility of Childhood Anthropometric Measures on Adult Glucose Homeostasis Measures: A 20-Year Cohort Study. *Int J Obes (Lond)* (2018) 42(10):1762–70. doi: 10.1038/s41366-018-0177-z
37. Wu F, Buscot MJ, Niinikoski H, Rovio SP, Juonala M, Sabin MA, et al. Age-Specific Estimates and Comparisons of Youth Tri-Ponderal Mass Index and Body Mass Index in Predicting Adult Obesity-Related Outcomes. *J Pediatr* (2020) 218:198–203 e6. doi: 10.1016/j.jpeds.2019.10.062
38. Karchynskaya V, Kopcakova J, Klein D, Gaba A, Madarasova-Geckova A, van Dijk JP, et al. Is BMI a Valid Indicator of Overweight and Obesity for Adolescents? *Int J Environ Res Public Health* (2020) 17(13):4815. doi: 10.3390/ijerph17134815
39. Zeng X, Zhang Y, Kwong JS, Zhang C, Li S, Sun F, et al. The Methodological Quality Assessment Tools for Preclinical and Clinical Studies, Systematic Review and Meta-Analysis, and Clinical Practice Guideline: A Systematic Review. *J Evid Based Med* (2015) 8(1):2–10. doi: 10.1111/jebm.12141
40. Wells GA, Shea B, O'Connell D, Peterson J, Welch V, Losos M, et al. *The Newcastle-Ottawa Scale (NOS) for Assessing the Quality If Nonrandomized Studies in Meta-Analyses*. Available at: http://www.ohri.ca/programs/clinical_epidemiology/oxford.asp (Accessed December 23, 2020).
41. Ye XF, Dong W, Tan LL, Zhang ZR, Qiu YL, Zhang J. Identification of the Most Appropriate Existing Anthropometric Index for Home-Based Obesity Screening in Children and Adolescents. *Public Health* (2020) 189:20–5. doi: 10.1016/j.puhe.2020.09.007
42. Alfaraidi H, Wicklow B, Dart AB, Sellers E, McGavock J, Thabane L, et al. The Tri-Ponderal Mass Index Is Associated With Adiposity in Adolescent Type 2 Diabetes Mellitus: A Cross-Sectional Analysis. *Sci Rep* (2021) 11(1):9111. doi: 10.1038/s41598-021-88705-7
43. Malavazos AE, Capitanio G, Milani V, Ambrogi F, Matelloni IA, Basilico S, et al. Tri-Ponderal Mass Index *vs* Body Mass Index in Discriminating Central

Obesity and Hypertension in Adolescents With Overweight. *Nutr Metab Cardiovasc Dis* (2021) 31(5):1613–21. doi: 10.1016/j.numecd.2021.02.013
44. Umano GR, Grandone A, Di Sessa A, Cozzolino D, Pedulla M, Marzuillo P, et al. Pediatric Obesity-Related Non-Alcoholic Fatty Liver Disease: Waist-to-Height Ratio Best Anthropometrical Predictor. *Pediatr Res* (2020) 90(1):166-70. doi: 10.1038/s41390-020-01192-w
45. Calcaterra V, Verduci E, Schneider L, Cena H, De Silvestri A, Vizzuso S, et al. Sex-Specific Differences in the Relationship Between Insulin Resistance and Adiposity Indexes in Children and Adolescents With Obesity. *Children (Basel)* (2021) 8(6):449. doi: 10.3390/children8060449
46. Wu YF, Fan HY, Chen YC, Kuo KL, Chien KL. Adolescent Tri-Ponderal Mass Index Growth Trajectories and Incident Diabetes Mellitus in Early Adulthood. *J Clin Endocrinol Metab* (2021) 106(8):e2919-27. doi: 10.1210/clinem/dgab235
47. De Lorenzo A, Soldati L, Sarlo F, Calvani M, Di Lorenzo N, Di Renzo L. New Obesity Classification Criteria as a Tool for Bariatric Surgery Indication. *World J Gastroenterol* (2016) 22(2):681–703. doi: 10.3748/wjg.v22.i2.681
48. LeMura LM, Maziekas MT. Factors That Alter Body Fat, Body Mass, and Fat-Free Mass in Pediatric Obesity. *Med Sci Sports Exerc* (2002) 34(3):487–96. doi: 10.1097/00005768-200203000-00016
49. Ben Jemaa H, Mankai A, Khlifi S, Minaoui R, Ghozzi D, Zediri M, et al. Development and Validation of Impedance-Based Equations for the Prediction of Total Body Water and Fat-Free Mass in Children Aged 8-11 Years. *Clin Nutr* (2019) 38(1):227–33. doi: 10.1016/j.clnu.2018.01.028
50. Vanderwall C, Randall Clark R, Eickhoff J, Carrel AL. BMI Is a Poor Predictor of Adiposity in Young Overweight and Obese Children. *BMC Pediatr* (2017) 17(1):135. doi: 10.1186/s12887-017-0891-z
51. Buss J. Limitations of Body Mass Index to Assess Body Fat. *Workplace Health Saf* (2014) 62(6):264. doi: 10.1177/216507991406200608
52. Weber DR, Moore RH, Leonard MB, Zemel BS. Fat and Lean BMI Reference Curves in Children and Adolescents and Their Utility in Identifying Excess Adiposity Compared With BMI and Percentage Body Fat. *Am J Clin Nutr* (2013) 98(1):49–56. doi: 10.3945/ajcn.112.053611
53. Vanderwall C, Eickhoff J, Randall Clark R, Carrel AL. BMI Z-Score in Obese Children Is a Poor Predictor of Adiposity Changes Over Time. *BMC Pediatr* (2018) 18(1):187. doi: 10.1186/s12887-018-1160-5
54. Ikeda JP, Crawford PB, Woodward-Lopez G. BMI Screening in Schools: Helpful or Harmful. *Health Educ Res* (2006) 21(6):761–9. doi: 10.1093/her/cyl144
55. Hermanussen M, Largo RH, Molinari L. Canalisation in Human Growth: A Widely Accepted Concept Reconsidered. *Eur J Pediatr* (2001) 160(3):163–7. doi: 10.1007/s004310000706
56. Abraham TM, Pedley A, Massaro JM, Hoffmann U, Fox CS. Association Between Visceral and Subcutaneous Adipose Depots and Incident Cardiovascular Disease Risk Factors. *Circulation* (2015) 132(17):1639–47. doi: 10.1161/CIRCULATIONAHA.114.015000
57. Panoulas VF, Ahmad N, Fazal AA, Kassamali RH, Nightingale P, Kitas GD, et al. The Inter-Operator Variability in Measuring Waist Circumference and Its Potential Impact on the Diagnosis of the Metabolic Syndrome. *Postgrad Med J* (2008) 84(993):344–7. doi: 10.1136/pgmj.2008.068825

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 10 November 2021
doi: 10.3389/fendo.2021.726154

Association of Serum Galectin-3-Binding Protein and Metabolic Syndrome in a Chinese Adult Population

Shihan Zhen[1], *Ruoxin Cai*[1], *Xuelian Yang*[1], *Yanan Ma*[2] *and Deliang Wen*[1]*

[1] Institute of Health Sciences, China Medical University, Shenyang, China, [2] School of Public Health, China Medical University, Shenyang, China

Background: Galectin-3-binding protein (GAL-3BP) is a ubiquitous and multifunctional secreted glycoprotein, which functions in innate immunity and has been highlighted as a potential mediator of adipose inflammation in obesity. In this study, we aimed to identify whether GAL-3BP is a novel biological marker for metabolic syndrome (MetS).

Methods: The biochemical and anthropometric variables of the 570 participants in this study were evaluated using standard procedures. Their serum GAL-3BP levels were measured using enzyme-linked immunosorbent assay (ELISA), while the association between the glycoprotein and MetS was analyzed using multiple logistic regression analyses. Moreover, an experimental MetS model was established. The expression of GAL-3BP in serum and adipose tissue was measured using ELISA and western blotting. Lipid accumulation was determined with the use of immunohistochemistry and immunofluorescent staining.

Results: The serum GAL-3BP level was found to be positively associated with MetS. The logistic regression analyses demonstrated that participants expressing the upper levels of GAL-3BP were more likely to develop MetS than those expressing less of the glycoprotein (OR = 2.39, 95%CI: 1.49, 3.83). The association between the serum GAL-3BP level and MetS was found preferentially in postmenopausal women (OR = 2.30, 95%CI: 1.31, 4.05). In addition, GAL-3BP was increased in the serum and visceral adipose tissue (VAT) of high fat diet (HFD) mice. Moreover, GAL-3BP was highly expressed in VAT macrophages.

Conclusions: This study confirmed serum GAL-3BP to be positively associated with MetS, highlighting it as a useful biological marker of MetS in Chinese participants.

Keywords: metabolic syndrome, inflammation, biomarker, sex difference, galectin-3-binding protein

OPEN ACCESS

Edited by:
Mostafa Qorbani,
Alborz University of Medical Sciences, Iran

Reviewed by:
Valeria Guglielmi,
University of Rome Tor Vergata, Italy
Ehsaneh Taheri,
Shahid Beheshti University of Medical Sciences, Iran
Victoria Sundblad,
Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina

***Correspondence:**
Deliang Wen
dlwen@cmu.edu.cn

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *16 June 2021*
Accepted: *23 September 2021*
Published: *10 November 2021*

Citation:
Zhen S, Cai R, Yang X, Ma Y and Wen D (2021) Association of Serum Galectin-3-Binding Protein and Metabolic Syndrome in a Chinese Adult Population. Front. Endocrinol. 12:726154. doi: 10.3389/fendo.2021.726154

INTRODUCTION

Galectin-3 binding protein (GAL-3BP) is a ubiquitous multifunctional secretory glycoprotein, which was initially identified as having innate immune function in humans following viral and bacterial infections (1). GAL-3BP has several targets, such as Galectin-1, Galectin-3, Galectin-7, Galectin-9, and GAL-3BP, which interact with extracellular matrix proteins and cell surface receptors such as β1-integrins, calcineurin, and NFATc1, thereby regulating cell–cell and cell–

matrix interactions (1–5). GAL-3BP is also known to regulate the activation of cyclophilin C, which regulates phagocytosis through the activation of NFAT in macrophages (6). GAL-3BP interacts with a group of target molecules through its multiple functional domains and participates in a wide range of physiological and pathological processes such as cell growth, cellular adhesion, inflammation, and visceral fat increase (1, 4, 7).

Recent studies have suggested that GAL-3BP plays a causal role in innate immunity (1), insulin resistance (8), and chronic low-grade inflammation (9). In humans, GAL-3BP is reported to be elevated in the plasma of obese individuals (10, 11) or those with several symptoms of metabolic syndrome (MetS) (11, 12). In addition, the glycoprotein is secreted from visceral adipose tissues (VAT) (13). In a mouse model, serum GAL-3BP levels increased under a high-fat high-cholesterol diet (14), and several studies have shown that serum GAL-3BP levels can predict the severity of liver disease, especially non-alcoholic fatty liver disease (NAFLD) (7, 14).

MetS refers to the cluster of biological factors that feature in type 2 diabetes mellitus, hypertension, dyslipidemia, and abdominal obesity (15) and is becoming a major public health issue (16). With the increase in obesity, the incidence of MetS in the Chinese population has increased rapidly from 29.65% in 2005 to 45.49% in 2014 (17). This increase also elevates the incidence of arthritis, diabetes, and cardiovascular disease (18, 19). Therefore, it is vital to examine the potential mechanisms that underlie MetS and identify biomarkers that will help to assess the risk of developing the syndrome.

Although the relationship between GAL-3BP and human obesity has been demonstrated (11), the significance of GAL-3BP as a biomarker for MetS has not been fully examined to date. The aims of the study were to investigate the clinical significance of serum GAL-3BP levels in determining the complex phenotype of MetS and evaluate whether GAL-3BP can act as a suitable biomarker for MetS by assessing the correlation between them.

MATERIALS AND METHODS

Human Subjects

The study was based on the Major Chronic Diseases Prevention and Control Cohort in Northeast China, a well-designed prospective cohort used to investigate environmental and genetic factors in non-communicable chronic disease. A face-to-face interview was conducted to collect information using a standardized questionnaire. The eligibility of the participants was defined as those who had resided in the area for at least 5 years, could partake in barrier-free communication, were compliant, and were 1) free from severe physical disabilities, cancer, cerebrovascular disease, severe liver and kidney diseases or psychological disorders or dementia over the past 6 months; 2) not currently diagnosed with a communicable disease; 3) not pregnant. All participants provided written informed consent. Between September and December 2019, a total of 675 participants from Yuhong district were enrolled using a multistage sampling technique. Sixty-nine participants were excluding because of hemolysis or chylous blood (fatty blood). Of the remaining 606 participants, 36 completed only the short questionnaire. After exclusions, the data for 570 participants were made available for the current investigation. Participants provided written informed consent to undergo venipuncture and were all told of the intended use of the samples. The research was approved by the Ethics Committee of China Medical University (CMU).

Waist circumference (WC) was measured at umbilicus level in the standing position. Systolic (SBP) and diastolic (DBP) blood pressure readings were taken using an automatic electronic sphygmomanometer (HEM-907; Omron, Tokyo, Japan). Blood was collected from the anterior humerus vein in the morning after 12-h of fasting without the intake of medication. Fasting plasma glucose (FPG), triglyceride (TG), low- and high-density lipoprotein-cholesterol (LDL- and HDL-C), aspartate aminotransferase (AST) and alanine aminotransferase (ALT) levels were determined using standard procedures.

Definition of Metabolic Syndrome

Participants with MetS were defined according to the criteria set out by the International Diabetes Federation (20). MetS was diagnosed when subjects presented with abdominal obesity (defined as WC ≥ 90 cm for males or ≥80 cm for females) and two or more of the following criteria: 1) high blood pressure (SBP ≥ 130mmHg or DBP ≥ 85mmHg); 2) elevated plasma glucose (FPG ≥ 5.6mmol/L); 3) elevated TG (TG ≥ 1.7mmol/L); or 4) low HDL-C (HDL-C < 1.04mmol/L for males or < 1.3mmol/L for females).

Covariates

Covariates including age, gender, nationality (Han or other), educational attainment (illiterate or primary school; junior middle school; high middle school; and college or higher), menopausal status (pre- or post-menopausal), AST (U/L), and ALT (U/L) were collected using face-to-face interviews and general information questionnaires.

Animals

C57BL/6 mice weighing approximately 20 g at the beginning of the experimental procedure were used. Mice were housed in a 12 h/12 h light/dark cycle and given distilled water and feed. Mice over 12–16 weeks-of-age were randomly divided into two groups. The control diet (CD) group was provided with a standard CD. The high fat diet (HFD) group was fed on a diet in which 60% of the calories were obtained from fat (Research diet #D12492) for 12 weeks. MetS was induced by feeding the animals with a HFD (Research Diets, New Brunswick, NJ), which was consistent with previously published work (21). Blood samples

Abbreviations: GAL-3BP, Galectin-3-binding protein; MetS, metabolic syndrome; WC, waist circumference; SBP, systolic blood pressure; DBP, diastolic blood pressure; FPG, fasting plasma glucose; TG, triglycerides; HDL-C, high-density lipoprotein-cholesterol; LDL-C, low-density lipoprotein-cholesterol; AST, aspartate aminotransferase; ALT, alanine aminotransferase; ECM, extracellular matrix; ELISA, enzyme linked immunosorbent assay; VAT, visceral adipose tissue; CD, Control diet; HFD, High fat diet.

were used to measure blood glucose, lipoprotein, and TG levels. Food intake, water consumption, weight, and body compositions were measured weekly, and the average food and water consumption was calculated accordingly. We used gonadal white adipose tissue for measurements. Before tissue collection, the mice fasted for 16 h and were weighed, before samples were excised and fixed in 4% paraformaldehyde buffer for histopathological assessment. Serum and various tissue samples were collected and frozen at −80°C. All animal procedures were approved by the Animal Ethics Committee of China Medical University.

Measurements of GAL-3BP

Human Galectin-3BP ELISA (2H-KMLJh314728) and Mouse Galectin-3BP ELISA (2M-KMLJM228552m) kits were purchased from CAMILO biological (Nanjing, China). Both kits were used in accordance with the manufacturer's instructions. Standards provided with the kits were diluted to produce a gradient of biomarker concentrations to obtain standard curves.

Western Blots

Visceral adipose tissues were prepared and lysed according to standard protocols. Antibodies to G3BP (ab181150, 1:1000) and GAPDH (ab8245, 1:2000) were purchased from Abcam. Blotting membranes were incubated with the primary antibody at 4°C overnight and the secondary anti-rabbit IgG (#32731; 1:10000; Thermo Scientific) at room temperature for 1 h. The resulting bands were visualized using a Tanon 5500 imaging system (Tanon, Shanghai, China). The results were quantified using ImageJ software (National Institute of Mental Health, USA).

Immunohistochemistry

Tissues were fixed overnight in 4% paraformaldehyde in PBS, dehydrated in a graded ethanol series, and washed with xylene. Tissues were embedded in paraffin and sectioned as 5 μm. Single-label immunohistochemistry was performed on adipose tissues. Macrophages were detected using a monoclonal antibody against F4/80 (ab6640, 1:100). Histopathological images were captured by immunofluorescence microscopy (80I, Nikon Corporation, Tokyo, Japan). Three sections per mouse were analyzed and n = 11–20.

Immunofluorescence Staining

After being deparaffinized in xylene and rehydrated using an ascending ethanol series, the slides were permeabilized with 0.1% Triton-X 100 for 5 min, blocked with 10% goat serum in PBST (PBS with 0.05% Tween 20) for 1 h at 37°C, and incubated with GAL-3BP (1:100) and F4/80 (1:100) at 4°C overnight. After washing with PBST, the coverslips were mounted with anti-fade reagent and 4′, 6′-diamidino-2-phenylindole (DAPI) (Life Technologies, Waltham, MA, USA). Images were acquired using a Leica DFC310 FX digital camera connected to a Leica DMI4000 B light microscope (Wetzlar, Germany).

Statistical Analysis

Descriptive information was presented is means with standard deviations. ANOVA tests for continuous variables and chi-square tests for categorical variables were used to compare participants with and without MetS. GAL-3BP concentration was divided into tertiles, and an increase from T1 to T3 was assumed (22). The upper strata of GAL-3BP levels were defined as T2 and T3, whereas and lower strata were defined as T1. The cut-off value in the present study were 45.13 (ng/ml).

Logistic regression models were used to estimate the odds ratios (OR) and 95% confidence intervals (95%CI) for MetS and serum GAL-3BP levels. The age-adjusted model was adjusted for age, and the multiple-adjusted model was adjusted for gender, national, educational attainment, AST, and ALT. A P-value < 0.05 indicated statistical significance. Statistical analyses were carried out using SPSS 25.0 (SPSS, Inc., Chicago, IL) and Stata 13.0 (StataCorp, College Station, TX, USA), while R (R studio, USA) and Graphpad Prism 5.0 (GraphPad Inc., La Jolla, CA) were adopted for graph preparation.

RESULTS

Table 1 shows the characteristics of the participants; 25.09% of participants had MetS. Compared with subjects without MetS, those with MetS exhibited higher values for WC, BMI, blood pressure, FPG, TG, HDL-C, LDL-C, and GAL-3BP. The median overall GAL-3BP concentration was 70.55 ng/ml (SD = 52.12, SEM = 2.18). The median GAL-3BP concentration for participants with MetS was 80.31 ng/ml (SD = 68.21, SEM = 5.70), whereas that for participants without MetS was 67.28 ng/ml (SD = 45.10, SEM = 2.18). The median GAL-3BP concentration in females was 72.20 ng/ml (SD = 55.35, SEM = 2.80), whereas that in males was 66.95 ng/ml (SD = 44.17, SEM = 3.30). Further, the median GAL-3BP concentration in females with MetS was 80.77 ng/ml (SD = 69.74, SEM = 6.56), whereas that in females without MetS was 68.71 ng/ml (SD = 48.00, SEM = 2.88) (p <0.05). The median GAL-3BP concentration in males with MetS was 78.59 ng/ml (SD = 63.19, SEM = 11.54), whereas that in males without MetS was 64.61 ng/ml (SD = 39.13, SEM = 3.21) (p >0.05). In addition, Gal-3BP levels in females and males were shown in **Supplemental Table 1**.

The Association Between GAL-3BP and MetS

After adjusting for confounders, participants with the higher GAL-3BP levels showed higher odds of having MetS than those in with lower levels of GAL-3BP (OR = 2.39, 95%CI: 1.49, 3.83). To further define any sex differences, we compared the results obtained for male and female subjects. Females with higher levels of GAL-3BP had higher odds of having MetS than those with lower levels of GAL-3BP (OR=2.31, 95%CI: 1.35, 3.94), whereas, males with higher GAL-3BP levels also showed higher odds of having MetS than those with lower levels of GAL-3BP (OR=3.02, 95%CI: 1.03, 8.34). Further, females were divided into premenopausal and postmenopausal groups. In the postmenopausal group, participants with higher GAL-3BP levels had a higher incidence of MetS than those with lower levels of the glycoprotein (OR=2.30, 95%CI: 1.31, 4.05). These results are shown in **Tables 2**, **3**.

TABLE 1 | Characteristics of study participants with and without MetS.

Characteristic	Participants without MetS (n = 427)	Participants with MetS (n = 143)	*P* value
Age, Mean (SD)	60.74 (9.95)	64.92 (7.48)	**<0.001**
Female, No. (%)	278 (65.11)	113 (79.02)	**0.002**
National (Han), No. (%)	368 (86.18)	130 (90.91)	0.141
Educational attainment, No. (%)			0.238
Illiterate or primary school	97 (22.72)	38 (26.57)	
Junior middle school	165 (38.64)	49 (34.27)	
High middle school	134 (31.38)	51 (35.66)	
College or higher	31 (7.26)	5 (3.5)	
Anthropometry			
WC, (cm), Mean. (SD)	78.84 (8.09)	89.04 (6.76)	**<0.001**
BMI, (kg/m2), Mean. (SD)	24.67 (2.92)	27.71 (3.13)	**<0.001**
SBP, (mmHg), Mean. (SD)	126.90 (14.47)	135.59 (11.81)	**<0.001**
DBP, (mmHg), Mean. (SD)	75.31 (8.80)	77.91 (9.33)	**0.003**
Laboratory examinations			
FPG, (mmol/L), Mean. (SD)	5.34 (1.21)	6.17 (1.78)	**<0.001**
TG, (mmol/L), Mean. (SD)	1.55 (1.33)	2.34 (1.36)	**<0.001**
HDL-C, (mmol/L), Mean. (SD)	1.31 (0.29)	1.22 (0.34)	**0.003**
LDL-C, (mmol/L), Mean. (SD)	3.25 (0.76)	3.45 (0.77)	**0.006**
AST, (U/L), Mean (SD)	26.36 (10.04)	26.22 (9.87)	0.882
ALT, (U/L), Mean (SD)	24.67 (17.75)	26.76 (13.47)	0.200
G3BP, (ng/ml), Mean (SD)	67.28 (45.10)	80.31 (68.21)	0.001

P-values < 0.05 are bold.

In addition, the association between GAL-3BP levels and MetS components were shown in **Supplemental Table 2**.

GAL-3BP Was Increased in the Serum and VAT of Mice on a High-Fat Diet

To confirm the GAL-3BP expression pattern in MetS mice, we fed the mice with a HFD. Data on the assessment of MetS parameter in mice were shown in **Supplemental Table 3**. As shown in **Figure 1A**, GAL-3BP was found to be increased significantly in the serum of the HFD group compared with that in the control group. We then assessed whether GAL-3BP was secreted from VAT and found that levels were increased in the HFD group compared with the control group (**Figures 1B, C**).

GAL-3BP Was Highly Expressed in VAT Macrophages

To verify the VAT cell types that secreted GAL-3BP, we tested the expression of the glycoprotein in the IHC. As shown in **Figure 2A**, in HFD group VAT, the adipocytes were bigger in the HFD group than the CD group. GAL-3BP was highly expressed as in a crown shape, which indicated that the macrophages may secrete the protein in the VAT of the HFD group. Next, using the macrophage marker F4/80 to locate the cells, we found that GAL-3BP was expressed in cells identified by F4/80 (**Figure 2B**). In the VAT of the HFD group, GAL-3BP and F4/80 were more highly expressed than in the CD group (**Figure 2B**).

DISCUSSION

In this study, we demonstrated that the serum GAL-3BP levels were positively correlated with the incidence of MetS in humans, particularly in postmenopausal females. Serum GAL-3BP may therefore serve as a useful biological marker for MetS. GAL-3BP was highly expressed in the VAT of MetS mice, suggesting that GAL-3BP expression may represent a biological process that

TABLE 2 | Association between Gal-3BP level and MetS.

	Level of GAL-3BP		*P* value
	Lower	Upper	
Total (n=570)			
Age-adjusted model	1 (Reference)	**2.40 (1.52, 3.80)**	**<0.001**
Multiple-adjusted model	1 (Reference)	**2.39 (1.49, 3.83)**	**<0.001**
Male (n=179)			
Age-adjusted model	1 (Reference)	**3.03 (1.09, 8.39)**	**0.033**
Multiple-adjusted model[a]	1 (Reference)	**3.02 (1.03, 8.34)**	**0.043**
Female (n=391)			
Age-adjusted model	1 (Reference)	**2.32 (1.37, 3.93)**	**0.002**
Multiple-adjusted model[b]	1 (Reference)	**2.31 (1.35, 3.94)**	**0.002**

OR, Odds ratio; CI, confidence interval; Age-adjusted model, adjusted for age (in years); Multiple-adjusted model, additional adjusted for gender, national, educational attainment, AST(U/L) and ALT(U/L). [a]Multiple-adjusted model adjusted for age (in years), national, educational attainment, AST(U/L) and ALT(U/L). [b]Multiple-adjusted model adjusted for age (in years), national, educational attainment, menopausal, AST(U/L) and ALT(U/L). P-values < 0.05 are bold.

TABLE 3 | Association between Gal-3BP level and MetS in female subjects.

	Level of GAL-3BP		*P* value
	Lower	Upper	
Premenopausal (n=63)			
Age-adjusted model	1 (Reference)	1.09 (0.19, 6.28)	0.927
Multiple-adjusted model	1 (Reference)	1.24 (0.17, 9.10)	0.833
Postmenopausal (n=328)			
Age-adjusted model	1 (Reference)	**2.42 (1.39, 4.22)**	**0.002**
Multiple-adjusted model	1 (Reference)	**2.30 (1.31, 4.05)**	**0.004**

OR, Odds ratio; CI, confidence interval; Age-adjusted model, adjusted for age (in years); Multiple-adjusted model, additional adjusted for national, educational attainment, AST(U/L) and ALT(U/L). P-values < 0.05 are bold.

underlies MetS. Moreover, we newly identified adipose tissue macrophages as a source of Gal3-BP under conditions of adipose tissue expansion.

In this study, we were able to demonstrate the usefulness of the serum GAL-3BP levels in reflecting the incidence of MetS. Associations between serum GAL-3BP, abdominal obesity and lipoprotein levels have been shown previously (9–11), and we also identified associations between GAL-3BP level, center obesity and high TG, while further demonstrating the positive association between GAL-3BP and MetS. In a previous report, GAL-3BP, a large oligomeric glycoprotein, was suggested to be the galectin-3 ligand (23). Previous studies have indicated that the level of galectin-3 is associated with visceral fat, lipoprotein levels, glucose homeostasis, and even the presence of MetS (24, 25). Therefore, GAL-3BP may affect the distribution of body fat, gluconeogenesis, hyperglycemia, and lipolysis, which may result in the reduced ability to maintain metabolic homeostasis.

Logistic regression analyses suggested that GAL-3BP is positively correlated with MetS. The sex difference may be due to the sexual dimorphisms in adipose tissue biology, including adipose distribution and function (26–28). Cai et al. reported GAL-3BP increase in NAFLD patients between three groups (PostM-NAFLD vs. PostM-Control, PreM-NAFLD vs. PreM-Control, and PostM-NAFLD vs. PreM-NAFLD). They hypothesized GAL-3BP may connect to NALFD and metabolic disorders (7). Our results also report the association between GAL-3BP and MetS in postmenopausal women. Subanalyses of females further suggested the presence of a robust association between GAL-3BP and MetS in postmenopausal females. Although the mechanism underlying menopausal status and an association between serum GAL-3BP and MetS is unclear, several studies have suggested that biological changes after menopause may lead to the reduced ability of adipose tissue to expand, leading to additional fat storage (27, 28). The change in adipose tissue expandability may underlie the significant association between serum GAL-3BP and MetS in postmenopausal women. In addition, GAL-3BP was shown to interact with Complement Factor D, Insulin like Growth Factor 1, and Albumin directly, and to network with Estrogen Receptor 1 (ESR1), Nitric Oxide Synthase 3 and INS (7). Several studies have suggested that estrogen is associated with MetS and its related factors in postmenopausal women (29, 30). In premenopausal women, intact estrogen dependency might be preserved in the myometrium, as well as in the uterine endometrium with characteristic stable expression of ESR1 with ESR2, whereas, in postmenopausal women with much lower estrogen levels, ESR1 is decreased (31). An imbalance between ESR1 in the adipose tissue could therefore affect the development of metabolic diseases (32). Hormones have critical functions in MetS pathogenesis and progression, and estrogens have critical functions in lipoprotein metabolism. Reduced estrogen in postmenopausal women may enhance the association between the Gal-3BP and MetS.

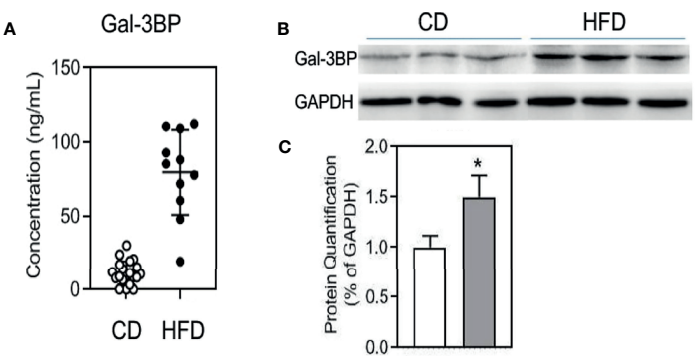


FIGURE 1 | GAL-3BP demonstrated positive correlations with MetS. **(A)** Serum GAL-3BP concentration. **(B)** Protein expressions of GAL-3BP and internal control GAPDH in Visceral Adipose Tissues. **(C)** Quantification of proteins with normalization to protein levels of GAPDH. Data are represented as mean ± SEM. n = 11-20, *$p < 0.05$ *vs* CD mice.

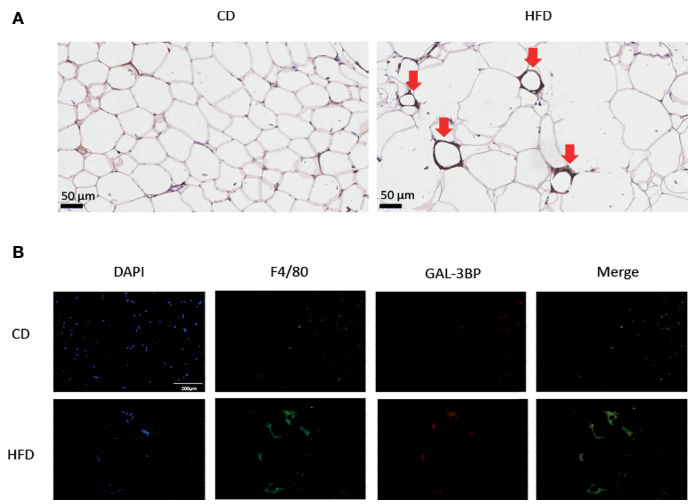


FIGURE 2 | The expression of GAL-3BP in the Visceral Adipose Tissues. **(A)** Representative images of F4/80 staining in VAT after HFD or CD exposure. 100 × **(B)** Representative images of immunofluorescent staining for F4/80 and GAL-3BP adducts. 100 ×. n = 5.

The association between GAL-3BP and ESR1 may thus result in postmenopausal MetS.

GAL-3BP was shown to be highly expressed in the VAT, which is consistent with previous findings (10, 11). Roelofsen et al. reported that GAL-3BP was secreted from VAT (13). We also found that serum GAL-3BP was highly expressed in the VAT of MetS mice. Typically, the increase in visceral fat has been verified to further boost insulin resistance, while MetS is probably induced by insulin resistance caused by the association between GAL-3BP levels and visceral adiposity. Moreover, recent data from reconstituted proteins *in vitro* have confirmed the association between GAL-3BP and adiponectin. GAL-3BP is a novel serum adiponectin binding protein and may abrogate the anti-inflammatory effects of adiponectin (9, 11). Hypoadiponectinemia is closely associated with hypertension, dyslipidemia, diabetes mellitus, and visceral fat obesity related to MetS (33). The present study showed that GAL-3BP was highly expressed in VAT macrophages. Inflammation may also account for these associations. Previous data have indicated that Gal-3BP has immunosuppressive as well as immunostimulatory functions *in vitro* (1); the glycoprotein has been implicated in inflammatory distress, immune response (7), and chronic low-grade inflammation (9). GAL-3BP is significantly positively associated with inflammatory markers, including IL6, IL-1β, together with TNFα (11, 34, 35), and these may participate in MetS pathogenesis related to GAL-3BP, since inflammation may probably result in insulin resistance. Furthermore, Gleissner et al. reported that GAL-3BP induces a pro-inflammatory transcriptome in human monocyte-derived macrophages (11). This is in line with our finding that GAL-3BP was highly expressed in macrophages in adipose tissues. Simultaneously, several studies have reported that GAL-3BP is a new biomarker for predicting chronic pancreatitis, non-alcoholic steatohepatitis (NASH), and NAFLD (7, 34, 36). Elevated GAL-3BP in patients with MetS is consistent with the presence of chronic low-grade inflammation as a key characteristic of MetS, pancreatitis and NASH (9, 34). We may compare GAL-3BP with inflammatory parameters in future.

Certain limitations to this study should be noted. First, this was a cross-sectional study to determine the significance of serum GAL-3BP as a biological marker of MetS. The cross-sectional design limited the usefulness of evaluating the serum GAL-3BP level as a biomarker in predicting the progression of MetS. A prospective study based on baseline stratified serum GAL-3BP levels may be necessary. Second, our study does not clearly demonstrate a mechanism underlying the association between GAL-3BP and MetS in participants. The murine model did not fully explain the source of Gal3-BP under adipose tissue expansion conditions in human subjects. We did not obtain adipose tissues from participants, who were community residents. We may delve into the mechanisms and try to link the findings in the future. Third, we did not include a perimenopausal group with oligomenorrea and perimenopausal symptoms. Participants going through menopausal process (perimenopausal women) were probably self-classified as menopausal. As menopause is a gradual process, the perimenopausal period should be included in future studies. Finally, we did not assess other adipokine (adiponectin or leptin, etc.) or other biomarkers levels, it is difficult to compare the GAL-3BP with other biomarkers. Therefore, we may compare GAL-3BP with adiponectins and other biomarkers in terms of the receiver operating characteristics for MetS in future.

Our results suggest a significant role for GAL-3BP in reflecting the complex phenotypes of MetS. In conclusion, the results from this study demonstrate that GAL-3BP levels show positive associations with MetS. This finding is particularly important because of the increasing risk of MetS seen in the Chinese population. Understanding the role of GAL-3BP in altering MetS could assist the development of diagnostic tools and treatments for obesity-related metabolic disorders. Nonetheless, further studies

should be carried out to clarify the role of GAL-3BP as a biomarker for MetS.

DATA AVAILABILITY STATEMENT

Data are available upon reasonable request. Requests to access the datasets should be directed to DW, dlwen@cmu.edu.cn.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by China Medical University. The patients/participants provided their written informed consent to participate in this study. The animal study was reviewed and approved by China Medical University.

AUTHOR CONTRIBUTIONS

YM and DW conceived and designed the study. SZ, RC, and XY collected, managed, and analyzed the data. SZ drafted the manuscript. All authors contributed to the article and approved the submitted version.

FUNDING

This research was supported by the National Key R&D Program of China (Grant #2018YFC1311600) and Liaoning Revitalization Talents Program (Grant #XLYC1808036).

ACKNOWLEDGMENTS

We would like to thank the participants in the study.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2021.726154/full#supplementary-material

REFERENCES

1. Loimaranta V, Hepojoki J, Laaksoaho O, Pulliainen AT. Galectin-3-Binding Protein: A Multitask Glycoprotein With Innate Immunity Functions in Viral and Bacterial Infections. *J Leukoc Biol* (2018) 104:777–86. doi: 10.1002/JLB.3VMR0118-036R
2. Stampolidis P, Ullrich A, Iacobelli S. LGALS3BP, Lectin Galactoside-Binding Soluble 3 Binding Protein, Promotes Oncogenic Cellular Events Impeded by Antibody Intervention. *Oncogene* (2015) 34:39–52. doi: 10.1038/onc.2013.548
3. Yamaguchi R, Hosaka M, Torii S, Hou N, Saito N, Yoshimoto Y, et al. Cyclophilin C-Associated Protein Regulation of Phagocytic Functions *via* NFAT Activation in Macrophages. *Brain Res* (2011) 1397:55–65. doi: 10.1016/j.brainres.2011.03.036
4. Ullrich A, Sures I, D'Egidio M, Jallal B, Powell TJ, Herbst R, et al. The Secreted Tumor-Associated Antigen 90K Is a Potent Immune Stimulator. *J Biol Chem* (1994) 269:18401–7. doi: 10.1016/S0021-9258(17)32322-0
5. Nonaka M, Ma BY, Imaeda H, Kawabe K, Kawasaki N, Hodohara K, et al. Dendritic Cell-Specific Intercellular Adhesion Molecule 3-Grabbing non-Integrin (DC-SIGN) Recognizes a Novel Ligand, Mac-2-Binding Protein, Characteristically Expressed on Human Colorectal Carcinomas. *J Biol Chem* (2011) 286:22403–13. doi: 10.1074/jbc.M110.215301
6. Jalkanen K, Leu T, Bono P, Salmi M, Jalkanen S, Smith DJ. Distinct Ligand Binding Properties of Mac-2-Binding Protein and Mouse Cyclophilin [Correction of Mousephilin] C-Associated Protein. *Eur J Immunol* (2001) 31:3075–84. doi: 10.1002/1521-4141(2001010)31:10<3075::AID-IMMU3075>3.0.CO;2-D
7. Cai H, Lu S, Chen Y, Das Mbbs Mrcog S, Niu Z, Zhuo G, et al. Serum Retinol Binding Protein 4 and Galectin-3 Binding Protein as Novel Markers for Postmenopausal Nonalcoholic Fatty Liver Disease. *Clin Biochem* (2018) 56:95–101. doi: 10.1016/j.clinbiochem.2018.04.017
8. Chen Y, Das S, Zhuo G, Cai H. Elevated Serum Levels of Galectin-3 Binding Protein Are Associated With Insulin Resistance in non-Diabetic Women After Menopause. *Taiwan J Obstet Gynecol* (2020) 59:877–81. doi: 10.1016/j.tjog.2020.09.014
9. Niinaga R, Yamamoto H, Yoshii M, Uekita H, Yamane N, Kochi I, et al. Marked Elevation of Serum M2BP-Adiponectin Complex in Men With Coronary Artery Disease. *Atherosclerosis* (2016) 253:70–4. doi: 10.1016/j.atherosclerosis.2016.08.024
10. Sugiura T, Dohi Y, Takase H, Yamashita S, Murai S, Tsuzuki Y, et al. Serum Levels of Mac-2 Binding Protein Increase With Cardiovascular Risk and Reflect Silent Atherosclerosis. *Atherosclerosis* (2016) 251:192–6. doi: 10.1016/j.atherosclerosis.2016.06.027
11. Gleissner CA, Erbel C, Linden F, Domschke G, Akhavanpoor M, Helmes CM, et al. Galectin-3 Binding Protein, Coronary Artery Disease and Cardiovascular Mortality: Insights From the LURIC Study. *Atherosclerosis* (2017) 260:121–9. doi: 10.1016/j.atherosclerosis.2017.03.031
12. Melin EO, Dereke J, Hillman M. Female Sex, High Soluble CD163, and Low HDL-Cholesterol Were Associated With High Galectin-3 Binding Protein in Type 1 Diabetes. *Biol Sex Differ* (2019) 10:51. doi: 10.1186/s13293-019-0268-0
13. Roelofsen H, Dijkstra M, Weening D, de Vries MP, Hoek A, Vonk RJ. Comparison of Isotope-Labeled Amino Acid Incorporation Rates (CILAIR) Provides a Quantitative Method to Study Tissue Secretomes. *Mol Cell Proteomics* (2009) 8:316–24. doi: 10.1074/mcp.M800254-MCP200
14. Iwata A, Kamada Y, Ebisutani Y, Yamamoto A, Ueda Y, Arai H, et al. Establishment of Mouse Mac-2 Binding Protein Enzyme-Linked Immunosorbent Assay and its Application for Mouse Chronic Liver Disease Models. *Hepatol Res* (2017) 47:902–9. doi: 10.1111/hepr.12819
15. Moore JX, Chaudhary N, Akinyemiju T. Metabolic Syndrome Prevalence by Race/Ethnicity and Sex in the United States, National Health and Nutrition Examination Survey, 1988-2012. *Prev Chronic Dis* (2017) 14:E24. doi: 10.5888/pcd14.160287
16. Kassi E, Pervanidou P, Kaltsas G, Chrousos G. Metabolic Syndrome: Definitions and Controversies. *BMC Med* (2011) 9:48. doi: 10.1186/1741-7015-9-48
17. Jiang B, Li B, Wang Y, Han B, Wang N, Li Q, et al. The Nine-Year Changes of the Incidence and Characteristics of Metabolic Syndrome in China: Longitudinal Comparisons of the Two Cross-Sectional Surveys in a Newly Formed Urban Community. *Cardiovasc Diabetol* (2016) 15:84. doi: 10.1186/s12933-016-0402-9
18. Wannamethee SG, Shaper AG, Lennon L, Morris RW. Metabolic Syndrome vs Framingham Risk Score for Prediction of Coronary Heart Disease, Stroke, and Type 2 Diabetes Mellitus. *Arch Internal Med* (2005) 165:2644–50. doi: 10.1001/archinte.165.22.2644
19. Ford ES. Risks for All-Cause Mortality, Cardiovascular Disease, and Diabetes Associated With the Metabolic Syndrome: A Summary of the Evidence. *Diabetes Care* (2005) 28:1769–78. doi: 10.2337/diacare.28.7.1769

20. Saely CH, Koch L, Schmid F, Marte T, Aczel S, Langer P, et al. Adult Treatment Panel III 2001 But Not International Diabetes Federation 2005 Criteria of the Metabolic Syndrome Predict Clinical Cardiovascular Events in Subjects Who Underwent Coronary Angiography. *Diabetes Care* (2006) 29:901–7. doi: 10.2337/diacare.29.04.06.dc05-2011
21. Micheli L, Lucarini E, Trallori E, Avagliano C, De Caro C, Russo R, et al. Extract: Alpha-Amylase Inhibition Against Metabolic Syndrome in Mice. *Nutrients* (2019) 11. doi: 10.3390/nu11081778
22. Greer KB, Falk GW, Bednarchik B, Li L, Chak A. Associations of Serum Adiponectin and Leptin With Barrett's Esophagus. *Clin Gastroenterol Hepatol* (2015) 13:2265–72. doi: 10.1016/j.cgh.2015.02.037
23. Grassadonia A, Tinari N, Iurisci I, Piccolo E, Cumashi A, Innominato P, et al. 90k (Mac-2 BP) and Galectins in Tumor Progression and Metastasis. *Glycoconj J* (2002) 19:551–6. doi: 10.1023/B:GLYC.0000014085.00706.d4
24. Nayor M, Wang N, Larson MG, Vasan RS, Levy D, Ho JE. Circulating Galectin-3 Is Associated With Cardiometabolic Disease in the Community. *J Am Heart Assoc* (2015) 5. doi: 10.1161/JAHA.115.002347
25. Pugliese G, Iacobini C, Pesce CM, Menini S. Galectin-3: An Emerging All-Out Player in Metabolic Disorders and Their Complications. *Glycobiology* (2015) 25:136–50. doi: 10.1093/glycob/cwu111
26. Tan CY, Vidal-Puig A. Adipose Tissue Expandability: The Metabolic Problems of Obesity may Arise From the Inability to Become More Obese. *Biochem Soc Trans* (2008) 36:935–40. doi: 10.1042/BST0360935
27. Gray SL, Vidal-Puig AJ. Adipose Tissue Expandability in the Maintenance of Metabolic Homeostasis. *Nutr Rev* (2007) 65:S7–12. doi: 10.1301/nr.2007.jun.S7-S12
28. Matsuo Y, Tanaka M, Yamakage H, Sasaki Y, Muranaka K, Hata H, et al. Thrombospondin 1 as a Novel Biological Marker of Obesity and Metabolic Syndrome. *Metab Clin Exp* (2015) 64:1490–9. doi: 10.1016/j.metabol.2015.07.016
29. Zhao L, Fan X, Zuo L, Guo Q, Su X, Xi G, et al. Estrogen Receptor 1 Gene Polymorphisms Are Associated With Metabolic Syndrome in Postmenopausal Women in China. *BMC Endocr Disord* (2018) 18:65. doi: 10.1186/s12902-018-0289-4
30. Lo JC, Zhao X, Scuteri A, Brockwell S, Sowers MR. The Association of Genetic Polymorphisms in Sex Hormone Biosynthesis and Action With Insulin Sensitivity and Diabetes Mellitus in Women at Midlife. *Am J Med* (2006) 119:S69–78. doi: 10.1016/j.amjmed.2006.07.009
31. Sakaguchi H, Fujimoto J, Aoki I, Tamaya T. Expression of Estrogen Receptor Alpha and Beta in Myometrium of Premenopausal and Postmenopausal Women. *Steroids* (2003) 68:11–9. doi: 10.1016/S0039-128X(02)00111-3
32. Gallagher CJ, Langefeld CD, Gordon CJ, Campbell JK, Mychaleckyj JC, Bryer-Ash M, et al. Association of the Estrogen Receptor-Alpha Gene With the Metabolic Syndrome and its Component Traits in African-American Families: The Insulin Resistance Atherosclerosis Family Study. *Diabetes* (2007) 56:2135–41. doi: 10.2337/db06-1017
33. Matsuzawa Y, Funahashi T, Kihara S, Shimomura I. Adiponectin and Metabolic Syndrome. *Arterioscler Thromb Vasc Biol* (2004) 24:29–33. doi: 10.1161/01.ATV.0000099786.99623.EF
34. Maekawa T, Kamada Y, Ebisutani Y, Ueda M, Hata T, Kawamoto K, et al. Serum Mac-2 Binding Protein Is a Novel Biomarker for Chronic Pancreatitis. *World J Gastroenterol* (2016) 22:4403–10. doi: 10.3748/wjg.v22.i17.4403
35. Gagno G, Padoan L, Stenner E, Beleù A, Ziberna F, Hiche C, et al. Galectin 3 and Galectin 3 Binding Protein Improve the Risk Stratification After Myocardial Infarction. *J Clin Med* (2019) 8. doi: 10.3390/jcm8050570
36. Kamada Y, Fujii H, Fujii H, Sawai Y, Doi Y, Uozumi N, et al. Serum Mac-2 Binding Protein Levels as a Novel Diagnostic Biomarker for Prediction of Disease Severity and Nonalcoholic Steatohepatitis. *Proteomics Clin Appl* (2013) 7:648–56. doi: 10.1002/prca.201200137

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 11 November 2021
doi: 10.3389/fendo.2021.774309

Circulating CTRP7 Is a Potential Predictor for Metabolic Syndrome

Wenjing Hu[1†], Bin Zhan[2†], Qinge Li[1], Gangyi Yang[3], Mengliu Yang[3], Minghong Tan[3], Shan Geng[2], Hua Liu[4], Chen Chen[5], Dongfang Liu[3] and Ling Li[1]*

[1] Key Laboratory of Diagnostic Medicine (Ministry of Education) and Department of Clinical Biochemistry, College of Laboratory Medicine, Chongqing Medical University, Chongqing, China, [2] Department of Endocrinology, The Thirteenth People's Hospital of Chongqing, Chongqing, China, [3] Department of Endocrinology, The Second Affiliated Hospital, Chongqing Medical University, Chongqing, China, [4] Department of Pediatrics, University of Mississippi Medical Center, Jackson, MS, United States, [5] Endocrinology, School of Biomedical Science (SBMS), Faculty of Medicine, University of Queensland, Brisbane, QLD, Australia

OPEN ACCESS

Edited by:
Patricia Khashayar, Ghent University, Belgium

Reviewed by:
Ying Zhao, Zhejiang University, China
Djordje S. Popovic, University of Novi Sad, Serbia
Aleksandra Klisic, Primary Health Care Center Podgorica, Montenegro

***Correspondence:**
Ling Li
liling@cqmu.edu.cn

[†]These authors have contributed equally to this work and share first authorship

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *11 September 2021*
Accepted: *18 October 2021*
Published: *11 November 2021*

Citation:
Hu W, Zhan B, Li Q, Yang G, Yang M, Tan M, Geng S, Liu H, Chen C, Liu D and Li L (2021) Circulating CTRP7 Is a Potential Predictor for Metabolic Syndrome. Front. Endocrinol. 12:774309. doi: 10.3389/fendo.2021.774309

Background: Previous animal studies have revealed that CTRP7 is related to energy metabolism. However, little is known regarding the relationship between CTRP7 and metabolic diseases in humans. Hence, this study was designed to explore the association between CTRP7 and MetS through a cross-sectional study and multiple intervention studies.

Methods: A total of 624 individuals were enrolled in this study. The levels of CTRP7 and APN were determined by ELISA kit. HEC, OGTT and lipid infusion were performed in heathy individuals to investigate the association of CTRP7 and glucose, insulin and FFA. Bioinformatics analysis was then undertaken to identify genes and signaling pathways associated with CTRP7. The relationship between CTRP7 with MetS components was also evaluated.

Results: In MetS patients, serum CTRP7 concentrations were significantly higher than in healthy controls, and was positively correlated with WC, BP, FBG, 2h-BG and TG, but negatively correlated with HDL-C and APN. Multivariate logistic regression analysis uncovered that CTRP7 was strongly correlated with the occurrence of MetS. In addition, circulating levels of CTRP7 in patients with two or more MetS components were higher than those with one MetS component. In the intervention studies, OGTTs resulted in a significant reduction in serum CTRP7 concentration. However, the increase in insulin levels caused by EHC and the increase of FFA caused by lipid-infusion led to the significant increase of serum CTRP7 concentration. Meanwhile, bioinformatics analysis revealed that CTRP7 was strongly associated with metabolism-related genes and signal pathways, which further illustrate the association of CTRP7 with whole-body metabolism.

Conclusions: Serum CTRP7 is increased in MetS patients, which may be a biomarker related to metabolic diseases.

Clinical Trial Registration Number: ChiCTR2000032878.

Keywords: CTRP7, MetS, insulin resistance, interventional tests, Bioinformatics

INTRODUCTION

Metabolic syndrome (MetS), also known as insulin resistance (IR) syndrome, was first described in 1988. It represents an aggregation of cardiovascular risk factors. At present, IR, obesity, impaired glucose tolerance (IGT), dyslipidemia, hypertension, and chronic low-grade inflammation are all considered characteristics of MetS (1–4).

MetS classification has important clinical significance and application value for screening metabolic diseases related to obesity (5). Because characteristics of MetS include chronic low-grade inflammation and IR, it is important to study the biomarkers for prevention and diagnosis as well as finding new drug targets. Recently, we and others have found that members of the CTRP family, such as c1q/tnf related protein subtype 6 (CTRP6), CTRP5, and CTRP15, are associated with the occurrence of MetS, type 2 diabetes mellitus (T2DM), and obesity (6–12). Therefore, the CTRP family may be an important biomarker of metabolic diseases in humans.

The biological function of CTRP7, a newly discovered member of the CTRP family, has been elusive since it was first identified (13). Previous animal studies found that CTRP7 mRNA expression in the fat of ob/ob diabetic mice was increased at eight-week-old (14). At the age of 12 weeks, blood glucose in ob/ob mice decreased due to a compensatory increase of insulin secretion. With decreased blood glucose, the expression of CTRP7 returned to the level of the control group (15). These findings suggest that CTRP7 may be involved in glucose metabolism *in vivo*. Another study reported that CTRP7 expression was upregulated in skeletal muscle in aged rats and down-regulated in caloric restricted animals. Therefore, it is further revealed that CTRP7 is related to energy metabolism (16). However, there are few studies on the relationship between CTRP7 and metabolic diseases in humans and animals, especially in MetS patients.

In this study, we examined serum CTRP7 and adiponectin (APN), an insulin sensitizer, levels in newly diagnosed MetS patients and healthy adults, and explored the association between CTRP7 and IR, APN, and metabolic parameters.

MATERIALS AND METHODS

Study Populations

A cohort of 624 people participated in the study, including 310 males and 314 females aged 21-82 years. These individuals included 328 newly diagnosed patients with MetS, and 296 age-matched normal adults. MetS patients come from outpatients and inpatients in the Department of Endocrinology and Metabolism. MetS was diagnosed according to the criteria of the Chinese Diabetes Association (CDS guidelines 2017) (17). Individuals who meet three or more of the following conditions were considered for the diagnosis of MetS: 1) Central obesity, waist circumference (WC) ≥ 90 cm for men or ≥85 for women; 2) Triglyceride (TG) ≥ 1.7 mmol/L; 3) High-density lipoprotein-cholesterol (HDL-C) < 1.04 mmol/L); 4) Blood pressure (BP) ≥ 130/85 mmHg or receiving antihypertensive drugs; 5) Fasting blood glucose (FBG) ≥ 5.6 mmol/L or 2-hour blood glucose (2h-BG) ≥ 7.8 mmol/L or T2DM. Exclusion criteria include patients with liver cirrhosis, heart, liver, or renal failure, steroid use, various malignant tumors, infection, or other diseases. Individuals who adhered to daily exercise, smoking and alcohol dependence were also excluded from this study.

Healthy participants were selected through advertising, routine physical examination, or from the community. The diagnosis of impaired glucose tolerance (IGT) and T2DM is based on WHO standards in 1998 (18). In our cohort, MetS patients were newly diagnosed and did not use drugs or lifestyle interventions. The healthy controls had normal blood glucose, no family history of T2DM and hypertension, no clinical evidence of disease, and no medication. This study was approved by the Human Research Ethics Committee of Chongqing Medical University and was registered at chictr.org (ChiCTR2000032878). In the current study, subjects who met the inclusion criteria were randomly assigned numbers, and individuals with or without Mets were selected randomly according to the number to eliminate the selection bias. all subjects signed informed consent. The study was performed in accordance with the Helsinki Declaration.

Anthropometric and Biochemical Measurements

Weight and height were measured using standardized equipment, and participants wore light indoor clothing without shoes. Body mass index (BMI) was calculated as weight divided by height squared (kg/m^2). Waist circumference (WC) was measured by the lower border of the ribs and the iliac crest. The waist-to-hip ratio (WHR) was calculated by WC and hip circumstance (HC). Blood pressure was measured with a mercury sphygmomanometer. Right arm blood pressure was measured three times in resting and sitting posture. The second and third average readings of blood pressure were taken for calculating. Body adiposity index (BAI) was calculated as [HC (cm)/(height (m))$^{1.5}$ – 18] (19). Visceral adiposity index (VAI) $_{Females}$= WC/[36.58 + (1.89 × BMI)] × (TG/0.81) × (1.52/HDL) or Visceral adiposity index (VAI) $_{Males}$= WC/[39.68 + (1.88× BMI)] × (TG/1.03) × (1.31/HDL) (20). After an overnight fasting, blood samples were collected, refrigerated, and serum samples were transported to the central laboratory for biochemical measurements within 12 h. Blood glucose and HbA1c were measured using a glucose oxidase method and HPLC, respectively. Insulin, free fatty acids (FFAs), and blood lipid were measured using an autoanalyzer (Hitachi 747; Hitachi, Tokyo, Japan) as described previously (21). Homeostasis model assessment of IR (HOMA-IR) was calculated using the following equations: HOMA-IR = fasting insulin (FIns, mU/L) × fasting blood glucose (FBG, mmol/L)/22.5 (22).

Abbreviations: MetS, metabolic syndrome; APN, adiponectin; OGTT, oral glucose tolerance test; EHC, euglycemic-hyperinsulinemic clamps; FFA, free fatty acid; WC, waist circumference; BP, blood pressure; FBG, fasting blood glucose; 2h-BG, 2-hour post–glucose load blood glucose; TG, triglyceride; HDL-C, high-density lipoprotein cholesterol.

Interventional Tests

Oral Glucose Tolerance Test

An OGTT was engaged in all populations, and blood for CTRP7, glucose, and insulin measurements were drawn at indicated times as reported previously (22).

Hyperinsulinemic-Euglycemic Clamps

HECs were engaged on 32 healthy subjects (15 male; age 25.1± 2.3 years; BMI: 22.5 ± 2.7kg/m^2) as previously reported (23). During HEC, regular insulin (1 mU/kg/min) was infused for 2 h, and a variable infusion of 20% glucose was administered to maintain blood glucose at baseline concentration. M-values were determined as the glucose infusion rate (GIR) during the stable period of the HEC and were related to body weight. Blood samples for CTRP7 and insulin measurement were obtained at indicated times (0, 80, 100,110 and 120 min). Serum samples were stored at -80°C for further analysis.

Lipid Infusion Study

32 healthy subjects were given a lipid infusion (20% Intralipid, 1.5 ml/min) for 4-hour. Blood samples were collected at different time points as previously published (22).

Cytokine Measurements

Circulating CTRP7 levels were determined with an ELISA kit (sk00396-09, Aviscerabio science Inc., MA. USA) according to the manufacturer's protocol. The detection limit of serum CTRP7 levels was 5-320 μg/L, and the intra- and inter-assay coefficients of variation (CV) were less than 5% and 10%, respectively. The ELISA kit has high sensitivity, good specificity for human CTRP7 detection without obvious cross-reaction, and interference. Circulating APN levels were also measured with an ELISA Kit following the manufacturer's protocol (Aviscera Bioscience, sk00010-02). Intra- and inter-assay CV were 8% and 10%, respectively.

Bioinformatics Analysis

A protein-protein interaction (PPI) network of CTRP7 gene was established by using the Database Search tool (version 11.0). An interaction score of 0.4 was considered a cut-off criterion, and the PPI was visualized. The cluster Profiler package was used for Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analyses (24). REACTOME enrichment analysis was completed by a STRING database (25). The list of annotated terms was obtained by GO, KEGG, and REACTION analysis. *P*-value < 0.05 was considered a statistical significance in GO, KEGG terms, and REACTOME analysis.

Statistical Analysis

All statistical analyses were performed using SPSS, version 20.0. All data were Mean ± SE or median of the interquartile range. The distribution of data was examined by Kolmogorov- Smirnov test. The differences between the two groups were compared by *t*-test or Mann-Whitney U test. The association between CTRP7 and APN as well as metabolic parameters was determined using correlation analysis. Multivariate regression analyses were performed to investigate the associations between variables. A Cochran-Armitage trend test was performed to analyze the tendency of serum CTRP7 levels associated with MetS. We used receiver operating characteristics (ROC) curve analysis to determine the cut-off point of CTRP7 for predicting MetS. In the OGTT, the area under the glucose curve (AUCg) was determined according to the trapezoidal rule. In statistical analyses, $p < 0.05$ was considered significant.

RESULTS

Serum CTRP7 Concentration Is Higher in Individuals With MetS or IR

Table 1 showed the main clinical and biochemical indicators in MetS patients and healthy controls. As expected, MetS patients have higher blood pressure (BP), obesity-related indicators (WC and BMI), glucose metabolism-related parameters [FBG, 2-h blood glucose after glucose overload (2h-BG), HbA1c and AUCg], triglyceride (TG), total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), FFA, FIns, 2-h serum insulin after glucose overload (2h-Ins), visceral adiposity index (VAI), and body adiposity index (BAI) and HOMA-IR than those of normal controls. However, high-density lipoprotein cholesterol (HDL-C) and APN levels were lower in MetS patients.

As shown in **Figure 1A**, the distribution of circulating CTRP7 levels in healthy subjects ranged from 52.4 to 378.1μg/L, and 90% of the healthy population was between 76.0 μg/L to 231.6 μg/L. Importantly, circulating CTRP7 levels were significantly higher in MetS patients than those in control subjects. After adjusting the gender and age of the study populations, the levels of CTRP7 in patients with MetS were still markedly higher than those in the control group (**Figure 1B** and **Table 1**). In contrast, serum APN concentrations, an insulin sensor, were lower in MetS patients compare with those in controls (**Table 1** and **Figure 1B**). There were no difference in serum CTRP7 levels between males and females [159.0 (117.4-209.2) *vs.* 150.0 (119.1-192.8)μg/L].

To investigate the relationship between CTRP7 and IR, study populations were divided into IR and non-IR according to HOMA-IR > 3 or ≤ 3 (24). The results showed that IR population had higher serum CTRP7 levels [195.4 (163.3-244.7) *vs.*130.6 (103.1-173.6) μg/L; $p < 0.01$, **Figure 1C**] and lower APN levels [5.19 (2.90-8.76) *vs.* 8.07 (4.67-1.39) mg/L, $p < 0.01$) compared with non-IR population (**Figure 1C**).

Relationship Between Serum CTRP7 and Other Indexes

Next, we engaged a linear correlation analysis. The results showed that CTRP7 was positively correlated with WC ($r = 0.36, p < 0.01$), SBP ($r = 0.25, p < 0.01$),DBP ($r = 0.25, p < 0.01$) FBG ($r = 0.65, p < 0.01$), 2h-BG ($r = 0.66, p < 0.01$), and TG ($r = 0.37, p < 0.01$), but negatively correlated with HDL-C ($r = -0.24, p < 0.01$) and APN ($r = -0.36, p < 0.01$; **Figure 1D**). In addition, multiple stepwise regression analysis uncovered that FBG, 2h-BG, WC, APN and DBP were independent impacted factors with serum CTRP7 concentration (**Figure 1E**). The multiple regression equation was: $Y_{\log\ (CTRP7)}=1.422 + 0.021X_{2h\text{-}BG} + 0.041X_{FBG} + 0.003\ X_{WC}\text{-}0.000023X_{APN} + 0.001\ X_{DBP}$.

TABLE 1 | Main clinical features and serum CTRP7 levels in MetS and control subjects.

Characteristics	Overall (n = 624)	MetS		*p*
		No (n = 296)	Yes (n = 328)	
Male	310	145	165	0.749
Age (yr)	53 (49-61)	52 (49-61)	54 (48-61)	0.217
WC (cm)	86.0 (80.0-91.0)	81.0 (77.0-86.0)	90.0 (85.0-94.0)	<0.001
BMI (Kg/m^2)	24.2 (22.6-26.7)	23.3 (22.0 -24.6)	25.7 (23.7-27.6)	<0.001
SBP (mmHg)	130 (120-143)	123 (115-133)	136 (127-152)	<0.001
DBP (mmHg)	81 (75-90)	78 (70-84)	86 (80-93)	<0.001
FBG (mmol/L)	6.15 (5.34-7.11)	5.45 (5.02-6.23)	6.70 (6.01-7.46)	<0.001
2h-BG (mmol/L)	9.30 (7.22-12.69)	7.45 (6.26-11.22)	12.09 (8.78-13.27)	<0.001
FIns (mU/L)	9.57 (8.26-12.06)	8.80 (7.93-9.74)	11.09 (9.00-13.15)	<0.001
2h-Ins (mU/L)	50.9 (34.1-74.4)	45.3 (31.8-61.1)	56.0 (37.0-85.2)	<0.001
TG (mmol/L)	1.82 (1.40-2.24)	1.48 (1.16-1.83)	2.07 (1.79-2.44)	<0.001
TC (mmol/L)	5.02 (4.53-5.36)	4.83 (4.46-5.36)	5.09 (4.64-5.36)	0.001
HDL-C (mmol/L)	1.23 (1.15-1.44)	1.34 (1.18-1.53)	1.20 (1.12-1.32)	<0.001
LDL-C (mmol/L)	2.85 (2.52-3.12)	2.71 (2.25-3.03)	2.94 (2.68-3.15)	<0.001
FFA (μmol/L)	0.60 (0.49-0.70)	0.54 (0.43-0.68)	0.64 (0.55-0.72)	<0.001
HbA1c (%)	6.3 (5.6-8.3)	5.8 (5.3-6.9)	7.7 (6.0-8.5)	<0.001
HOMA-IR	2.55 (1.99-3.65)	2.10 (1.83-2.62)	3.31 (2.35-4.23)	<0.001
AUC_g	21.1 (16.8-28.7)	17.3 (14.6-23.4)	26.6 (20.2-31.0)	<0.001
AUC_i	85.4 (63.7-114.2)	84.3 (66.7-101.2)	87.0 (59.0-131.9)	0.080
VAI	2.27 (1.58-2.97)	1.67 (1.16-2.34)	2.67 (2.17-3.47)	<0.001
BAI	28.9 (26.3-31.8)	27.9 (25.6-30.6)	29.9 (27.2-32.6)	<0.001
APN (mg/L)	6.81 (4.14-12.35)	8.46 (5.16-14.13)	5.66 (3.22-9.81)	<0.001
CTRP7 (μg/L)	155.1 (119.0-197.5)	127.5 (102.1-160.5)	188.7 (139.7-224.3)	<0.001
CTRP7 (adjusted)*	— —	137.4 ± 3.3	190.9 ± 3.2	<0.001

*Values are given as mean ± SE or median (Interquartile Range). MetS, Metabolic Syndrome; BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; FBG, fasting blood glucose; 2h-BG, 2-h blood glucose after glucose overload; FIns, fasting plasma insulin; 2h-Ins, 2-h serum insulin after glucose overload; TG, triglyceride; TC, total cholesterol; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; FFA, free fatty acid; HOMA-IR, homeostasis model assessment of insulin resistance; AUC_g, the area under the curve of glucose during oral glucose tolerance test; AUCi, the area under the curve of glucose during insulin tolerance test; VAI, visceral adiposity index; BAI, body adiposity index; APN, adiponectin; *Mean ± standard error by general linear model with adjustment of age and Sex.*

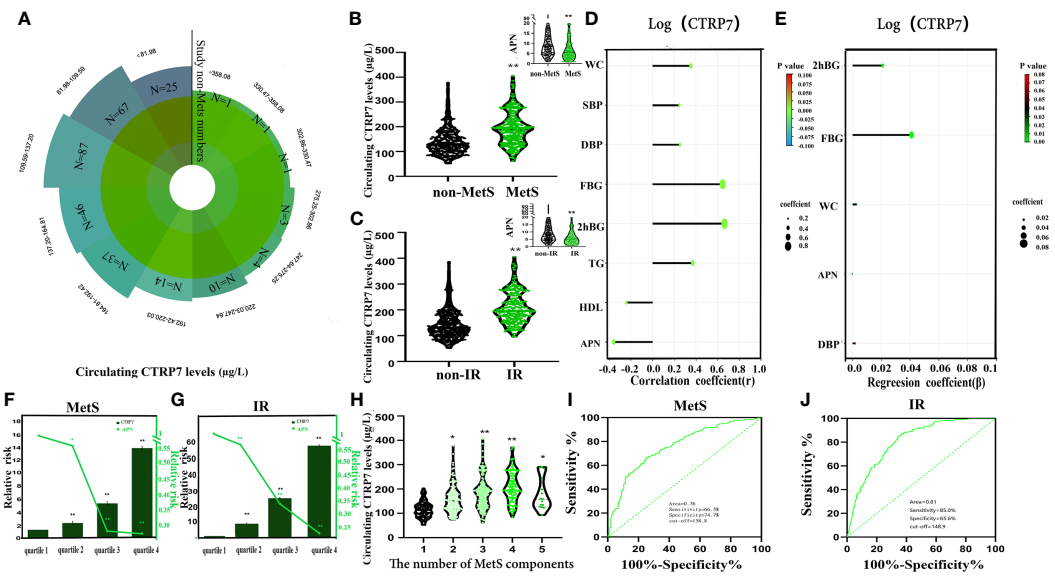


FIGURE 1 | Circulating CTRP7 level and its relationship with metabolic markers and Mets in the study population. **(A)** Distribution of serum CTRP7 in the healthy population. **(B)** Serum CTRP7 and APN levels in MetS and healthy subjects. **(C)** Serum CTRP7 levels, according to HOMA-IR (IR, HOMA-IR > 3; non-IR, HOMA-IR ≤ 3). **(D)** Simple correlation analysis of variables associated with circulating CTRP7. **(E)** Multiple regression analysis of variables associated with circulating CTRP7. **(F, G)** The odds ratio of having MetS **(F)** and IR **(G)** in different quartiles of serum CTRP7 and APN. **(H)** Circulating CTRP7 levels to the number of MetS components. **(I, J)** ROC curve analyses for the prediction of MetS **(I)** and IR **(J)** according to serum CTRP7 levels. MetS, metabolic syndrome; IR, insulin resistance. Data are expressed as mean ± SE or median (Interquartile Range). *$p < 0.05$, **$p < 0.01$ *vs.* non-MetS or non-IR. quartile 1 or number 1.

CTRP7 Is Related to the MetS and IR

Next, we engaged a multivariate logistic regression analysis to investigate the association of CTRP7 with MetS and IR. This analysis uncovered that circulating CTRP7 was markedly associated with MetS and IR, even if controlling for age and gender. However, BMI and lipids may contribute the most (**Table 2**). Using the row mean score difference and Cochran-Armitage test, we found that the increase of serum CTRP7 concentration showed a linear trend, and CTRP7 was independently related to MetS and IR (**Table S1**). In addition, we divided CTRP7 and APN concentration into four quartiles (quartile 1, <119.0μg/L; quartile 2, 119.0-155.0 μg/L; quartile 3,155.0-197.5μg/L; quartile 4, > 197.5μg/L for CTRP7 and tertile1, < 4.14mg/L; quartile 2, 4.14-6.81mg/L; quartile 3, 6.81-12.35 mg/L; quartile 4, >12.35 mg/L for APN). The relative risk of MetS and IR was calculated by logistic regression analysis. We found that the odds ratios for MetS was higher in quartiles 2, 3 and 4 of CTRP7 concentration than that of quartile 1 (95% CI 1.45-3.83 for quartile 2; 95% CI 3.32 - 8.88 for quartile 3 and 95% CI 8.15-24.33 for quartile 4 *vs.* quartile 1, all $p < 0.01$), while the odds ratio of MetS in the quartile 2, 3 and 4 of APN concentration was lower than that in quartile 1 (95% CI 0.34 - 0.88 for quartile 2; 95% CI 0.17 - 0.45 for quartile 3 and 95% CI 0.17- 0.44 for quartile 4; *vs.* quartile 1, $p < 0.01$ or 0.05) (**Figure 1F**). Additionally, the odds ratio of CTRP7 and APN concentration for predicting the development of IR was similar to that of MetS (95% CI 3.65 - 21.71 for quartile 2, 95% CI 10.42 - 59.92 for quartile 3, and 95% CI 23.93-140.45 for quartile 4 *vs*. quartile 1 for CTRP7; 95% CI 0.36-0.88 for quartile 2, 95% CI 0.21- 0.54 for quartile 3 and 95% CI 0.14-0.37 for quartile 4 *vs*. quartile 1 for APN, all $p <$ 001) (**Figure 1G**). When circulating CTRP7 levels were stratified by MetS components including BP, blood lipids, abdominal obesity, and FBG, the circulating CTRP7 levels in patients with two or more MetS components were higher than those with one MetS component (**Figure 1H**). Patients with 2, 3, 4, or more MetS components had CTRP7 concentration for 118.2 (96.5-142.6), 150.1 (117.4-184.3), 180.6 (133.2 -207.6), 195.9 (163.8 -257.4) and 158.1(126.5-245.2) μg/L, respectively. We further used the ROC curves of circulating CTRP7 to predict the occurrence of MetS and IR. The area under the ROC curves for MetS (AUC_{MetS}) and IR (AUC_{IR}) was 0.76 with 66.5% sensitivity and 74.7% specificity for MetS (**Figure 1I**) and 0.81 with 85% sensitivity and 65.6% specificity for IR (**Figure 1J**), respectively. The optimal cut-off points of CTRP7 for MetS and IR were 158.8 μg/L and 148.9μg/L, respectively.

Alternations of Serum CTRP7 Concentration in Different Intervention Studies

The different intervention study designs are shown in **Figure 2A**. To evaluate whether serum CTRP7 is affected by blood glucose and insulin, we first conducted an OGTT study in normal men and women. In response to OGTT induced increases in blood glucose and insulin levels, the serum CTRP7 levels were significantly reduced in these subjects (**Figure 2B**). There was no significant difference in serum CTRP7 concentration between normal men and women (**Figures 2B, C**).

To further identify the regulatory role of blood glucose or insulin on circulating CTRP7, HECs were performed in 15 young men and 17 young women (**Figures 2D, E**). During the HEC, the blood glucose was clamped at the basal level (~ 5mmol), and insulin levels were significantly elevated from 48.3 ± 13.2 to 348.3 ± 52.4 pmol/L, indicating hyperinsulinemia *in vivo*. Our EHC results showed that exogenous increased insulin led to an obvious increase in circulating CTRP7 levels (from 135.6 ± 14.0 to 180.9 ± 12.2μg/L for men; 129.8 ± 29.2 to 164.7 ± 25.0 μg/L for women).

To further explore the relationship between CTRP7 and IR, we performed a lipid infusion to increase serum FFA levels and induce an acute IR *in vivo* (**Figures 2F, G**). The lipid infusion-induced increases in serum FFA levels and significantly increased the levels of circulating CTRP7 [from 127.8 (117.4-133.1) to 163.4 (150.5-181.4) μg/L for men; 119.3 (108.1-129.8) to 165.1 (149.3-174.2) μg/L for women]. Therefore, we believe that FFA-induced IR promotes the release of CTRP7 *in vivo*.

Bioinformatics Analysis

To further explore the relationship between CTRP7 and metabolic disorders, we performed bioinformatics analysis using Internet big data. As shown in **Figure 3A**, a PPI network was constructed. Ten genes (proteins) were involved in this PPI network, including ZC3H10, PECR, CBLN3, PLTP, CLEC19A, TMEM69, FAM132A, CCDC137, CD36, and LAMB4. Among them, some genes were related to lipid metabolisms, such as CD36 and FAM132A (26–28). For GO analysis, we used $p < 0.05$ as the screening condition and arranged the results from a large degree to a small degree. GO analysis revealed that in biological processes, the top 10 proteins include the positive regulation of

TABLE 2 | Association of circulating CTRP7 with IR and MetS in fully adjusted models.

Model adjust	MetS			IR		
	OR	95% CI	*P*	OR	95% CI	*P*
Age	2.401	2.029-2.843	<0.001	3.194	2.621-3.893	<0.001
Age, Sex	2.410	2.035-2.855	<0.001	3.335	2.717-4.094	<0.001
Age, Sex, BP	2.206	1.841-2.643	<0.001	3.104	2.523-3.819	<0.001
Age, Sex, BP, BMI	1.880	1.549-2.282	<0.001	2.907	2.347-3.600	<0.001
Age, Sex, BP, BMI, WC	1.912	1.556-2.350	<0.001	2.875	2.319-3.564	<0.001
Age, Sex, BP, BMI, WC, Lipids	1.006	1.001-1.011	0.012	1.014	1.010-1.018	<0.001

Results of multivariate logistic regression analysis are presented as the odds ratio of being in IR and MetS status increase in serum CTRP7. CI, confidence interval; OR, odds ratio.

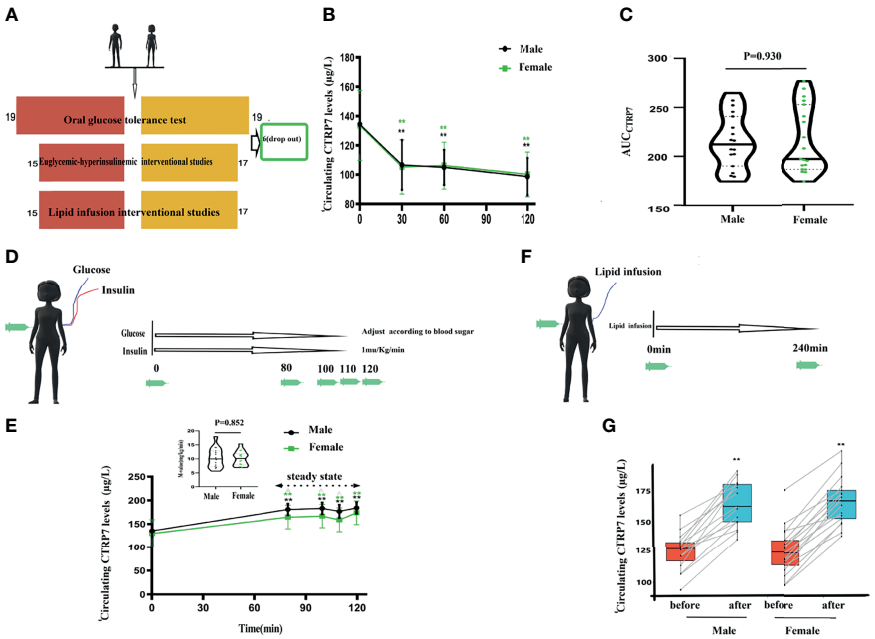


FIGURE 2 | Circulating CTRP7 levels in the interventional studies of healthy individuals. **(A)** Schematic diagram of intervention study design. **(B)** Circulating CTRP7 concentrations during the OGTT. **(C)** Area under the curve of CTRP7 during the OGTT (AUC_{CTRP7}). **(D)** Schematic diagram of the EHC. **(E)** Time course of serum CTRP7 alternations in healthy individuals during the EHC. **(F)** Schematic diagram of lipid infusion. **(G)** Time course of serum CTRP7 alternations in healthy subjects during lipid infusion. OGTT, oral glucose tolerance test; HEC, hyperinsulinemic-euglycemic clamp. Data are means ± SD. $^{**}p < 0.01$ *vs.* baseline, or female.

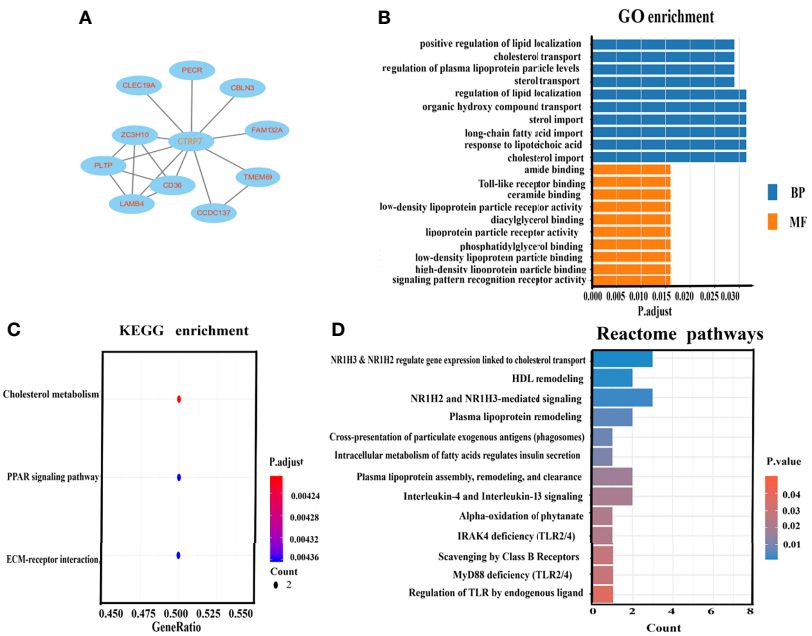


FIGURE 3 | Bioinformatic analysis for CTRP7-related genes and signaling pathways. **(A)** Protein-protein interaction (PPI) network. **(B)** GO enrichment analysis for biological process (BP) and molecular function (MF). **(C)** KEGG enrichment analysis of the pathways. The gradual color represents the P-value. The size of the bubble represents the gene number. **(D)** REACTOME analysis for enriched pathways. The X-axis represents the number of involved genes. The Y-axis represents the pathway terms. GO, Gene Ontology; KEGG, Kyoto Encyclopedia of Genes and Genomes.

lipid localization, cholesterol transport, regulation of plasma lipoprotein particle levels, sterol transport, regulation of lipid localization, organic hydroxy compound transport, sterol import, long-chain fatty acid import, response to lipoteichoic acid, cholesterol import. In the case of cellular components, no protein is enriched. Finally, in the case of molecular function, the top 10 proteins include the amide binding, Toll-like receptor binding, ceramide binding, low-density lipoprotein particle receptor activity, diacylglycerol binding, lipoprotein particle receptor activity, phosphatidylglycerol binding, low-density lipoprotein particle binding, high-density lipoprotein particle binding, signaling pattern recognition receptor activity [**Figure 3B**].

To explore the relationship between CTRP7 and signal pathway, we conducted a KEGG analysis. $p < 0.05$ was used as the screening condition, and the *p*-values were ranked from large to small. We found that CTRP7 related proteins were mainly enriched in cholesterol metabolism PPAR signaling pathway, ECM receptor interaction, etc. (**Figure 3C**). In addition, REACTOME enrichment analysis revealed that CTRP7-related genes were mainly enriched in metabolic-related signal pathways, such as cholesterol transport, HDL remodeling, lipoprotein remodeling, plasma lipoprotein clearance, and the relation of insulin secretion, etc. (**Figure 3D**).

DISCUSSION

In this study, we found that serum CTRP7 levels were significantly increased in MetS and IR individuals, and CTRP7 levels were associated with the disorder of glucose and lipid metabolism. Blood glucose, WC, APN, and BP were independent factors of circulating CTRP7. In addition, logistic regression analysis showed that high serum CTRP7 concentration was significantly associated with the occurrence of MetS and IR. We also found no significant difference in circulating CTRP7 levels between men and women, indicating that there may be no correlation between CTRP7 and sex hormones in study population. In the intervention studies, the OGTT test showed to a decrease in serum CTRP7 level, while the HEC test showed an increase in CTRP7 level. In addition, lipid infusion also increased serum CTRP7 levels. Bioinformatics analysis further showed that CTRP7 was associated with genes and signaling pathways related to glucose and lipid metabolism.

Cytokines, especially adipocytokines, and genetic and environmental factors play an important role in the pathogenesis of MetS (28, 29). It has been well documented that APN is an insulin sensitizer, and plays an important role in the pathogenesis of IR, diabetes, and metabolic disease (30–32). Low circulating APN levels are associated with MetS components, such as hyperglycemia, hyperinsulinemia, and dyslipidemia (33–36). In the current study, we find that increased serum CTRP7 levels are significantly associated with decreased circulating APN levels in MetS patients, as well as with other MetS components such as BP, blood glucose, WC and TG, etc. Logistic regression and ROC curve analysis reveals that circulating CTRP7 is significantly related to the occurrence of MetS.

Similar to our results, a small sample study in obese individuals (n = 37) showed that the level of serum CTRP7 in obese individuals was significantly increased and positively correlated with BMI, glucose, insulin, and HOMA-IR (11). In addition, one study reported decreased circulating CTRP7 levels in coronary artery disease (CAD) patients and may serve as a biomarker of CAD (37). Another study revealed that the production of CTRP7 in muscle tissue was increased and further increased by caloric restriction in old animals (16). These data further suggest that CTRP7 is a secretory protein related to metabolism. Given the previous study in obese individuals and our above results, including the reverse change in serum CTRP7 and APN concentration, and the negative correlation between CTRP7 and APN, we believe that CTRP7 has a negative regulatory effect on metabolism and insulin sensitivity. Therefore, it may be a metabolic inhibitor *in vivo*.

To further explore the factors regulating the secretion and release of CTRP7 *in vivo*, we conducted a variety of intervention studies. As a response to the oral glucose challenge, serum CTRP7 levels decreased significantly during the OGTT. Results from OGTT indicated a relevant link between CTRP7 secretion and glucose and insulin levels *in vivo*. Furthermore, HEC resulted in a significant increase in circulating CTRP7 levels when blood glucose was maintained at basal levels. We considered that increased insulin levels may promote the secretion or release of CTRP7 *in vivo*. Therefore, combined with the results of the OGTT and EHC, we believe that hyperglycemia inhibits the secretion and release of CTRP7. On the other hand, hyperinsulinemia stimulated its secretion and release. However, the inhibitory effect of blood glucose on CTRP7 may be stronger than the role of insulin on the secretion and release of CTRP7.

It is generally believed that the increase of FFA can lead to IR (36). To further explore whether elevated serum FFA has an effect on circulating CTRP7 levels, we performed a 4-hour intralipid infusion plus EHC in normal individuals. The results showed that the increased serum FFAs stimulated the secretion and release of CTRP7, resulting in a significant increase of circulating CTRP7 levels. This result also suggested that CTRP7 is related to acute-IR induced by elevated FFAs and maybe a potential nutrient sensor involved in lipid metabolism.

Finally, we used the network gene database and bioinformatics platform to evaluate the association of CTRP7 and other metabolism-related genes and signal pathways. Bioinformatics analysis showed that CTRP7 was related to lipid-metabolism genes, such as CD36 and FAM132A. KEGG analysis also showed that CTRP7 related proteins were mainly enriched in cholesterol metabolism PPAR signaling pathway and ECM receptor interaction. Therefore, our bioinformatics analysis further revealed that CTRP7 is a gene related to metabolism. This result supports our cohort and intervention studies. We thus consider that CTRP7 may be used as a biomarker of MetS and other metabolic diseases.

There are also some limitations in the current study, including 1) the population included in this study was limited to the Han people. Therefore, our results need to be confirmed in the population of different races; 2) although we strictly controlled the selection criteria for the study cohorts, we cannot completely

exclude residual confounding factors; 3) our cross-sectional study did not reflect the changes of circulating CTRP7 levels in the development of MetS and the impact after treatment. Therefore, a long-term follow-up study is necessary; Although it is difficult to find a direct causal relationship with the results of a cross-sectional study, we believe that the data provided by this study provide an interesting avenue for further study of the association of CTRP7 with glucose and lipid metabolism.

In conclusion, our data show that MetS patients have high circulating CTRP7 levels, and lower APN levels when compared to controls. Circulating CTRP7 is associated with metabolism and MetS, and is regulated by glucose, insulin and FFA. The novelty of this study is that circulating CTRP7 levels in MetS patients were determined for the first time, and the relationship between CTRP7 and glucose and lipid metabolism as well as insulin sensitivity was evaluated by various intervention methods. Therefore, our data highlight the role of CTRP7 in MetS and its potential use as a predictive biomarker of MetS in the future.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/**Supplementary Material**. Further inquiries can be directed to the corresponding author.

ETHICS STATEMENT

This study was approved by the Human Research Ethics Committee of Chongqing Medical University. The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

WH, QL, SG, DL, MT, and LL researched and analyzed data. BZ, CC, and HL reviewed and edited the manuscript. GY and MY wrote and edited the manuscript, and is the guarantor of this work and, as such, had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. All authors contributed to the article and approved the submitted version.

FUNDING

This work was supported by research grants from the National Natural Science Foundation of China (81300670) and from the Science and Technology Program of the Health Bureau of Chongqing (2019ZDXM039). Natural Science Foundation Project of Chongqing CSTC (cstc2020jcyj-msxmX0952).

ACKNOWLEDGMENTS

We thank patients and healthy individuals who made this study possible.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2021.774309/full#supplementary-material

REFERENCES

1. Kahn R, Buse J, Ferrannini E, Stern M. The Metabolic Syndrome: Time for a Critical Appraisal. Joint Statement From the American Diabetes Association and the European Association for the Study of Diabetes. *Diabetologia* (2005) 48(9):1684–99. doi: 10.1007/s00125-005-1876-2
2. Ladeiras-Lopes R, Moreira HT, Bettencourt N, Fontes-Carvalho R, Sampaio F, Ambale-Venkatesh B, et al. Metabolic Syndrome Is Associated With Impaired Diastolic Function Independently of MRI-Derived Myocardial Extracellular Volume: The MESA Study. *Diabetes* (2018) 67(5):1007–12. doi: 10.2337/db17-1496
3. Peng Q, Karvonen-Gutierrez CA, Randolph JF, Nan B, McConnell D, Harlow SD. Age at Onset of Metabolic Syndrome Among Women With and Without Polycystic Ovary Syndrome-Like Status. *J Clin Endocrinol Metab* (2019) 104 (5):1429–39. doi: 10.1210/jc.2018-01428
4. Mi Q, Li Y, Wang M, Yang G, Zhao X, Liu H. Circulating C1q/TNF-Related Protein Isoform 15 Is a Marker for the Presence of Metabolic Syndrome. *Diabetes Metab Res Rev* (2019) 35(1):e3085. doi: 10.1002/dmrr.3085
5. Klunder-Klunder M, Flores-Huerta S, Garcia-Macedo R, Peralta-Romero J, Cruz M. Adiponectin in Eutrophic and Obese Children as a Biomarker to Predict Metabolic Syndrome and Each of Its Components. *BMC Public Health* (2013) 13:88. doi: 10.1186/1471-2458-13-88
6. Peterson JM, Wei Z, Wong GW. C1q/TNF-Related Protein-3 (CTRP3), a Novel Adipokine That Regulates Hepatic Glucose Output. *J Biol Chem* (2010) 285(51):39691–701. doi: 10.1074/jbc.M110.180695
7. Shapiro L, Scherer PE. The Crystal Structure of a Complement-1q Family Protein Suggests an Evolutionary Link to Tumor Necrosis Factor. *Curr Biol: CB* (1998) 8(6):335–8. doi: 10.1016/s0960-9822(98)70133-2
8. Kopp A, Bala M, Weigert J, Buchler C, Neumeier M, Aslanidis C, et al. Effects of the New Adiponectin Paralogous Protein CTRP-3 and of LPS on Cytokine Release From Monocytes of Patients With Type 2 Diabetes Mellitus. *Cytokine* (2010) 49(1):51–7. doi: 10.1016/j.cyto.2009.10.001
9. Wang M, Tang X, Li L, Liu D, Liu H, Zheng H, et al. C1q/TNF-Related Protein-6 Is Associated With Insulin Resistance and the Development of Diabetes in Chinese Population. *Acta Diabetol* (2018) 55(12):1221–9. doi: 10.1007/s00592-018-1203-2
10. Wong GW, Krawczyk SA, Kitidis-Mitrokostas C, Ge G, Spooner E, Hug C, et al. Identification and Characterization of CTRP9, A Novel Secreted Glycoprotein, From Adipose Tissue That Reduces Serum Glucose in Mice and Forms Heterotrimers With Adiponectin. *FASEB J: Off Publ Fed Am Societies Exp Biol* (2009) 23(1):241–58. doi: 10.1096/fj.08-114991
11. Petersen PS, Lei X, Wolf RM, Rodriguez S, Tan SY, Little HC, et al. CTRP7 Deletion Attenuates Obesity-Linked Glucose Intolerance, Adipose Tissue Inflammation, and Hepatic Stress. *Am J Physiol Endocrinol Metab* (2017) 312(4):E309–25. doi: 10.1152/ajpendo.00344.2016
12. Jiang F, Yang M, Zhao X, Liu R, Yang G, Liu D, et al. C1q/TNF-Related Protein5 (CTRP5) as a Biomarker to Predict Metabolic Syndrome and Each of Its Components. *Int J Endocrinol* (2018) 2018:7201473. doi: 10.1155/2018/7201473

13. Wong GW, Wang J, Hug C, Tsao TS, Lodish HF. A Family of Acrp30/adiponectin Structural and Functional Paralogs. *Proc Natl Acad Sci USA* (2004) 101(28):10302–07. doi: 10.1073/pnas.0403760101
14. Coleman DL, Hummel KP. The Influence of Genetic Background on the Expression of the Obese (Ob) Gene in the Mouse. *Diabetologia* (1973) 9 (4):287–93. doi: 10.1007/BF01221856
15. Wong GW, Krawczyk SA, Kitidis-Mitrokostas C, Revett T, Gimeno R, Lodish HF. Molecular, Biochemical and Functional Characterizations of C1q/TNF Family Members: Adipose-Tissue-Selective Expression Patterns, Regulation by PPAR-Gamma Agonist, Cysteine-Mediated Oligomerizations, Combinatorial Associations and Metabolic Functions. *Biochem J* (2008) 416 (2):161–77. doi: 10.1042/BJ20081240
16. Rohrbach S, Aurich AC, Li L, Niemann B. Age-Associated Loss in Adiponectin-Activation by Caloric Restriction: Lack of Compensation by Enhanced Inducibility of Adiponectin Paralogs CTRP2 and CTRP7. *Mol Cell Endocrinol* (2007) 277(1-2):26–34. doi: 10.1016/j.mce.2007.07.005
17. Chinese Diabetes Society. Guidelines for the Prevention and Treatment of Type 2 Diabetes in China (2017). *Chin J Diabetes Mellitus* (2018) 10(1):4–67. doi: 10.19538/j.nk2018040108
18. Alberti KG, Zimmet PZ. Definition, Diagnosis and Classification of Diabetes Mellitus and its Complications. Part 1: Diagnosis and Classification of Diabetes Mellitus Provisional Report of a WHO Consultation. *Diabetic Med: J Br Diabetic Assoc* (1998) 15(7):539–53. doi: 10.1002/(SICI)1096-9136(199807)15:7<539::AID-DIA668>3.0.CO;2-S
19. Bergman RN, Stefanovski D, Buchanan TA, Sumner AE, Reynolds JC, Sebring NG, et al. A Better Index of Body Adiposity. *Obes (Silver Spring)* (2011) 19 (5):1083–9. doi: 10.1038/oby.2011.38
20. He Y, Hu W, Yang G, Guo H, Liu H, Li L. Adipose Insulin Resistance and Circulating Betatrophin Levels in Women With PCOS. *BioMed Res Int* (2020) 2020:1253164. doi: 10.1155/2020/1253164
21. Xu X, Zhang T, Mokou M, Li L, Li P, Song J, et al. Follistatin-Like 1 as a Novel Adipomyokine Related to Insulin Resistance and Physical Activity. *J Clin Endocrinol Metab* (2020) 105(12):dgaa629. doi: 10.1210/clinem/dgaa629
22. Li K, Liao X, Wang K, Mi Q, Zhang T, Jia Y, et al. Myonectin Predicts the Development of Type 2 Diabetes. *J Clin Endocrinol Metab* (2018) 103(1):139–47. doi: 10.1210/jc.2017-01604
23. Hu W, Li L, Yang M, Luo X, Ran W, Liu D, et al. Circulating Sfrp5 Is a Signature of Obesity-Related Metabolic Disorders and Is Regulated by Glucose and Liraglutide in Humans. *J Clin Endocrinol Metab* (2013) 98(1):290–8. doi: 10.1210/jc.2012-2466
24. Yang S, Dai H, Hu W, Geng S, Li L, Li X, et al. Association Between Circulating Follistatin-Like-1 and Metabolic Syndrome in Middle-Aged and Old Population: A Cross-Sectional Study. *Diabetes Metab Res Rev* (2021) 37 (2):e3373. doi: 10.1002/dmrr.3373
25. Liu J, Li H, Sun L, Wang Z, Xing C, Yuan Y. Aberrantly Methylated-Differentially Expressed Genes and Pathways in Colorectal Cancer. *Cancer Cell Int* (2017) 17:75. doi: 10.1186/s12935-017-0444-4
26. Bell-Anderson KS, Funnell AP, Williams H, Mat Jusoh H, Scully T, Lim WF, et al. Loss of Kruppel-Like Factor 3 (KLF3/BKLF) Leads to Upregulation of the Insulin-Sensitizing Factor Adipolin (FAM132A/CTRP12/C1qdc2). *Diabetes* (2013) 62(8):2728–37. doi: 10.2337/db12-1745
27. Valsesia A, Saris WH, Astrup A, Hager J, Masoodi M. Distinct Lipid Profiles Predict Improved Glycemic Control in Obese, Nondiabetic Patients After a Low-Caloric Diet Intervention: The Diet, Obesity and Genes Randomized Trial. *Am J Clin Nutr* (2016) 104(3):566–75. doi: 10.3945/ajcn.116.137646
28. Kotnik P, Fischer Posovszky P, Wabitsch M. Endocrine and Metabolic Effects of Adipose Tissue in Children and Adolescents. *Zdr Varst* (2015) 54(2):131–8. doi: 10.1515/sjph-2015-0020
29. Magana Gomez JA, Moreno-Mascareno D, Angulo Rojo CE, de la Pena GD. Association of Total and High Molecular Weight Adiponectin With Components of Metabolic Syndrome in Mexican Children. *J Clin Res Pediatr Endocrinol* (2020) 12(2):180–8. doi: 10.4274/jcrpe.galenos.2019.2019.0113
30. Bloomgarden ZT. Adiposity and Diabetes. *Diabetes Care* (2002) 25(12):2342–9. doi: 10.2337/diacare.25.12.2342
31. Kershaw EE, Flier JS. Adipose Tissue as an Endocrine Organ. *J Clin Endocrinol Metab* (2004) 89(6):2548–56. doi: 10.1210/jc.2004-0395
32. Arner P. Insulin Resistance in Type 2 Diabetes – Role of the Adipokines. *Curr Mol Med* (2005) 5(3):333–9. doi: 10.2174/1566524053766022
33. Liu YL, Liang HR, Liu HT, Li SY, Zhou YY, Cheng HL, et al. Association of Serum Adiponectin Levels With Artherosclerosis and the Metabolic Syndrome in Obese Children. *J Pediatr Endocrinol Metab* (2010) 23(8):743–51. doi: 10.1515/jpem.2010.122
34. Li G, Xu L, Zhao Y, Li L, Fu J, Zhang Q, et al. Leptin-Adiponectin Imbalance as a Marker of Metabolic Syndrome Among Chinese Children and Adolescents: The BCAMS Study. *PloS One* (2017) 12(10):e0186222. doi: 10.1371/journal.pone.0186222
35. Mi J, Munkonda MN, Li M, Zhang MX, Zhao XY, Fouejeu PC, et al. Adiponectin and Leptin Metabolic Biomarkers in Chinese Children and Adolescents. *J Obes* (2010) 2010:892081. doi: 10.1155/2010/892081
36. Dresner A, Laurent D, Marcucci M, Griffin ME, Dufour S, Cline GW, et al. Effects of Free Fatty Acids on Glucose Transport and IRS-1-Associated Phosphatidylinositol 3-Kinase Activity. *J Clin Invest* (1999) 103(2):253–9. doi: 10.1172/JCI5001
37. Zhang Y, Liu C, Liu J, Guo R, Yan Z, Liu W, et al. Implications of C1q/TNF-Related Protein Superfamily in Patients With Coronary Artery Disease. *Sci Rep* (2020) 10(1):878. doi: 10.1038/s41598-020-57877-z

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 16 November 2021
doi: 10.3389/fendo.2021.762250

Utility of Three Adiposity Indices for Identifying Left Ventricular Hypertrophy and Geometric Remodeling in Chinese Children

Huan Wang[1], *Min Zhao*[2], *Costan G. Magnussen*[3,4,5] *and Bo Xi*[1]*

[1] Department of Epidemiology/Shandong Provincial Clinical Research Center for Emergency and Critical Care Medicine, School of Public Health/Qilu Hospital, Cheeloo College of Medicine, Shandong University, Jinan, China, [2] Department of Toxicology and Nutrition, School of Public Health, Cheeloo College of Medicine, Shandong University, Jinan, China, [3] Menzies Institute for Medical Research, University of Tasmania, Hobart, TAS, Australia, [4] Research Centre of Applied and Preventive Cardiovascular Medicine, University of Turku, Turku, Finland, [5] Centre for Population Health Research, University of Turku and Turku University Hospital, Turku, Finland

OPEN ACCESS

Edited by:
Ozra Tabatabaei-Malazy, Tehran University of Medical Sciences, Iran

Reviewed by:
Jun Ma, Peking University, China
Dianjianyi Sun, Peking University, China

***Correspondence:**
Bo Xi
xibo2010@sdu.edu.cn

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *21 August 2021*
Accepted: *01 November 2021*
Published: *16 November 2021*

Citation:
Wang H, Zhao M, Magnussen CG and Xi B (2021) Utility of Three Adiposity Indices for Identifying Left Ventricular Hypertrophy and Geometric Remodeling in Chinese Children. Front. Endocrinol. 12:762250. doi: 10.3389/fendo.2021.762250

Background: Previous studies have shown that waist-to-height ratio (WHtR) performed similarly well when compared to body mass index (BMI) and waist circumference (WC) for identifying cardiovascular risk factors. However, to our knowledge, the performance of these three adiposity indices for identifying left ventricular hypertrophy (LVH) and left ventricular geometric (LVG) remodeling in youth has not been assessed. We aimed to determine the utility of BMI, WC and WHtR for identifying LVH and LVG in Chinese children.

Methods: This study included 1,492 Chinese children aged 6-11 years. Adiposity indices assessed were BMI, WC and WHtR. LVH and high relative wall thickness (RWT) were defined using sex- and age-specific 90th percentile values of left ventricular mass index and RWT, respectively, based on the current population. LVG remodeling included concentric remodeling (CR), eccentric hypertrophy (EH) and concentric hypertrophy (CH), which was defined based on the combination of LVH and high RWT.

Results: The magnitude of association of central obesity defined by WHtR with LVH [odds ratio (*OR*) =10.09, 95% confidence interval (*CI*) =6.66-15.29] was similar with general obesity defined by BMI (*OR*=10.49, 95% *CI*=6.97-15.80), and both were higher than central obesity defined by WC (*OR*=6.87, 95% *CI*=4.57-10.33). Compared with BMI, WHtR had better or similar predictive utility for identifying LVH, EH, and CH [the area under the curve (AUC): 0.84 *vs.* 0.79; 0.84 *vs.* 0.77; 0.87 *vs.* 0.88, respectively]; WC had worse or similar discriminatory utility with AUCs of 0.73, 0.70, 0.83, respectively.

Conclusion: WHtR performed similarly or better than BMI or WC for identifying LVH and LVG remodeling among Chinese children. WHtR provides a simple and convenient measure of central obesity that might improve the discrimination of children with cardiac structural damage.

Keywords: waist-to-height ratio, body mass index, waist circumference, left ventricular hypertrophy, geometric remodeling, children

INTRODUCTION

The prevalence of pediatric obesity has greatly increased worldwide, particularly in low- and middle-income countries (1, 2). In China, the prevalence of general obesity (3) and abdominal obesity (4) among children and adolescents has markedly increased over the past three decades. Obesity is related to cardiovascular (CV) risk (including hyperglycemia, elevated blood pressure, dyslipidemia, metabolic syndrome, and insulin resistance) (5) and short-term target organ damage (6, 7) in childhood. For example, obesity increases the risk of left ventricular hypertrophy (LVH) and left ventricular geometric (LVG) remodeling (8–11) (markers of cardiac structural damage), which are independent predictors of cardiovascular disease (CVD) (12, 13).

LVH was independently associated with the long term adverse CV events, such as coronary heart disease, other CVD related death, and heart failure (14). Specific abnormal LVG remodeling also provided distinct prognostic information. For example, hypertensive patients with concentric hypertrophy (CH) had the highest CV events and all-cause mortality, followed by those with eccentric hypertrophy (EH) and concentric remodeling (CR), compared with those with normal geometry (15). Importantly, participants converting from CR to normal geometry had decreased risk of all-cause mortality (16). LVH has been the most common target organ damage in children and adolescents with hypertension (17), and obesity is strongly associated with abnormal LVG modeling (8–11). Therefore, assessing the presence of LVH and LVG in the early life using a simple and effective obesity-related indicator could be helpful to prevent target organ damage such as LVG remodeling in the short term and the CVD outcomes in the long term.

Although BMI is universally used to determine obesity-related comorbidity and mortality, as an index of obesity it has several limitations. For example, sex- and age-specific cut-offs (18, 19) complicate the use of BMI to define obesity in practice, and BMI does not accurately discriminate body fat distribution (20). In contrast, WC and WHtR are markers of abdominal adiposity that are more closely linked with metabolic disturbances (21), and more strongly associated with CVD outcomes (22) and all-cause mortality (23). Sex- and age-specific cut-offs of WC are also required to define central obesity, whereas WHtR is standardized for height and indirectly adjusts for the effect of age, which is a simple and pragmatic index to correctly assess central obesity (24).

Although recent studies have reported that BMI, WC and WHtR performed similarly to predict common CV risk factors in children and adolescents (25–30), how these adiposity indices comparison when identifying LVH and LVG remodeling in children is largely unknown. A simplified and effective method to identify preclinical cardiac remodeling in the pediatric population is important. Therefore, we aimed to assess the utility of BMI, WC and WHtR for identifying LVH and LVG remodeling (including CR, EH and CH) and to determine the optimal cut-off of WHtR in Chinese children aged 6-11 years.

METHODS

Subjects

This cross-sectional study included 1,492 children aged 6-11 years. Participants were recruited from one primary school in Huantai County, Zibo City, Shandong Province, China, using a convenient clustering sampling method from November 2017 to January 2018. Detailed information has been described elsewhere (31). Included participants provided written informed consent to participate in this study after knowing the aim and procedures. All children underwent physical measurements [height, weight, WC, and blood pressure (BP)] and an echocardiography examination at the school. A structured questionnaire was filled out by the children and their parents/guardians jointly at home. The study was approved by the Ethics Committees of the School of Public Health, Shandong University (Approval number: 20160308).

Measurements

Height, weight and WC were measured twice in accordance with a uniform procedure and repeated a third time if the first two values differed by more than 1.0 cm (height), 0.5 kg (weight) or 1.0 cm (WC), with mean of the multiple measurements used for data analysis. A calibrated electronic weighing scale with automatic stadiometer (HGM-300; China) was used to measure height and weight, with children required to stand erect in bare feet and in light clothes. A non-elastic plastic tape was used to measure WC in a horizontal plane directly on the skin above 1 cm of the umbilicus after a normal exhalation. BMI was calculated as weight (kg) divided by the square of height (m^2). WHtR was calculated as the ratio of WC (cm)/height (cm). The clinically validated and calibrated upper-arm electronic sphygmomanometer (Omron HEM-7012; Japan) was used to measure the BP in seated position, which is accurate at measuring BP among children aged under 18 years (32). Three BP readings were measured consecutively at one visit, with replicate readings performed after approximately 20-seconds,

by trained staff following recommendations proposed by the Chinese working group of blood pressure measurement (33). Mean values of the last two BP readings were used for data analysis. Given the effect of children's growth, the Z-scores for BMI, WC, WHtR and BP (the original values minus means specific for sex and age then divided by the standard deviations specific for sex and age) were calculated to reflect the sex- and age-specific distribution for these indices in this population and could be used for direct comparisons between different samples (e.g., with different ages and sexes).

A portable color Doppler Ultrasonography (CX30; USA) with 2-4 MHz convex array transducers was used to assess left ventricle structure. One experienced sonographer acquired all images following recommendations for cardiac chamber quantification (34). Left ventricular internal dimension (LVID), interventricular septal thickness (IVST) and left ventricular posterior wall thickness (LVPWT) were measured during diastole. In this study, the intra-class correlation coefficients for repeated measurements on the same 20 participants by the one operator were 0.92 for IVST and 0.95 for LVPWT. Left ventricular mass (LVM, g) was calculated as $0.8\times1.04\times[(LVID + IVST + LVPWT)^3\text{-}(LVID)^3] + 0.6$ according to the Devereux's formula (35). Left ventricular mass index (LVMI) was calculated as LVM (g) divided by height to the power of 2.7 ($m^{2.7}$) (36). Relative wall thickness (RWT) was calculated as (LVPWT + IVST)/LVID (37).

A self-reported structured questionnaire was administered to collect information on lifestyle variables, including daily sleep duration, daily screen time, daily physical activity time, frequency of daily vegetable/fruit intake and weekly frequency of soft drink intake. Short sleep duration (< 9 hours per day) (38), long screen time (> 2 hours per day) (39), insufficient physical activity (< 1 hour per day) (40) and insufficient intake of vegetable/fruit (< 5 servings per day) (41) were defined according to pediatric recommendations. Finally, more frequent intake of soft drink (≥ 1 time per week) was defined based the distribution of frequency for our study population. The frequency of soft drink intake in the past 30 days was assessed using a self-designed questionnaire with 6 options: 'never', 'less than once per month', '1-3 times per month', '1-2 times per week', '3-5 times per week', and 'nearly everyday'. According to the distribution of frequency among this study population, we defined the more frequent intake of soft drink as more than once per week.

Definitions of Obesity, LVH and LVG Remodeling

BMI-obese and WC-obese were defined using sex- and age-specific cut-offs of BMI and WC for Chinese children (19, 42, 43); WHtR-obese was defined as WHtR ≥ 0.5 (44). LVH was defined as LVMI ≥ sex- and age-specific 90th percentile and high RWT was defined as RWT ≥ sex- and age-specific 90th percentile for this population. LVG patterns were further categorized as: normal geometry (normal LVMI and normal RWT); CR (normal LVMI and high RWT); EH (LVH and normal RWT); and CH (LVH and high RWT) (45). In sensitivity analysis, sex- and age-specific 95th percentile values of LVMI and RWT for this population were used to re-define LVH, CR, EH and CH.

Statistical Analysis

Continuous variables are expressed as means (standard deviations) and categorical variables as numbers (%); group differences are examined by the student's t-test or chi-square test as appropriate. The proportions of LVH and LVG remodeling across obesity status subgroups defined by BMI, WC and WHtR were compared using the chi-square test. Multivariable logistic regression models were used to examine the association of obesity with LVH and LVG remodeling; adjusted odds ratios (*ORs*) and 95% confidence intervals (95% *CIs*) were estimated after adjusting for sex, age, daily sleep duration, daily screen time, daily physical activity time, daily frequency of vegetable/fruit intake, weekly frequency of soft drink intake, Z-scores for systolic and diastolic BP. The receiver operating characteristic (ROC) curve analysis was used to compare the performance of BMI, WC and WHtR for identifying LVH and LVG remodeling, with BMI as the referent, and the area under the ROC curve (AUC), sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) were calculated. Generally, an AUC value < 0.7 is considered poor, 0.7-0.8 as acceptable and > 0.8 as good (46). The optimal cut-off of WHtR in the present population was determined by maximizing the Youden index (sensitivity + specificity - 1) (47). All statistical analyses were conducted using SAS version 9.4 and R version 4.0.2 with "pROC" package (48); a two-sided P value < 0.05 was considered statistically significant.

RESULTS

Participant Characteristics

A total of 1,492 children (boys: 53.2%) aged 6-11 years were included in this study. Among them, 146 children had LVH. Compared to children without LVH, those with LVH had higher levels, on average, of BMI, WC, WHtR, BMI Z-score, WC Z-score, WHtR Z-score, systolic and diastolic BP, and Z-scores for systolic and diastolic BP (all P <0.05, **Table 1**). Besides, sex- and age-adjusted means for BMI, WC, WHtR were still higher among children with LVH than those without LVH (BMI: 22.44 *vs.* 17.74 kg/m ^{2}; WC: 72.88 *vs.* 61.91 cm; WHtR: 0.53 *vs.* 0.45). About two thirds of children with LVH were classified as obese, irrespective of the indices (BMI, WC or WHtR) used to define obesity, which was around three times as high as the proportion for those without LVH. A larger proportion of children without LVH (16.8%) had a short sleep duration (<9 hours/day) than those with LVH (8.9%); 8.2% of children with LVH had an exceeded screen time (>2 hours/day), nearly twice as high as those without LVH (4.3%) (**Table 1**). Children with CH had the highest levels of BMI, WC, WHtR, BMI Z-score, WC Z-score, WHtR Z-score and systolic BP, and those with normal geometry had the lowest levels. The proportions of obesity in children with abnormal geometry (CR: 31.2-42.2%; EH: 60.0-65.0%; CH: 80.4-

TABLE 1 | Characteristics of participants according to the presence of left ventricular hypertrophy.

Characteristics	Total (n = 1492)	LVH (n = 146)	Normal (n = 1346)	*P* value*
Age, years	8.90 (1.51)	8.87 (1.55)	8.91 (1.51)	0.750
Boys	793 (53.2)	78 (53.4)	715 (53.1)	0.944
BMI, kg/m ^{2}	18.20 (3.45)	22.42 (4.46)	17.74 (2.99)	<0.001
WC, cm	62.98 (9.80)	72.80 (13.19)	61.92 (8.73)	<0.001
WHtR	0.46 (0.06)	0.53 (0.06)	0.45 (0.05)	<0.001
BMI Z-score	0.00 (1.00)	1.24 (1.17)	-0.14 (0.88)	<0.001
WC Z-score	0.00 (1.00)	1.05 (1.18)	-0.11 (0.90)	<0.001
WHtR Z-score	0.00 (1.00)	1.28 (1.04)	-0.14 (0.89)	<0.001
SBP, mmHg	106.34 (9.20)	108.63 (9.79)	106.09 (9.10)	0.002
DBP, mmHg	63.62 (6.68)	65.29 (7.58)	63.44 (6.55)	0.005
SBP Z-score	0.00 (1.00)	0.26 (0.98)	-0.03 (1.00)	0.001
DBP Z-score	0.00 (1.00)	0.26 (1.08)	-0.03 (0.98)	0.001
BMI-obese	327 (21.9)	97 (66.4)	230 (17.1)	<0.001
WC-obese	470 (31.5)	105 (71.9)	365 (27.1)	<0.001
WHtR-obese	363 (24.3)	99 (67.8)	264 (19.6)	<0.001
Sleep duration <9 hours/day	239 (16.0)	13 (8.9)	226 (16.8)	0.014
Screen time > 2 hours/day	70 (4.7)	12 (8.2)	58 (4.3)	0.034
Physical activity time < 1 hour/day	859 (57.6)	88 (60.3)	771 (57.3)	0.487
Intake of vegetable/fruit < 5 servings/day	1214 (81.4)	126 (86.3)	1088 (80.8)	0.107
Intake of soft drink ≥ 1 time/week	91 (6.1)	9 (6.2)	82 (6.1)	0.972

**Differences in characteristics between children with and without LVH were assessed using t test or chi-square test.*
Continuous variables are expressed as means (standard deviations) and categorical variables as numbers (%).
LVH, left ventricular hypertrophy; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure.

87.0%) were much higher than those with normal geometry (15.8-25.8%) (**Supplementary Table 1**).

Association of Indices of Obesity With LVH and LVG Remodeling

The prevalence of LVH, CR, EH and CH was higher in children with obesity, regardless of the measures (BMI, WC or WHtR) used to define obesity. Among those without obesity, CR was the prominent phenotype of LVG remodeling, while EH was the prominent phenotype in those with obesity (**Table 2** and **Supplementary Table 2**). The magnitude of the association of WHtR-obese with LVH (*OR* =10.09, 95% *CI* =6.66-15.29) was similar to the association observed for BMI-obese (*OR*=10.49, 95% *CI*=6.97-15.80), with both stronger than WC-obese (*OR*=6.87, 95% *CI*=4.57-10.33). WHtR-obese and BMI-obese also had a similar magnitude of association with LVG remodeling, which were both stronger than WC-obese. Besides, the continuous Z-scores for BMI, WC and WHtR were also positively associated with LVH and LVG remodeling (**Table 3**). Sensitivity analysis showed similar associations of obesity with LVH and LVG remodeling (**Supplementary Table 3**).

Utility of Adiposity Indices for Identifying LVH and LVG Remodeling

Compared with BMI (AUC=0.79, 95% *CI*: 0.75-0.84), WC (AUC=0.73, 95% *CI*: 0.68-0.79) had worse predictive utility for identifying LVH, while WHtR (AUC=0.84, 95% *CI*: 0.81-0.88) outperformed BMI (**Table 4** and **Figure 1**). BMI, WC and WHtR had similarly poor discriminatory utility for CR (all AUCs below 0.7) (**Table 4** and **Figure 1**). For identifying EH, WC

TABLE 2 | Prevalence of left ventricular hypertrophy and left ventricular geometric remodeling according to obesity status, n (%).

Obesity status	LVH	LVG remodeling		
		CR	EH	CH
BMI				
Normal (n=1165)	49 (4.2)	75 (6.4)	40 (3.4)	9 (0.8)
Obese (n=327)	97 (29.7)	34 (10.4)	60 (18.4)	37 (11.3)
P value*	<0.001	0.015	<0.001	<0.001
WC				
Normal (n=1022)	41 (4.0)	63 (6.2)	35 (3.4)	6 (0.6)
Obese (n=470)	105 (22.3)	46 (9.8)	65 (13.8)	40 (8.5)
P value*	<0.001	0.013	<0.001	<0.001
WHtR				
Normal (n=1129)	47 (4.2)	70 (6.2)	38 (3.4)	9 (0.8)
Obese (n=363)	99 (27.3)	39 (10.7)	62 (17.1)	37 (10.2)
P value*	<0.001	0.004	<0.001	<0.001

**Differences in the prevalence of LVH or LVG between non-obese and obese groups were assessed using chi-square test.*
LVH, left ventricular hypertrophy; LVG, left ventricular geometric; CR, concentric remodeling; EH, eccentric hypertrophy; CH, concentric hypertrophy; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio.

TABLE 3 | Association of obesity with left ventricular hypertrophy and left ventricular geometric remodeling.

	Model 1		Model 2		Model 3	
	OR (95% *CI*)	*P* value	*OR* (95% *CI*)	*P* value	*OR* (95% *CI*)	*P* value
LVH						
BMI-obese	9.73 (6.70-14.14)	<0.001	9.76 (6.70-14.23)	<0.001	10.49 (6.97-15.80)	<0.001
WC-obese	6.92 (4.73-10.13)	<0.001	6.75 (4.60-9.90)	<0.001	6.87 (4.57-10.33)	<0.001
WHtR-obese	9.58 (6.52-14.08)	<0.001	9.75 (6.60-14.39)	<0.001	10.09 (6.66-15.29)	<0.001
BMI Z-score	3.38 (2.82-4.07)	<0.001	3.41 (2.83-4.11)	<0.001	3.96 (3.20-4.91)	<0.001
WC Z-score	2.80 (2.36-3.32)	<0.001	2.81 (2.36-3.34)	<0.001	3.15 (2.58-3.85)	<0.001
WHtR Z-score	3.69 (3.04-4.48)	<0.001	3.76 (3.08-4.58)	<0.001	4.30 (3.44-5.38)	<0.001
CR						
BMI-obese	2.42 (1.57-3.74)	<0.001	2.49 (1.61-3.86)	<0.001	2.16 (1.36-3.44)	0.001
WC-obese	2.11 (1.41-3.14)	<0.001	2.21 (1.47-3.31)	<0.001	1.95 (1.26-3.00)	0.003
WHtR-obese	2.62 (1.71-4.02)	<0.001	2.74 (1.78-4.22)	<0.001	2.45 (1.56-3.86)	<0.001
BMI Z-score	1.64 (1.34-2.00)	<0.001	1.67 (1.37-2.05)	<0.001	1.59 (1.28-1.99)	<0.001
WC Z-score	1.74 (1.44-2.10)	<0.001	1.78 (1.47-2.16)	<0.001	1.73 (1.39-2.15)	<0.001
WHtR Z-score	1.54 (1.26-1.89)	<0.001	1.58 (1.28-1.94)	<0.001	1.49 (1.19-1.86)	<0.001
EH						
BMI-obese	8.11 (5.27-12.46)	<0.001	8.21 (5.32-12.68)	<0.001	8.18 (5.12-13.09)	<0.001
WC-obese	5.40 (3.51-8.31)	<0.001	5.26 (3.41-8.11)	<0.001	4.97 (3.13-7.91)	<0.001
WHtR-obese	8.27 (5.31-12.89)	<0.001	8.51 (5.44-13.33)	<0.001	8.28 (5.14-13.35)	<0.001
BMI Z-score	3.26 (2.66-4.01)	<0.001	3.30 (2.68-4.07)	<0.001	3.69 (2.90-4.68)	<0.001
WC Z-score	2.75 (2.26-3.35)	<0.001	2.77 (2.27-3.39)	<0.001	2.99 (2.37-3.77)	<0.001
WHtR Z-score	3.67 (2.95-4.55)	<0.001	3.75 (3.01-4.69)	<0.001	4.17 (3.25-5.35)	<0.001
CH						
BMI-obese	21.94 (10.40-46.28)	<0.001	21.74 (10.26-46.07)	<0.001	26.02 (11.72-57.76)	<0.001
WC-obese	19.06 (8.00-45.39)	<0.001	19.03 (7.95-45.55)	<0.001	21.44 (8.69-52.87)	<0.001
WHtR-obese	20.24 (9.48-43.21)	<0.001	20.56 (9.56-44.23)	<0.001	23.17 (10.38-51.73)	<0.001
BMI Z-score	4.63 (3.44-6.21)	<0.001	4.64 (3.43-6.28)	<0.001	5.81 (4.11-8.21)	<0.001
WC Z-score	3.73 (2.80-4.98)	<0.001	3.72 (2.77-4.99)	<0.001	4.48 (3.20-6.28)	<0.001
WHtR Z-score	4.50 (3.32-6.08)	<0.001	4.56 (3.33-6.23)	<0.001	5.47 (3.84-7.78)	<0.001

OR, odds ratio; CI, confidence interval; LVH, left ventricular hypertrophy; CR, concentric remodeling; EH, eccentric hypertrophy; CH, concentric hypertrophy; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio.

Model 1: Adjusted for sex and age.

Model 2: Model 1 + daily sleep duration, daily screen time, daily physical activity, frequency of daily vegetable/fruit intake and frequency of weekly soft drink intake.

Model 3: Model 2 + Z-scores for systolic and diastolic blood pressure.

(AUC=0.70, 95% *CI*: 0.63-0.77) was inferior to BMI (AUC=0.77, 95% *CI*: 0.71-0.82), and WHtR (AUC=0.84, 95% *CI*: 0.80-0.88) had the best performance (**Table 4** and **Figure 1**). For identifying CH, WC (AUC=0.83, 95% *CI*: 0.76-0.90) had a worse utility in comparison to BMI (AUC=0.88, 95% *CI*: 0.82-0.94) and WHtR (AUC=0.87, 95% *CI*: 0.82-0.92) (**Table 4** and **Figure 1**). Z-scores of these three adiposity indices showed largely similar discriminatory utility as those using the original scales (**Table 4** and **Figure 2**). Similar results were observed in the sensitivity analysis (**Supplementary Table 4**).

In addition, the optimal cut-offs of WHtR to identify LVH, CR, EH and CH were around 0.50 for boys (0.50, 0.50, 0.48, 0.51, respectively), while the corresponding optimal cut-offs were somewhat lower than 0.50 for girls (0.46, 0.49, 0.46, 0.47, respectively) (**Figure 3**). Except for CR, the sensitivity of WHtR was above 0.80, suggesting that WHtR could perform well to correctly identify children with LVH, EH and CH (**Table 4**).

DISCUSSION

In this population-based study of Chinese children aged 6-11 years, we found that the magnitude of association of central obesity defined by WHtR with LVH and LVG remodeling was similar with obesity defined by BMI, and both were stronger than central obesity defined by WC; WHtR performed similarly or better than BMI or WC for discriminating LVH and LVG remodeling; and WHtR cut-offs of ~0.5 for boys and <0.5 for girls provided the best discrimination of those with LVH and LVG remodeling. Our findings contribute to the evidence base that support the use of WHtR to identify children at risk of cardio-metabolic outcomes by extending them to include subclinical cardiac structural damage.

Consistent with previous studies (10, 49–51), we found obesity was associated with LVH, and the magnitude of the association was stronger for central obesity defined by WHtR and general obesity defined by BMI than central obesity defined by WC. A cross-sectional study including 281 outpatient children aged 6-16 years found that WHtR was significantly associated with LVH, while BMI or WC was not (51). However, another cross-sectional study among 96 children and adolescents aged 7-15 years showed that WHtR was less strongly correlated with LVMI among these three adiposity indices (52). Differences in demographic characteristics (e.g., different distributions of sex and age) and various methods for measurements of anthropometric and cardiac structural indices (e.g., different positions and devices during measurements) might explain these inconsistent results.

TABLE 4 | Utility of adiposity indices for identifying left ventricular hypertrophy and left ventricular geometric remodeling.

	AUC (95% *CI*)	*P* value*	Sensitivity, %	Specificity, %	PPV, %	NPV, %
LVH						
BMI	0.79 (0.75-0.84)	Ref.	63.0	87.2	34.7	95.6
WC	0.73 (0.68-0.79)	<0.001	55.5	88.6	34.6	94.8
WHtR	0.84 (0.81-0.88)	0.001	89.7	63.7	21.2	98.3
BMI Z-score	0.82 (0.78-0.86)	Ref.	73.3	80.8	29.2	96.5
WC Z-score	0.78 (0.73-0.82)	<0.001	70.6	78.5	26.2	96.1
WHtR Z-score	0.85 (0.82-0.88)	0.007	91.1	65.1	22.1	98.5
CR						
BMI	0.61 (0.56-0.67)	Ref.	54.1	67.4	12.8	94.3
WC	0.63 (0.57-0.68)	0.193	62.4	61.8	12.6	94.9
WHtR	0.60 (0.54-0.66)	0.493	34.9	82.8	15.1	93.5
BMI Z-score	0.62 (0.56-0.67)	Ref.	52.3	65.6	11.8	94.0
WC Z-score	0.65 (0.59-0.70)	0.011	73.4	49.5	11.4	95.5
WHtR Z-score	0.61 (0.55-0.66)	0.282	35.8	81.9	14.8	93.5
EH						
BMI	0.77 (0.71-0.82)	Ref.	60.0	86.0	25.8	96.4
WC	0.70 (0.63-0.77)	<0.001	50.0	92.0	33.6	95.8
WHtR	0.84 (0.80-0.88)	<0.001	92.0	61.9	16.3	99.0
BMI Z-score	0.80 (0.75-0.85)	Ref.	67.0	81.8	23.0	96.8
WC Z-score	0.75 (0.70-0.81)	0.001	64.0	79.9	20.5	96.5
WHtR Z-score	0.84 (0.81-0.88)	0.001	91.0	65.4	17.5	98.9
CH						
BMI	0.88 (0.82-0.94)	Ref.	91.3	75.7	12.2	99.6
WC	0.83 (0.76-0.90)	<0.001	82.6	76.2	11.4	99.2
WHtR	0.87 (0.82-0.92)	0.866	80.4	83.4	15.2	99.1
BMI Z-score	0.90 (0.85-0.95)	Ref.	84.8	86.5	18.9	99.4
WC Z-score	0.86 (0.81-0.92)	0.033	84.8	80.1	13.7	99.3
WHtR Z-score	0.89 (0.85-0.94)	0.741	80.4	85.6	17.2	99.2

LVH, left ventricular hypertrophy; CR, concentric remodeling; EH, eccentric hypertrophy; CH, concentric hypertrophy; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio; AUC, area under the operating characteristic curve; CI, confidence interval; PPV, positive predictive value; NPV, negative predictive value; Ref, referent.
**Comparisons of AUCs with BMI as the referent.*

In our study, obesity was also associated with LVG remodeling, and the magnitude of the association of obesity with CH was the strongest, followed by EH and CR. A study including 62 normotensive children aged 8-11 years showed that elevated WHtR increased the odds of any phenotype of LVG remodeling (8). A study among 526 children aged 6-15 years found that neither obesity (defined by BMI) nor WC Z-score was associated with CR, whereas obesity and WC Z-score was independently associated with EH and CH (11). Another study including 343 African American youths with a mean age of 13.8 years reported that obesity (defined by BMI) was associated with CR independent of BP, but not with CH (53). The heterogeneity of ethnicity, the definition of LVG remodeling patterns and the consideration of potential covariates might partially explain these inconsistent findings.

Previous studies have compared the predictive utility of BMI, WC and WHtR for common CV risk factors in children and adolescents (25–30, 54–56) with most reporting similar performance among these three adiposity indices (25–30). However, few studies have assessed the performance for identifying LVH and LVG remodeling among children. A cross-sectional study of 10,907 adults from China reported no discrimination utility of BMI, WC or WHtR for CR (all AUCs <0.5), and similarly fair performance for EH and CH (AUCs ranging from 0.63 to 0.72) (57). Another study including 281 white children aged 6-16 years reported that WHtR performed best for identifying LVH compared with BMI or WC, with AUCs (95% *CI*) of 0.711 (0.650-0.733), 0.680 (0.616-0.743) and 0.657 (0.593-0.722), respectively (51). Likewise, our study reported that WHtR performed similarly or better than BMI or WC for identifying LVH and LVG remodeling; suggesting the use of WHtR to screen CV risk factors in practice could improve the ability to identify youth at high risk of cardiac remodeling. It has been shown that WHtR could outperform BMI and WC to predict total and trunk adiposity in children and adolescents (58). And increased adiposity may contribute to cardiac remodeling by hemodynamic and metabolic pathways, including increase in stroke volume and cardiac output, disorder of cardiac metabolism, activation of sympathetic nervous system, and secretion of adipokines by the adipose tissue (10, 59).

However, we found that all three adiposity indices had low predictive utility of CR with the AUCs of around 0.6, suggesting that a single index alone might have limited value to predict this remodeling phenotype in children. In addition, generally low PPVs but high NPVs suggest the better utility for identifying those without abnormal cardiac structures, which might be explained by the low prevalence of LVH/LVG remodeling among children.

It has been generally accepted that a WHtR cut-off of 0.5 might be useful in predicting CV risk in youth, independent of sex, age or ethnicity (44, 60), whereas a study including children

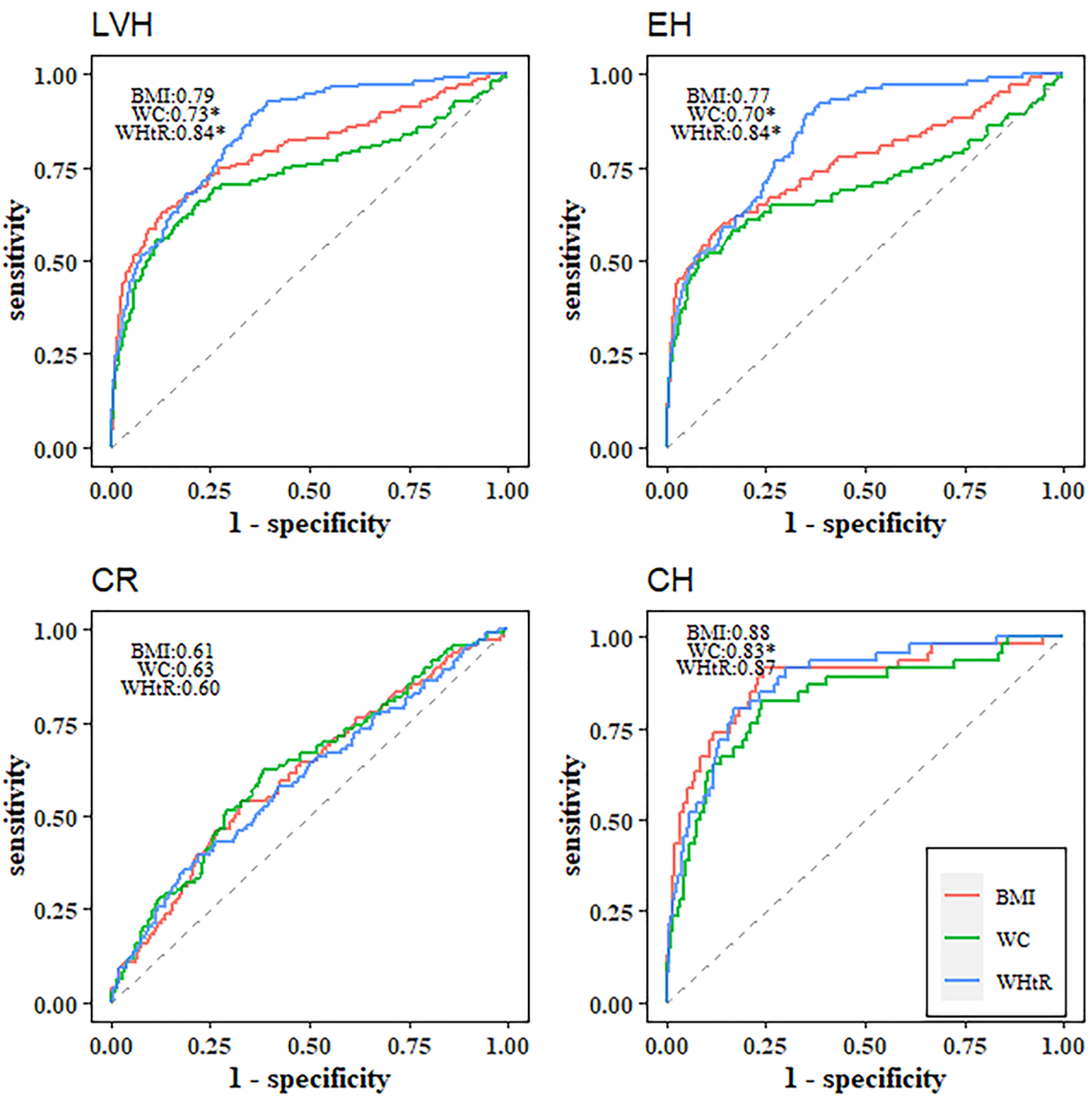


FIGURE 1 | Receiver operating characteristic curves of BMI, WC and WHtR for identifying left ventricular hypertrophy and left ventricular geometric remodeling. *Statistically significant difference in area under the operating characteristic curves as compared with BMI. LVH, left ventricular hypertrophy; CR, concentric remodeling; EH, eccentric hypertrophy; CH, concentric hypertrophy; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio.

and adolescents aged 7-19 years from Europe (13,172; boys: 49.7%) and southern China (14,566; boys: 50.3%) reported that a WHtR cut-off of 0.5 was not suitable for diverse ethnic groups, with a lower threshold of WHtR proposed, especially for girls from southern China (61). Another multicenter study involving 8,130 children and adolescents aged 7-18 years (boys: 53.2%) from China suggested that the optimal WHtR cut-off of 0.467 would be more accurate to identify the clustering of CV risk factors in the pediatric population, and that the optimal cut-offs varied across sex and age with lower cut-offs for girls and those aged 12 years or older (62). In our study, the optimal cut-offs of WHtR were ~0.5 for boys and <0.5 for girls, which may support the sex dependent cut-offs of WHtR. Besides, there were some differences in basic characteristics between boys and girls in our study (**Supplementary Table 5**). For example, boys had higher BMI, WC, WHtR, and SBP than girls, and boys tended to have unhealthy eating habits (such as insufficient intake of vegetable/fruit and more frequent intake of soft drink) compared with girls. Sex hormones (e.g., estrogens and androgens) and genetic mechanisms could regulate glucose and lipid homeostasis, energy metabolism and gene expression in a sexually dimorphic manner resulting in sex-specific cardio-metabolic disorders (63). Therefore, sex dependent cut-offs for identifying LVH/LVG are necessary according to these

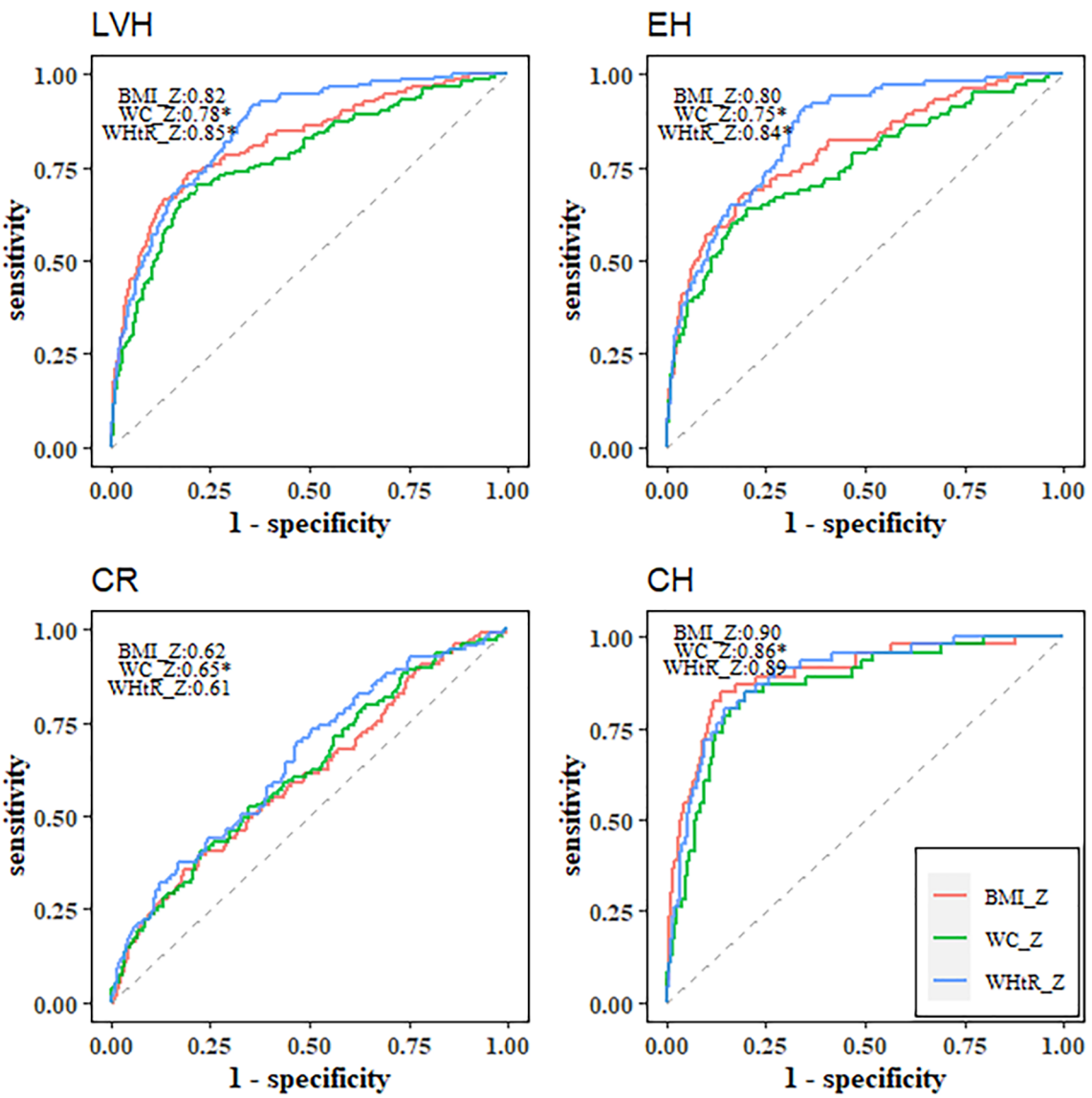


FIGURE 2 | Receiver operating characteristic curves of BMI Z-score, WC Z-score and WHtR Z-score for identifying left ventricular hypertrophy and left ventricular geometric remodeling. *Statistically significant difference in area under the operating characteristic curves as compared with BMI Z-score. LVH, left ventricular hypertrophy; CR, concentric remodeling; EH, eccentric hypertrophy; CH, concentric hypertrophy; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio.

different sex-specific features in cardio-metabolic disorders. Importantly, a simple and effective index for screening obesity in youth could be useful to identify obesity-related CV risk factors and to prevent the target organ damage in the early stage, and further cohort studies with a large sample size are needed to confirm our findings.

The strengths of our study were that we compared the discriminatory capability of BMI, WC and WHtR and determined the optimal cut-off of WHtR based on more clinically relevant markers of target organ damage, LVH and LVG remodeling, among children in a relatively large sample. However, our study has several limitations. First, the design of this study was cross-sectional and the interpretation of predictive utility of these three adiposity indices should be made with caution as we are unable to discount reverse causation. Second, the present study only included children aged 6-11 years from one primary school, which limits the generalizability of our findings. Third, despite a standard questionnaire being used to collect lifestyle variables, only diet frequency was assessed without more details about food quality and quantity, and puberty status was not evaluated in the present study. Therefore, it is possible that our estimates might be biased due to unmeasured or residual confounding.

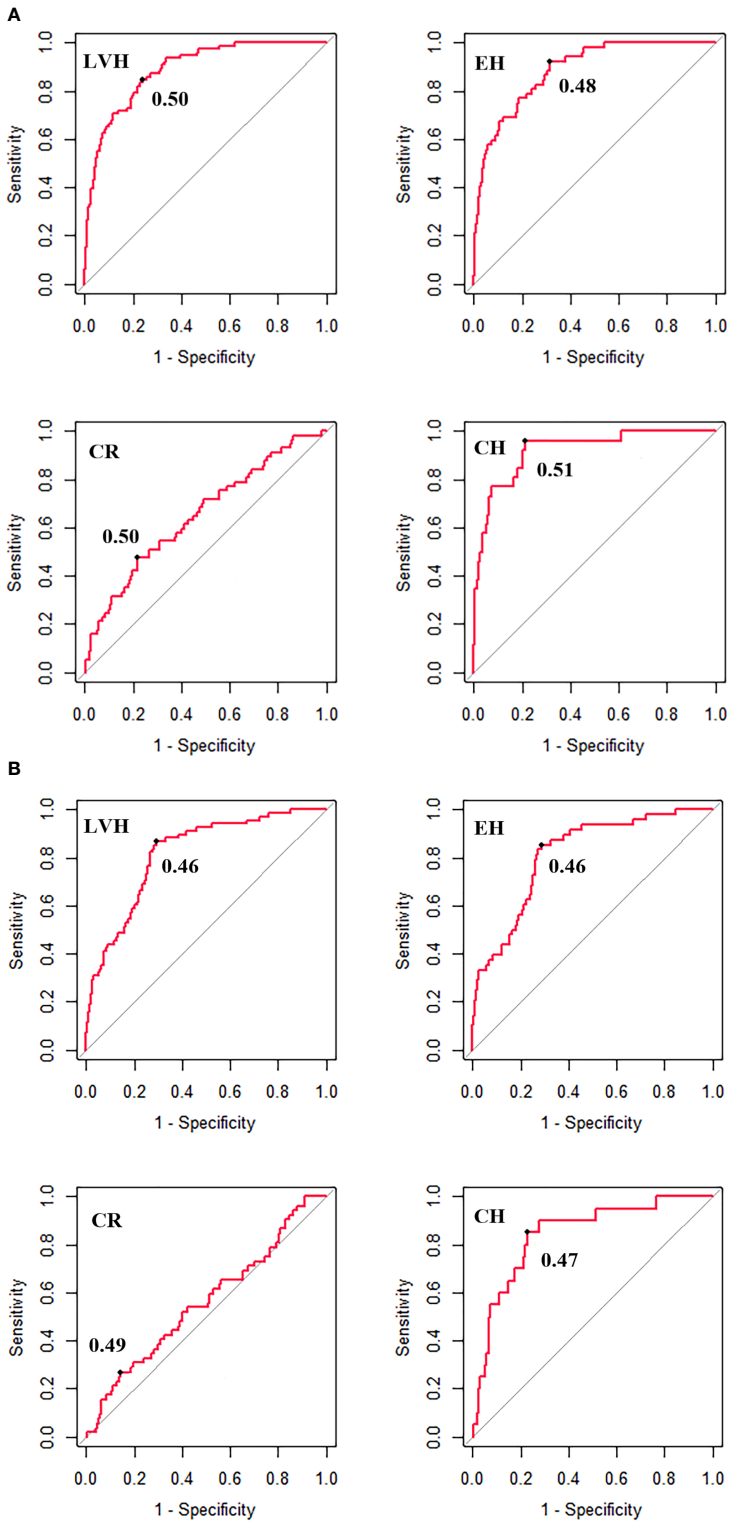


FIGURE 3 | The optimal cut-off of WHtR for identifying left ventricular hypertrophy and left ventricular geometric remodeling for boys **(A)** and girls **(B)**. LVH, left ventricular hypertrophy; CR, concentric remodeling; EH, eccentric hypertrophy; CH, concentric hypertrophy; BMI, body mass index; WC, waist circumference; WHtR, waist-to-height ratio. The optimal cut-off is the threshold corresponding to the maximum of Youden Index (sensitivity + specificity -1).

In summary, WHtR performed similarly well or better than BMI or WC for identifying LVH and LVG remodeling, with a WHtR cut-off of ~0.5 for boys and <0.50 for girls shown to have the best discriminatory utility. Our data suggest that WHtR could be used to identify children at risk of having subclinical cardiac structural damage.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/**Supplementary Material**. Further inquiries can be directed to the corresponding author.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by Ethics Committees of the School of Public Health, Shandong University (Approval number: 20160308). Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

AUTHOR CONTRIBUTIONS

BX conceptualized and designed the study, supervised the data collation, statistical analyses, and reviewed and revised the manuscript. HW did the statistical analyses, drafted the initial manuscript, and reviewed and revised the manuscript. CM, MZ, and BX reviewed and revised the manuscript. All authors contributed to the article and approved the submitted version.

FUNDING

This work was supported by the National Natural Science Foundation of China (81722039, 81673195).

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2021.762250/full#supplementary-material

REFERENCES

1. NCD Risk Factor Collaboration (NCD-RisC). Worldwide Trends in Body-Mass Index, Underweight, Overweight, and Obesity From 1975 to 2016: A Pooled Analysis of 2416 Population-Based Measurement Studies in 128.9 Million Children, Adolescents, and Adults. *Lancet* (2017) 390(10113):2627–42. doi: 10.1016/s0140-6736(17)32129-3
2. Yang L, Bovet P, Ma C, Zhao M, Liang Y, Xi B. Prevalence of Underweight and Overweight Among Young Adolescents Aged 12-15 Years in 58 Low-Income and Middle-Income Countries. *Pediatr Obes* (2019) 14(3):e12468. doi: 10.1111/ijpo.12468
3. Song Y, Agardh A, Ma J, Li L, Lei Y, Stafford RS, et al. National Trends in Stunting, Thinness and Overweight Among Chinese School-Aged Children, 1985-2014. *Int J Obes (Lond)* (2019) 43(2):402–11. doi: 10.1038/s41366-018-0129-7
4. Ma S, Hou D, Zhang Y, Yang L, Sun J, Zhao M, et al. Trends in Abdominal Obesity Among Chinese Children and Adolescents, 1993–2015. *J Pediatr Endocrinol Metab* (2021) 34(2):163–9. doi: 10.1515/jpem-2020-0461
5. Ayer J, Charakida M, Deanfield JE, Celermajer DS. Lifetime Risk: Childhood Obesity and Cardiovascular Risk. *Eur Heart J* (2015) 36(22):1371–6. doi: 10.1093/eurheartj/ehv089
6. Park MH, Skow Á, De Matteis S, Kessel AS, Saxena S, Viner RM, et al. Adiposity and Carotid-Intima Media Thickness in Children and Adolescents: A Systematic Review. *BMC Pediatr* (2015) 15:161. doi: 10.1186/s12887-015-0478-5
7. Jing L, Nevius CD, Friday CM, Suever JD, Pulenthiran A, Mejia-Spiegeler A, et al. Ambulatory Systolic Blood Pressure and Obesity Are Independently Associated With Left Ventricular Hypertrophic Remodeling in Children. *J Cardiovasc Magn Reson* (2017) 19(1):86. doi: 10.1186/s12968-017-0401-3
8. Giannisi F, Keivanidou A, Sakellari I, Balala S, Hassapidou M, Hitoglou-Makedou A, et al. Anthropometric and Biochemical Markers as Possible Indicators of Left Ventricular Abnormal Geometric Pattern and Function Impairment in Obese Normotensive Children. *Diagnostics (Basel)* (2020) 10 (7):468. doi: 10.3390/diagnostics10070468
9. Jing L, Binkley CM, Suever JD, Umasankar N, Haggerty CM, Rich J, et al. Cardiac Remodeling and Dysfunction in Childhood Obesity: A Cardiovascular Magnetic Resonance Study. *J Cardiovasc Magn Reson* (2016) 18(1):28. doi: 10.1186/s12968-016-0247-0
10. Brady TM. The Role of Obesity in the Development of Left Ventricular Hypertrophy Among Children and Adolescents. *Curr Hypertens Rep* (2016) 18(1):3. doi: 10.1007/s11906-015-0608-3
11. Pieruzzi F, Antolini L, Salerno FR, Giussani M, Brambilla P, Galbiati S, et al. The Role of Blood Pressure, Body Weight and Fat Distribution on Left Ventricular Mass, Diastolic Function and Cardiac Geometry in Children. *J Hypertens* (2015) 33(6):1182–92. doi: 10.1097/hjh.0000000000000552
12. Lavie CJ, Patel DA, Milani RV, Ventura HO, Shah S, Gilliland Y. Impact of Echocardiographic Left Ventricular Geometry on Clinical Prognosis. *Prog Cardiovasc Dis* (2014) 57(1):3–9. doi: 10.1016/j.pcad.2014.05.003
13. Armstrong AC, Gidding S, Gjesdal O, Wu C, Bluemke DA, Lima JA. LV Mass Assessed by Echocardiography and CMR, Cardiovascular Outcomes, and Medical Practice. *JACC Cardiovasc Imaging* (2012) 5(8):837–48. doi: 10.1016/j.jcmg.2012.06.003
14. Kawel-Boehm N, Kronmal R, Eng J, Folsom A, Burke G, Carr JJ, et al. Left Ventricular Mass at MRI and Long-Term Risk of Cardiovascular Events: The Multi-Ethnic Study of Atherosclerosis (MESA). *Radiology* (2019) 293(1):107–14. doi: 10.1148/radiol.2019182871
15. Koren MJ, Devereux RB, Casale PN, Savage DD, Laragh JH. Relation of Left Ventricular Mass and Geometry to Morbidity and Mortality in Uncomplicated Essential Hypertension. *Ann Intern Med* (1991) 114(5):345–52. doi: 10.7326/0003-4819-114-5-345
16. Milani RV, Lavie CJ, Mehra MR, Ventura HO, Kurtz JD, Messerli FH. Left Ventricular Geometry and Survival in Patients With Normal Left Ventricular Ejection Fraction. *Am J Cardiol* (2006) 97(7):959–63. doi: 10.1016/j.amjcard.2005.10.030
17. National High Blood Pressure Education Program Working Group on High Blood Pressure in Children and Adolescents. The Fourth Report on the Diagnosis, Evaluation, and Treatment of High Blood Pressure in Children and Adolescents. *Pediatrics* (2004) 114(Suppl2):555–76.
18. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a Standard Definition for Child Overweight and Obesity Worldwide: International Survey. *BMJ* (2000) 320(7244):1240–3. doi: 10.1136/bmj.320.7244.1240
19. Li H, Zong XN, Ji CY, Mi J. Body Mass Index Cut-Offs for Overweight and Obesity in Chinese Children and Adolescents Aged 2-18 Years. *Chin J Epidemiol* (2010) 31(6):616–20. doi: 10.3760/cma.j.issn.0254-6450.2010.06.004
20. Freedman DS, Wang J, Maynard LM, Thornton JC, Mei Z, Pierson RN, et al. Relation of BMI to Fat and Fat-Free Mass Among Children and Adolescents. *Int J Obes (Lond)* (2005) 29(1):1–8. doi: 10.1038/sj.ijo.0802735
21. Lee BJ, Yim MH. Comparison of Anthropometric and Body Composition Indices in the Identification of Metabolic Risk Factors. *Sci Rep* (2021) 11 (1):9931. doi: 10.1038/s41598-021-89422-x

22. Xue R, Li Q, Geng Y, Wang H, Wang F, Zhang S. Abdominal Obesity and Risk of CVD: A Dose-Response Meta-Analysis of Thirty-One Prospective Studies. *Br J Nutr* (2021) 126(9):1420–30. doi: 10.1017/S0007114521000064
23. Jayedi A, Soltani S, Zargar MS, Khan TA, Shab-Bidar S. Central Fatness and Risk of All Cause Mortality: Systematic Review and Dose-Response Meta-Analysis of 72 Prospective Cohort Studies. *BMJ* (2020) 370:m3324. doi: 10.1136/bmj.m3324
24. Taylor RW, Williams SM, Grant AM, Taylor BJ, Goulding A. Predictive Ability of Waist-to-Height in Relation to Adiposity in Children Is Not Improved With Age and Sex-Specific Values. *Obes (Silver Spring)* (2011) 19 (5):1062–8. doi: 10.1038/oby.2010.217
25. Quadros TMB, Gordia AP, Silva LR. Anthropometry and Clustered Cardiometabolic Risk Factors in Young People: A Systematic Review. *Rev Paul Pediatr* (2017) 35(3):340–50. doi: 10.1590/1984-0462/;2017;35;3;00013
26. Lo K, Wong M, Khalechelvam P, Tam W. Waist-to-Height Ratio, Body Mass Index and Waist Circumference for Screening Paediatric Cardio-Metabolic Risk Factors: A Meta-Analysis. *Obes Rev* (2016) 17(12):1258–75. doi: 10.1111/obr.12456
27. Gomes TN, Nevill A, Katzmarzyk PT, Pereira S, Dos Santos MM, Buranarugsa R, et al. Identifying the Best Body-Weight-Status Index Associated With Metabolic Risk in Youth. *Scand J Med Sci Sports* (2018) 28(11):2375–83. doi: 10.1111/sms.13249
28. Zhao M, Bovet P, Ma C, Xi B. Performance of Different Adiposity Measures for Predicting Cardiovascular Risk in Adolescents. *Sci Rep* (2017) 7:43686. doi: 10.1038/srep43686
29. Sardinha LB, Santos DA, Silva AM, Grøntved A, Andersen LB, Ekelund U. A Comparison Between BMI, Waist Circumference, and Waist-to-Height Ratio for Identifying Cardio-Metabolic Risk in Children and Adolescents. *PloS One* (2016) 11(2):e0149351. doi: 10.1371/journal.pone.0149351
30. Bauer KW, Marcus MD, El ghormli L, Ogden CL, Foster GD. Cardio-Metabolic Risk Screening Among Adolescents: Understanding the Utility of Body Mass Index, Waist Circumference and Waist to Height Ratio. *Pediatr Obes* (2015) 10(5):329–37. doi: 10.1111/ijpo.267
31. Yang LL, Zhang Q, Zhang YQ, Sun JH, Zhao M, Xi B. Design of Huantai Childhood Cardiovascular Health Cohort Study. *Chin J Prev Med* (2020) 54 (12):1461–4. doi: 10.3760/cma.j.cn112150-20200610-00857
32. Meng LH, Hou DQ, Shan XY, Mi J. Accuracy Evaluation of Omron Hem-7012 Electronic Sphygmomanometers in Measuring Blood Pressure of Children and Adolescents. *Chin J Hypertens* (2013) 21(2):158–62. doi: 10.16439/j.cnki.1673-7245.2013.02.036
33. Wang W, Zhang WZ, Sun NL, Lin JX, Chen LY, Wu KG, et al. Chinese Guidelines for Blood Pressure Measurement. *Chin J Hypertens* (2011) 19 (12):1101–15. doi: 10.16439/j.cnki.1673-7245.2011.12.004
34. Lang RM, Bierig M, Devereux RB, Flachskampf FA, Foster E, Pellikka PA, et al. Recommendations for Chamber Quantification: A Report From the American Society of Echocardiography's Guidelines and Standards Committee and the Chamber Quantification Writing Group, Developed in Conjunction With the European Association of Echocardiography, a Branch of the European Society of Cardiology. *J Am Soc Echocardiogr* (2005) 18 (12):1440–63. doi: 10.1016/j.echo.2005.10.005
35. Devereux RB, Alonso DR, Lutas EM, Gottlieb GJ, Campo E, Sachs I, et al. Echocardiographic Assessment of Left Ventricular Hypertrophy: Comparison to Necropsy Findings. *Am J Cardiol* (1986) 57(6):450–8. doi: 10.1016/0002-9149(86)90771-x
36. de Simone G, Daniels SR, Devereux RB, Meyer RA, Roman MJ, Dedivitiis O, et al. Left-Ventricular Mass and Body Size in Normotensive Children and Adults-Assessment of Allometric Relations and Impact of Overweight. *J Am Coll Cardiol* (1992) 20(5):1251–60. doi: 10.1016/0735-1097(92)90385-z
37. Reichek N, Devereux RB. Reliable Estimation of Peak Left Ventricular Systolic Pressure by M-Mode Echographic-Determined End-Diastolic Relative Wall Thickness: Identification of Severe Valvular Aortic Stenosis in Adult Patients. *Am Heart J* (1982) 103(2):202–3. doi: 10.1016/0002-8703(82)90493-8
38. Paruthi S, Brooks LJ, D'Ambrosio C, Hall WA, Kotagal S, Lloyd RM, et al. Recommended Amount of Sleep for Pediatric Populations: A Consensus Statement of the American Academy of Sleep Medicine. *J Clin Sleep Med* (2016) 12(6):785–6. doi: 10.5664/jcsm.5866
39. American Academy of Pediatrics. Council on Communications and Media. Children, Adolescents, and the Media. *Pediatrics* (2013) 132(5):958–61. doi: 10.1542/peds.2013-2656
40. WHO. Global Recommendations on Physical Activity for Health. Available at: https://www.who.int/dietphysicalactivity/factsheet_recommendations/en/ (Accessed 2021 28th March).
41. WHO. Effectiveness of Interventions and Programmes Promoting Fruit and Vegetable Intake. Available at: https://www.who.int/dietphysicalactivity/publications/f&v_promotion_effectiveness.pdf?ua=1 (Accessed 2021 28th March).
42. Zong XN, Li H, Zhang YQ. Percentile Reference Value of Waist Circumference for Chinese Children Aged 3-7 Years. *Chin J Epidemiol* (2020) 41(8):1286–90. doi: 10.3760/cma.j.cn112338-20190827-00629
43. Ma GS, Ji CY, Ma J, Mi J, Sung RY, Xiong F, et al. Waist Circumference Reference Values for Screening Cardiovascular Risk Factors in Chinese Children and Adolescents Aged 7-18 Years. *Chin J Epidemiol* (2010) 31 (6):609–15. doi: 10.3760/cma.j.issn.0254-6450.2010.06.003
44. Ashwell M, Gibson S. A Proposal for a Primary Screening Tool: 'Keep Your Waist Circumference to Less Than Half Your Height'. *BMC Med* (2014) 12:207. doi: 10.1186/s12916-014-0207-1
45. Ganau A, Devereux RB, Roman MJ, de Simone G, Pickering TG, Saba PS, et al. Patterns of Left Ventricular Hypertrophy and Geometric Remodeling in Essential Hypertension. *J Am Coll Cardiol* (1992) 19(7):1550–8. doi: 10.1016/0735-1097(92)90617-v
46. Mandrekar JN. Receiver Operating Characteristic Curve in Diagnostic Test Assessment. *J Thorac Oncol* (2010) 5(9):1315–6. doi: 10.1097/JTO.0b013e3181ec173d
47. Zou KH, O'Malley AJ, Mauri L. Receiver-Operating Characteristic Analysis for Evaluating Diagnostic Tests and Predictive Models. *Circulation* (2007) 115 (5):654–7. doi: 10.1161/CIRCULATIONAHA.105.594929
48. Robin X, Turck N, Hainard A, Tiberti N, Lisacek F, Sanchez JC, et al. Proc: An Open-Source Package for R and S+ to Analyze and Compare Roc Curves. *BMC Bioinf* (2011) 12:77. doi: 10.1186/1471-2105-12-77
49. Mehta SK. Waist Circumference to Height Ratio and Left Ventricular Mass in Children and Adolescents. *Cardiol Young* (2016) 26(4):658–62. doi: 10.1017/S1047951115000803
50. Mehta SK, Richards N, Lorber R, Rosenthal GL. Abdominal Obesity, Waist Circumference, Body Mass Index, and Echocardiographic Measures in Children and Adolescents. *Congenit Heart Dis* (2009) 4(5):338–47. doi: 10.1111/j.1747-0803.2009.00330.x
51. Di Bonito P, Moio N, Sibilio G, Cavuto L, Sanguigno E, Forziato C, et al. Cardiometabolic Phenotype in Children With Obesity. *J Pediatr* (2014) 165 (6):1184–9. doi: 10.1016/j.jpeds.2014.08.007
52. Rodicio MM, Domenech de Miguel V, Guinda Jiménez M, Cigárran Guldris S, López Franco MM, Estany Gestal A, et al. Early Cardiac Abnormalities in Obese Children and Their Relationship With Adiposity. *Nutrition* (2018) 46:83–9. doi: 10.1016/j.nut.2017.09.001
53. Dhuper S, Abdullah RA, Weichbrod L, Mahdi E, Cohen HW. Association of Obesity and Hypertension With Left Ventricular Geometry and Function in Children and Adolescents. *Obes (Silver Spring)* (2011) 19(1):128–33. doi: 10.1038/oby.2010.134
54. Mai TMT, Gallegos D, Jones L, Tran QC, Tran TMH, van der Pols JC. The Utility of Anthopometric Indicators to Identify Cardiovascular Risk Factors in Vietnamese Children. *Br J Nutr* (2020) 123(9):1043–55. doi: 10.1017/S0007114520000203
55. Aguilar-Morales I, Colin-Ramirez E, Rivera-Mancia S, Vallejo M, Vázquez-Antona C. Performance of Waist-to-Height Ratio, Waist Circumference, and Body Mass Index in Discriminating Cardio-Metabolic Risk Factors in a Sample of School-Aged Mexican Children. *Nutrients* (2018) 10(12):1850. doi: 10.3390/nu10121850
56. Ma L, Cai L, Deng L, Zhu Y, Ma J, Jing J, et al. Waist Circumference Is Better Than Other Anthropometric Indices for Predicting Cardiovascular Disease Risk Factors in Chinese Children–a Cross-Sectional Study in Guangzhou. *J Atheroscler Thromb* (2016) 23(3):320–9. doi: 10.5551/jat.31302
57. Chang Y, Guo X, Li T, Li S, Guo J, Sun Y. A Body Shape Index and Body Roundness Index: Two New Body Indices to Identify Left Ventricular Hypertrophy Among Rural Populations in Northeast China. *Heart Lung Circ* (2016) 25(4):358–64. doi: 10.1016/j.hlc.2015.08.009
58. Brambilla P, Bedogni G, Heo M, Pietrobelli A. Waist Circumference-to-Height Ratio Predicts Adiposity Better Than Body Mass Index in Children

and Adolescents. *Int J Obes (Lond)* (2013) 37(7):943–6. doi: 10.1038/ijo.2013.32
59. Abel ED, Litwin SE, Sweeney G. Cardiac Remodeling in Obesity. *Physiol Rev* (2008) 88(2):389–419. doi: 10.1152/physrev.00017.2007
60. Yoo EG. Waist-to-Height Ratio as a Screening Tool for Obesity and Cardiometabolic Risk. *Korean J Pediatr* (2016) 59(11):425–31. doi: 10.3345/kjp.2016.59.11.425
61. Nawarycz T, So HK, Choi KC, Sung RY, Li AM, Nelson EA, et al. Waist-to-Height Ratio as a Measure of Abdominal Obesity in Southern Chinese and European Children and Adolescents. *Int J Obes (Lond)* (2016) 40(7):1109–18. doi: 10.1038/ijo.2015.251
62. Dou Y, Jiang Y, Yan Y, Chen H, Zhang Y, Chen X, et al. Waist-to-Height Ratio as a Screening Tool for Cardiometabolic Risk in Children and Adolescents: A Nationwide Cross-Sectional Study in China. *BMJ Open* (2020) 10(6):e037040. doi: 10.1136/bmjopen-2020-037040
63. Gerdts E, Regitz-Zagrosek V. Sex Differences in Cardiometabolic Disorders. *Nat Med* (2019) 25(11):1657–66. doi: 10.1038/s41591-019-0643-8

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 04 January 2022
doi: 10.3389/fendo.2021.795044

Metabolic Syndrome and Its Components Are Associated With Altered Amino Acid Profile in Chinese Han Population

Shuiya Sun[1†], Dongjuan He[1,2†], Cheng Luo[1,2], Xihua Lin[1], Jiahua Wu[1], Xueyao Yin[1], Chengfang Jia[1], Qianqian Pan[1], Xuehong Dong[1], Fenping Zheng[1], Hong Li[1] and Jiaqiang Zhou[1*]*

[1] Department of Endocrinology, Sir Run Run Shaw Hospital, Zhejiang University School of Medicine, Hangzhou, China, [2] Department of Endocrinology, People's Hospital of Quzhou, Quzhou, China

OPEN ACCESS

Edited by:
Ozra Tabatabaei-Malazy, Tehran University of Medical Sciences, Iran

Reviewed by:
Jarlei Fiamoncini, University of São Paulo, Brazil
Basmah Eldakhakhny, King Abdulaziz University, Saudi Arabia
Ghada M. A. Ajabnoor, King Abdulaziz University, Saudi Arabia

***Correspondence:**
Jiaqiang Zhou
zjq8866@zju.edu.cn
Hong Li
srrshnfm@zju.edu.cn

[†]These authors have contributed equally to this work

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: 14 October 2021
Accepted: 08 December 2021
Published: 04 January 2022

Citation:
Sun S, He D, Luo C, Lin X, Wu J, Yin X, Jia C, Pan Q, Dong X, Zheng F, Li H and Zhou J (2022) Metabolic Syndrome and Its Components Are Associated With Altered Amino Acid Profile in Chinese Han Population. Front. Endocrinol. 12:795044. doi: 10.3389/fendo.2021.795044

Objective: Recent studies have found that the levels of plasma amino acids, such as branched-chain amino acids and aromatic amino acids, were associated with visceral obesity, insulin resistance, future development of diabetes and cardiovascular diseases. However, few studies have involved a Chinese Han population. This study aimed to examine the association between amino acid profile and metabolic syndrome (MetS) and its components in the Chinese Han population.

Methods: This is a cross-sectional study, which enrolled a cohort of 473 participants from a community. We employed the isotope internal standard method to determine the plasma concentrations of 28 amino acids using high-performance liquid chromatography-tandem mass spectrometry (LC/MS). Participants were divided into MetS (n = 72) and non-MetS groups (n = 401) to analyze the association between amino acids and MetS and its components.

Results: The prevalence of MetS was 15.2% according to the criteria. Plasma concentrations of isoleucine (Ile), leucine (Leu), valine (Val), tyrosine (Tyr), tryptophan (Trp), phenylalanine (Phe), glutamic acid (Glu), aspartic acid (Asp), alanine (Ala), histidine (His), methionine (Met), asparagine (Asn), and proline (Pro) were significantly higher in the MetS group than those in the non-MetS group ($P < 0.05$), but taurine (Tau) was significantly lower ($P < 0.05$). When MetS components were increased, the concentrations of these 13 amino acids significantly increased ($P < 0.05$), but Tau concentration was significantly decreased ($P < 0.05$). We extracted the amino acid profile by principal component analysis (PCA), PC1 and PC2, which extracted from the 14 amino acids, were significantly associated with MetS (odds ratio, 95% confidence interval: 1.723, 1.325–2.085 and 1.325, 1.043–1.684, respectively). A total of 260 non-MetS participants were followed up effectively, and 42 participants developed new-onset MetS within 5 years. We found that the amino acid profile of PC1 was linked to the occurrence of future MetS. Decreased Tau was correlated with the future development of MetS.

Conclusion: Participants with MetS exhibit an abnormal amino acid profile, and its components gradually increase when these amino acids are altered. Amino acid PCA profile can be employed for assessing and monitoring MetS risk. Finally, decreased Tau may be linked to the future development of MetS.

Keywords: amino acid, metabolic syndrome, component, biomarkers, amino acid profile

INTRODUCTION

Rapid lifestyle and dietary changes have contributed to a rise in the global prevalence of metabolic syndrome (MetS). MetS is a cluster of risk factors that increase the risk of an individual developing heart disease, diabetes, stroke, and chronic neurodegenerative disease (1–4). MetS diagnosis increases the relative risk for cardiovascular disease over 5 to 10 years by approximately 2-fold and type 2 diabetes mellitus (T2DM) by at least 5-fold (5). Current research on MetS pathogenesis mainly focuses on abdominal obesity, lipotoxicity, and insulin resistance. Recent studies have demonstrated that insulin resistance is strongly linked to amino acid metabolism, and it is believed that plasma amino acid levels may increase during insulin resistance. Branched-chain and aromatic amino acids (BCAAs and AAAs), in particular, are closely associated with the risk of visceral obesity, insulin resistance, and development of DM in the future. Plasma amino acid alterations in the early stage of lifestyle-related diseases are due to obesity and insulin resistance-related inflammation, and these alterations are reversed by appropriate (nutritional, pharmaceutical, or surgical) interventions that improve insulin sensitivity (6–10).

Accumulating evidence suggests that some amino acids could regulate various metabolic processes, including glucose and lipid metabolism. Evidence from American, Northern European, and Japanese populations link BCAAs and AAAs to insulin resistance, T2DM, and cardiometabolic risk. Research has demonstrated that the BCAAs and AAAs are positively correlated with body mass index (BMI), waist circumference, visceral fat, systolic blood pressure (SBP), diastolic blood pressure (DBP), fasting blood glucose, insulin and triglyceride levels, and insulin resistance, but inversely linked to high-density lipoprotein-cholesterol (HDL-c) in cross-sectional analyses of large prospective cohort studies (11–15). Interactions of excess BCAAs and lipids may cause β-cell dysfunction, accelerating the transition from an obese, insulin-resistant state to T2DM (16). Numerous studies have demonstrated that humans possess amino acid sensors that detect changes in amino acid levels and trigger corresponding metabolic responses, such as those mediated by serine/threonine-protein kinase general control non-derepressible 2 (GCN2), activating transcription factor 4 (ATF4), mechanistic target of rapamycin (mTOR), and 5′-adenosine monophosphate-activated protein kinase (AMPK) (17–19). Tremblay and Marette (20) demonstrated that BCAAs could downregulate glucose transport *via* the mTOR kinase pathway in skeletal muscle cells. Although little is known about the mechanisms underlying the increase in AAAs, it has been hypothesized that Tyr aminotransferase is repressed during states of insulin resistance and DM, resulting in elevations of circulating Tyr and Phe (21). Glycine (Gly) levels are lower among obese subjects compared with those with normal weight (22), and Gly has been inversely associated with BMI, waist circumference (WC) (23), and insulin resistance (24). Serine (Ser) and Asn are inversely linked to insulin resistance (24). Ornithine (Orn), His, and Pro are positively correlated with an adverse cardiometabolic risk profile in cross-sectional analyses. In addition, Pro and His have been positively associated with insulin resistance (24). Ntzouvani et al. (14) investigated potential patterns in amino acid plasma concentrations using principal component analysis (PCA). They discovered that MetS participants had significantly higher levels of BCAAs, AAAs, Glu, Asp, and Ala, and these amino acid patterns were significantly linked to MetS.

Tau is a sulfur-containing amino acid widely distributed in many tissues and organs. It is involved in various physiological processes. Many studies revealed that Tau effectively reduces cholesterol, triglycerides, blood glucose, and blood pressure (25–28). In a clinical trial with non-diabetic men, who were overweight or obese, oral Tau ameliorates lipid-induced functional β-cell decompensation and insulin resistance by reducing oxidative stress (29). Wu et al. (30) demonstrated that ameliorated hepatic insulin resistance by Tau might be associated with the inhibiting c-Jun N-terminal kinase (JNK1) activation and improving insulin signaling in the liver.

Although studies in other countries have revealed a correlation between amino acids and MetS, few studies existed involving the Chinese Han people. Accordingly, this study discussed the correlation between amino acid profile and MetS and its components in Chinese Han people.

MATERIALS AND METHODS

Research Subjects and Groups

Our study enrolled participants through community-based survey of MetS prevalence. Inclusion criteria include the following: residents from a community, aged 40–65 years, part of the Han population, and have not been intervened with MetS-related components previously. Exclusion criteria include the following: a history of previous cardiovascular events, the use of oral/intravenous glucocorticoids, liver cirrhosis and ascites, kidney damage, hyperthyroidism or hypothyroidism, malignancies, pregnant, or lactating women. We conducted physical examinations, biochemical examinations, and oral glucose tolerance tests. A total of 473 residents were surveyed, and the inclusion criteria for MetS followed the 2009 guidelines

of the International Diabetes Federation (IDF) and the American Heart Association/National Heart, Lung, and Blood Institute (AHA/NHLBI) (31). We divided participants into MetS and non-MetS groups and compared the amino acid levels between the two groups.

Diagnostic Criteria

MetS was defined according to the 2009 guidelines of the IDF and AHA/NHLBI, which include three of the following five items: 1) elevated waist circumference: population and country-specific, a waist circumference of ≥90 cm (Chinese men) or ≥80 cm (Chinese women); 2) elevated triglycerides (or drug treatment for elevated triglycerides) ≥150 mg/dl (1.7 mmol/L); 3) reduced HDL-c (or drug treatment for reduced HDL-c) <40 mg/dl (1.0 mmol/L) in men and <50 mg/dl (1.3 mmol/L) in women; 4) elevated blood pressure (or antihypertensive drug treatment in a patient with a history of hypertension): systolic ≥130 mmHg and/or diastolic ≥85 mmHg; and 5) elevated fasting glucose (or drug treatment of elevated glucose) ≥100 mg/dl (5.6 mmol/L).

Detection Method

Participants who were not diagnosed with diabetes received a 75-g oral glucose tolerance test (OGTT), whereas those who were previously diagnosed with diabetes were administered a 100-g carbohydrate (steamed bread meal) test. Venous blood samples were obtained at 0 and 2 h following either OGTT or steamed bread meal test. Following standard blood processing, serum and plasma aliquots were stored at −80°C until subsequent use. Using Abbott C16000 automatic biochemical analyzer (Chicago, IL, USA), several biochemical tests were analyzed, including fasting blood glucose (FBG), triglyceride (TG), total cholesterol (TC), low-density lipoprotein-cholesterol (LDL-c), HDL-c, alanine aminotransferase (ALT), aspartate aminotransferase (AST), serum creatinine (CREA), urea nitrogen (BUN), uric acid (UA), and urine albumin-to-creatinine ratio (UACR). Glycosylated hemoglobin (HbA1c) was detected using high-performance liquid chromatography (Hemoglobin Testing System; Bio-Rad, CA, USA). Serum levels of insulin were measured by radioimmunoassay using an insulin detection kit (Beijing North Institute of Biological Technology, China). The homeostatic model assessment of insulin resistance (HOMA-IR) value was employed to evaluate the level of insulin sensitivity and was calculated as follows: fasting blood glucose (mmol/L) × fasting serum insulin (FINS; mU/L)/22.5 (32). Plasma concentrations of 28 amino acids were identified using LC/MS with isotope internal standard method by Beijing Emino Medical Research (Beijing, China). This method is accurate, reliable, and highly reproducible (33, 34). The main instruments used were as follows: the liquid phase model was an HPLC Ultimate3000 (Dionex Liquid Factory, CA, USA), and the mass spectrometer model was a 3200 Q TRAP LC-MS/MS (AB Company, CA, USA).

All participants underwent physical examinations by physicians using standard procedures, including taking measurements of their height, weight, WC, hip circumference (HC), and blood pressure. Blood pressure was measured three times (2 min between each measurement), and the average value was calculated. BMI was calculated by dividing body weight by height squared. Body fat percentage (Fat%) was measured using bioelectrical impedance analysis (TBF-300, Tanita Co., Tokyo, Japan). MRI scans were performed at the umbilicus level between L4 and L5 with the subject in a supine position. The abdominal visceral fat area (VFA) and abdominal subcutaneous fat area (SFA) were calculated using the SliceOmatic software (version 4.2). Smokers were defined as those who smoked at least one cigarette per day during the past year or recently stopped smoking (within the last 12 months); the remaining participants were defined as non-smokers. Alcohol drinkers were defined as those who consumed alcohol more than 3 days a week. Moderate exercise was defined as 30 min of exercise each time, 3–5 days a week.

All participants were interviewed face-to-face by trained medical staff to collect demographic data, baseline lifestyle, and health status data using a standardized questionnaire. This study was approved by the Ethics Committee of Sir Run Run Shaw Hospital. Written informed consent was obtained from all participants.

Statistical Methods

Data were analyzed using SPSS 26.0 (SPSS Inc., Chicago, IL, USA) and GraphPad Prism (version 7.0; GraphPad Software, Inc.). Categorical variables are expressed as frequencies [*n* (%)]. All continuous variables are reported as mean ± standard deviation (SD), while not normally distributed variables are expressed as median value (interquartile range). The Student's *t*-test was used to estimate differences in the distribution of demographic characteristics between case and control subjects for continuous variables. The Wilcoxon–Mann–Whitney *U* test was used for not normally distributed variables. One-way analysis of variance was used for intergroup comparisons of MetS components. The association between MetS and categorical variables was evaluated using Fischer's exact test. A single-factor logistic regression model was employed to assess the correlation between plasma amino acids and MetS to calculate OR value and the corresponding 95% confidence interval, and the difference was statistically significant with $P < 0.05$. Following that, logistic regression was deployed to evaluate the link between amino acids and MetS and its components after adjusting for potential covariates, including age, gender, alcohol drinking, current smoking status, and moderate exercise.

Furthermore, Pearson's correlation coefficients were calculated by using amino acids and metabolic-related variables. The side dendrogram represents the hierarchical clustering of 14 amino acids based on Pearson correlation. Due to the strong correlation between amino acids, PCA extracted the main factors to investigate the link between the amino acid profile and MetS.

RESULTS

Population Characteristics

Table 1 summarizes the clinical and biochemical characteristics of all participants, with an average age of 53.20 ± 6.7 years. The

TABLE 1 | Comparison of general clinical characteristics and biochemical indicators between the MetS group and non-MetS group.

Variables	Non-MetS group *n* = 401	MetS group *n* = 72	*P*-value
Female, *N* (%)	238 (59.35%)	45 (62.50%)	0.617
Current smoker, *N* (%)	118 (29.35%)	22 (30.56%)	0.982
Alcohol drinker, *N* (%)	163 (40.65%)	32 (44.44%)	0.689
Moderate exercise, *N* (%)	184 (45.77%)	35 (48.61%)	0.655
Age (years)	53.15 ± 6.64	56.13 ± 6.40	<0.001*
BMI (kg/m^2)	23.04 ± 2.55	26.81 ± 2.58	<0.001*
WC (cm)	77.03 ± 8.11	89.49 ± 6.23	<0.001*
WHR	0.86 ± 0.06	0.93 ± 0.05	<0.001*
Fat% (%)	28.36 ± 6.63	35.06 ± 6.91	<0.001*
SFA (cm^2)	145.6 (113.05, 192.05)	210 (167.93, 258.30)	<0.001*
VFA (cm^2)	65.08 (46.21, 103.3)	123.1 (96.06, 151.5)	<0.001*
SBP (mmHg)	120.57 ± 15.35	133.91 ± 15.14	<0.001*
DBP (mmHg)	80.58 ± 13.27	86.00 ± 9.49	0.001*
FPG (mmol/L)	5.02 ± 1.10	5.95 ± 1.81	<0.001*
2h-PG (mmol/L)	6.07 ± 3.14	8.77 ± 4.33	<0.001*
FINS (mIU/L)	10.85 (7.65, 14.16)	16.45 (10.96, 22.88)	<0.001*
2 h insulin (mIU/L)	40.25 (25.86, 62.29)	58.87 (37.86, 100.14)	<0.001*
HOMA-IR	2.37 (1.70, 3.25)	3.66 (2.47, 5.54)	<0.001*
HbA1c (%)	5.67 ± 0.70	6.04 ± 1.00	<0.001*
CHOL (mmol/L)	5.55 ± 1.04	5.86 ± 1.08	0.024*
TG (mmol/L)	1.25 (0.92, 1.65)	1.86 (1.40, 2.60)	<0.001*
LDL-c (mmol/L)	2.41 ± 0.56	2.52 ± 0.63	0.125
HDL-c (mmol/L)	1.48 ± 0.37	1.31 ± 0.31	<0.001*
ALT (IU/L)	18 (14, 26)	24.5 (19, 35.75)	<0.001*
AST (IU/L)	20 (17, 23)	21 (18, 25)	0.005*
CREA (mg/dl)	0.79 ± 0.15	0.80 ± 0.19	0.557
BUN (mmol/L)	16.50 ± 3.84	16.90 ± 5.13	0.447
UA (mg/dl)	4.67 ± 1.39	5.40 ± 1.31	<0.001*
UACR (mg/mmol)	4.59 (3.11, 6.79)	6.99 (4.59, 17.54)	0.001*

*Statistical differences between MetS and no MetS are shown as *P < 0.05.*

MetS, metabolic syndrome; BMI, body mass index; WC, waist circumference; WHR, waist-to-hip ratio; Fat%, body fat percentage; SFA, subcutaneous fat area; VFA, visceral fat area; SBP, Systolic blood pressure; DBP, diastolic blood pressure; FPG, fasting blood glucose; 2h-PG, 2-h postprandial glucose; FINS, fasting serum insulin; HOMA-IR, homeostasis model assessment for insulin resistance; HbA1c, glycosylated hemoglobin A1c; TC, total cholesterol; TG, triglyceride; LDL-c, low-density lipoprotein-cholesterol; HDL-c, high-density lipoprotein-cholesterol; ALT, alanine aminotransferase; AST, aspartate aminotransferase; CREA, serum creatinine; BUN, serum urea nitrogen; UA, uric acid; UACR, urine albumin-to-creatinine ratio.

mean age for MetS and non-MetS groups were 56.13 ± 6.40 and 53.15 ± 6.64 years, respectively. Age, BMI, WC, waist-to-hip ratio (WHR), Fat%, SFA, VFA, SBP, DBP, FPG, 2h-PG, FINS, 2h-insulin, HOMA-IR, HbA1c, CHOL, TG, ALT, AST, uric acid (UA), and UACR were higher in the MetS group than in the non-MetS group, but HDL-c was lower. The difference was statistically significant ($P < 0.05$; **Table 1**).

Altered Plasma Amino Acid Levels in MetS

Plasma levels of Ile, Leu, Val, Tyr, Trp, Phe, Glu, Asp, Ala, His, Met, Asn, and Pro were all significantly higher, whereas Tau was significantly lower in the MetS group than those in the non-MetS group, with a statistically significant difference ($P < 0.05$; **Table 2**). The other 14 amino acids were unrelated to MetS.

Altered Plasma Amino Acid Profile With MetS Components

All participants were divided into groups according to the number of MetS components (WC, blood glucose, increased TG, decreased HDL-c, and blood pressure). The groups were as follows: 0-component group, 1-component group, 2-components group, and 3–5-components group. The participant who had 3–5 components belonged to the MetS group. As the number of components increased, plasma levels of Ile, Leu, Val, Tyr, Trp, Phe, Glu, Asp, Ala, His, Met, Asn, and Pro increased progressively, but Tau decreased. Compared with the 0-component group, one-way analysis of variance revealed that the other groups exhibited statistically significant higher plasma levels of Ile, Leu, Val, Tyr, Trp, Phe, Glu, Asp, Ala, His, Met, Asn, and Pro and lower plasma levels of Tau ($P < 0.05$; **Table 3**). The other 14 amino acids were unrelated to MetS components.

Amino Acid Profiles Extracted by PCA Were Associated With MetS and Its Components

Pearson's correlation coefficients were calculated between the 14 amino acids and the metabolic variables linked to MetS (**Figure 1**). As illustrated in **Figure 1**, Ile, Leu, Val, Tyr, Trp, Glu, Ala, and Met showed strong positive correlations with BMI, WC, WHR, VFA, insulin, SBP, DBP, TG, and UA, but a strong negative correlation with HDL-c. Only Tau had negative correlations with metabolic variables. The side dendrogram represents the hierarchical clustering of the 14 amino acids based on Pearson correlation, and we can see a strong correlation with the amino acids (**Figure 2**). Multivariate regression analysis cannot be used, and the PCA method was

TABLE 2 | Comparison of amino acid determination results between the MetS group and non-MetS group (unit: μmol/L).

	Non-MetS group *n* = 401	MetS group *n* = 72	*P*-value	OR (95% CI)
Isoleucine (Ile)	54.26 ± 12.87	62.50 ± 17.57	<0.001*	1.040 (1.022–1.058)
Leucine (Leu)	133.03 ± 23.87	150.34 ± 28.49	<0.001*	1.026 (1.016–1.037)
Valine (Val)	187.09 ± 35.53	210.27 ± 45.38	<0.001*	1.015 (1.009–1.022)
Tyrosine (Tyr)	72.68 ± 15.10	81.01 ± 18.72	<0.001*	1.031 (1.015–1.046)
Tryptophan (Trp)	30.63 ± 4.94	33.77 ± 6.09	<0.001*	1.113 (1.061–1.168)
Phenylalanine (Phe)	138.91 ± 42.40	158.88 ± 46.43	<0.001*	1.009 (1.004–1.015)
Glutamic (Glu)	36.18 ± 5.99	39.88 ± 7.13	<0.001*	1.090 (1.049–1.134)
Aspartic (Asp)	6.92 ± 1.74	7.70 ± 1.98	<0.001*	1.243 (1.092–1.415)
Alanine (Ala)	202.92 ± 42.25	233.58 ± 38.49	<0.001*	1.017 (1.011–1.023)
Lysine (Lys)	150.43 ± 33.38	156.95 ± 31.31	0.124	1.006 (0.998–1.013)
Arginine (Arg)	124.23 ± 28.03	130.50 ± 29.44	0.083	1.007 (0.999–1.016)
Histidine (His)	97.20 ± 26.30	109.21 ± 38.16	0.001*	1.013 (1.005–1.022)
Methionine (Met)	44.05 ± 11.16	48.16 ± 13.09	0.008*	1.029 (1.008–1.050)
Threonine (Thr)	83.44 ± 23.23	84.64 ± 21.22	0.684	1.002 (0.992–1.013)
Glycine (Gly)	396.11 ± 86.29	391.84 ± 87.80	0.700	0.999 (0.996–1.002)
Serine (Ser)	101.23 ± 17.70	103.37 ± 19.94	0.354	1.007 (0.993–1.020)
Taurine (Tau)	1.57 ± 0.57	1.41 ± 0.52	0.019*	0.559 (0.345–0.905)
Asparagine (Asn)	49.16 ± 11.18	54.33 ± 14.41	<0.001*	1.034 (1.014–1.054)
Citrulline (Cit)	36.08 ± 9.81	37.22 ± 12.27	0.383	1.011 (0.987–1.035)
Ornithine (Orn)	153.15 ± 49.64	161.69 ± 44.26	0.173	1.003 (0.999–1.008)
Glutamine (Gln)	1065.36 ± 314.14	1073.22 ± 274.96	0.842	1.000 (0.999–1.001)
Cysteine (Cys)	143.39 ± 74.01	152.48 ± 66.94	0.331	1.002 (0.998–1.005)
Homocysteine (tHcy)	4.95 ± 1.12	4.68 ± 1.26	0.064	0.809 (0.648–1.010)
α-Aminobutyric acid	2.04 ± 0.66	2.04 ± 0.77	0.992	0.997 (0.689–1.443)
Hydroxyproline	19.44 ± 7.51	20.80 ± 9.19	0.172	1.022 (0.990–1.054)
1-Methylhistidine	33.23 ± 13.48	35.81 ± 14.79	0.141	1.013 (0.996–1.031)
3-Methylhistidine	1.66 ± 2.08	1.92 ± 2.03	0.329	1.054 (0.947–1.173)
Proline (Pro)	308.82 ± 92.76	336.59 ± 83.17	0.018*	1.003 (1.000–1.005)

*Statistical differences between MetS and no MetS are shown as *P < 0.05.*

used to investigate their potential patterns. The varimax rotation method was performed to produce interpretable components. Components with an eigenvalue ≥1.0 were extracted. Items with a factor loading ≥ $\left|0.4\right|$ were considered as composing a given factor. Factor scores were calculated using the regression method and used in subsequent analyses investigating correlations between extracted factors and MetS components. The amino acid profile, including Ile, Leu, Val, Tyr, Trp, Glu, Asp, Ala, His, Met, Asn, and Pro, denoted "PC1," was the dominant factor (accounting for 43.29% of the variance in PCA, **Table 4**). This amino acid profile was linked to MetS and its components, related to MetS, abdominal obesity, abnormal glucose,

TABLE 3 | Amino acid association with MetS components.

	0 component	1 component	2 components	3–5 components
	n = 124	*n* = 165	*n* = 112	*n* = 72
Ile	50.58 ± 11.60	55.16 ± 12.81**	56.96 ± 13.67***	65.31 ± 17.15***
Leu	125.48 ± 22.54	135.18 ± 23.47***	139.41 ± 24.10***	153.39 ± 28.15***
Val	175.45 ± 34.77	192.22 ± 35.02***	196.96 ± 36.39***	208.67 ± 44.91***
Tyr	68.72 ± 14.92	73.67 ± 13.63**	77.60 ± 17.34***	79.68 ± 17.59***
Trp	29.76 ± 5.41	30.92 ± 4.62	31.64 ± 4.65**	33.75 ± 6.50***
Phe	139.42 ± 47.50	139.33 ± 40.37	140.09 ± 35.97	167.43 ± 49.32***
Glu	34.91 ± 6.40	36.51 ± 5.50*	37.70 ± 5.90***	39.76 ± 7.51***
Asp	6.64 ± 1.78	7.04 ± 1.63	7.03 ± 1.61	7.98 ± 2.29***
Ala	185.69 ± 40.88	209.05 ± 40.20***	216.81 ± 41.42***	236.25 ± 34.73***
His	94.27 ± 26.63	97.23 ± 26.62	102.68 ± 26.98*	107.57 ± 38.77*
Met	42.96 ± 12.09	44.43 ± 10.05	45.77 ± 11.90*	46.89 ± 12.94*
Asn	48.55 ± 12.96	49.12 ± 9.72	51.07 ± 11.56	52.86 ± 14.35*
Pro	304.41 ± 116.67	302.49 ± 75.54	326.38 ± 79.00*	335.00 ± 86.17**
Tau	1.59 ± 0.66	1.57 ± 0.51	1.50 ± 0.51	1.39 ± 0.51**

*All participants have been divided into four groups according to MetS components and the 14 amino acids associated with MetS in each group. One-way analysis of variance was used, compared with the 0-component group; the statistical differences of other groups are shown as *P < 0.05, **P < 0.01, and ***P < 0.001.*
Ile, isoleucine; Leu, leucine; Val, valine; Tyr, tyrosine; Trp, tryptophan; Phe, phenylalanine; Glu, glutamic; Asp, aspartic; Ala, alanine; His, histidine; Met, methionine; Asn, asparagine; Pro, proline; Tau, taurine.

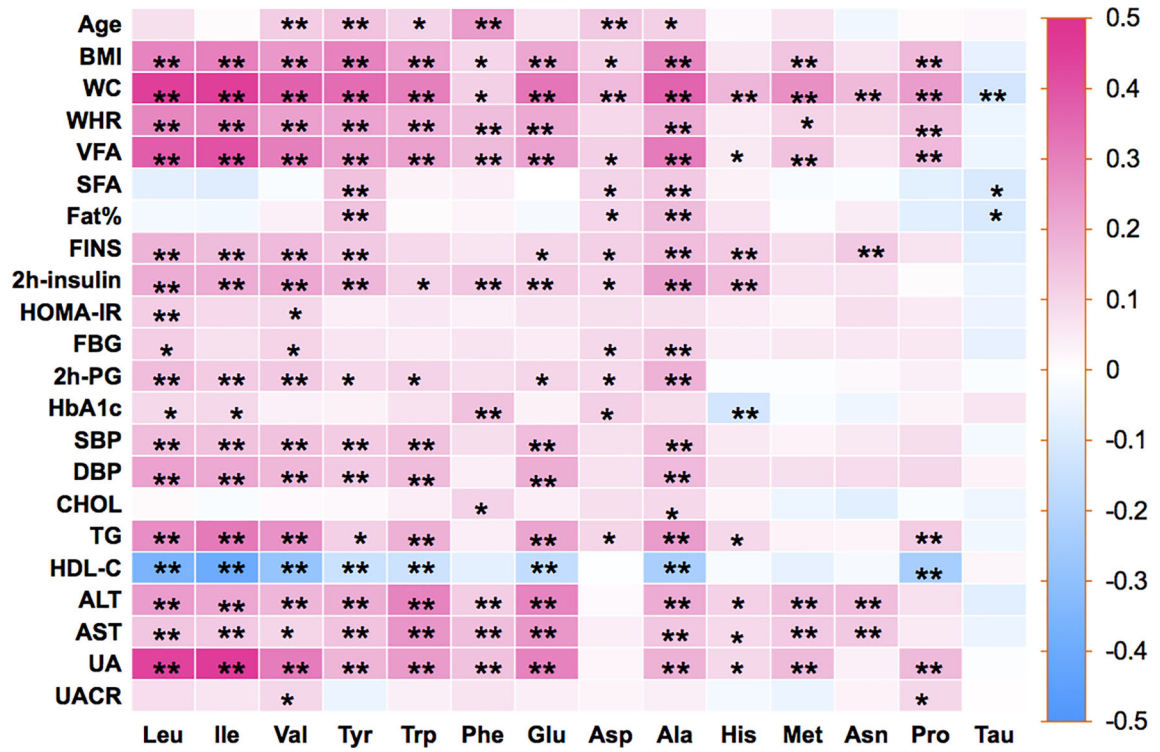


FIGURE 1 | Pearson's correlation coefficients were calculated between 14 amino acids and metabolic-related variables. BMI, body mass index; WC, waist circumference; WHR, waist-to-hip ratio; Fat%, body fat percentage; VFA, visceral fat area; SFA, subcutaneous fat area; FINS, fasting serum insulin; HOMA-IR, homeostasis model assessment for insulin resistance; FPG, fasting blood glucose; 2h-PG, 2-h postprandial glucose; HbA1c, glycosylated hemoglobin A1c; SBP, systolic blood pressure; DBP, diastolic blood pressure; TC, total cholesterol; TG, triglyceride; HDL-c, high-density lipoprotein-cholesterol; ALT, alanine aminotransferase; AST, aspartate aminotransferase; UA, uric acid; UACR, urine albumin-to-creatinine ratio; Ile, isoleucine; Leu, leucine; Val, valine; Tyr, tyrosine; Trp, tryptophan; Phe, phenylalanine; Glu, glutamic; Asp, aspartic; Ala, alanine; His, histidine; Met, methionine; Asn, asparagine; Pro, proline; Tau, taurine. Statistical differences between 14 amino acids and metabolic-related variables are shown as $^{*}P < 0.05$ and $^{**}P < 0.01$.

dyslipidemia, and elevated blood pressure after adjustment for age, gender, current smoking, alcohol drinking, and moderate exercise ($P < 0.05$; **Table 5**). Additionally, we identified "PC2"—a Tau-related amino acid profile—as being associated with MetS, abnormal glucose, and dyslipidemia ($P < 0.05$; **Table 5**). PC3 was unrelated to MetS.

Amino Acid Profiles Were Associated With Future Development of MetS Within 5 Years

To confirm the correlation of amino acid profiles and future development of MetS, we followed up 401 non-MetS. Finally, 260 participants were followed up effectively from 2010 until 2015 (another 141 participants were lost to follow-up due to moving to another house, requesting to withdraw from the study, etc.), and 42 participants developed new-onset MetS. The basal clinical and biochemical characteristics were compared according to whether participants developed new-onset MetS or not within 5 years. We found that the baselines of BMI, WC, WHR, Fat%, VFA, SBP, FINS, 2h-insulin, HOMA-IR, TG, and UA were significantly higher in the subsequent new-onset MetS group than in the subsequent non-MetS group, while HDL-c was significantly lower ($P < 0.05$; **Table 6**). Ile, Leu, Tyr, and Ala were elevated, and Tau was reduced in the new-onset MetS group at baseline, exhibiting a statistically significant difference ($P < 0.05$; **Table 7**). After adjusting for age, gender, current smoking, alcohol drinking, and moderate exercise, the "PC1" and Tau were significantly correlated with MetS ($P < 0.05$). After adjusting for BMI, WC, WHR, Fat%, VFA, SBP, FINS, 2h-insulin, TG, HDL-c, and UA, Tau was significantly negatively correlated with MetS ($P = 0.016$). Decreased Tau could be associated with future development of MetS within 5 years.

DISCUSSION

Using the 2009 diagnostic criteria of the IDF and AHA/NHLBI, we examined the association between amino acids and MetS. First, we found evidence that amino acid profiles are beneficial in identifying individuals who are at high risk of MetS in a Chinese Han population. It has been demonstrated that increased levels of Ile, Leu, Val, Tyr, Trp, Phe, Glu, Asp, Ala, His, Met, Asn, and Pro and decreased Tau levels were linked to an increased risk of MetS and its components.

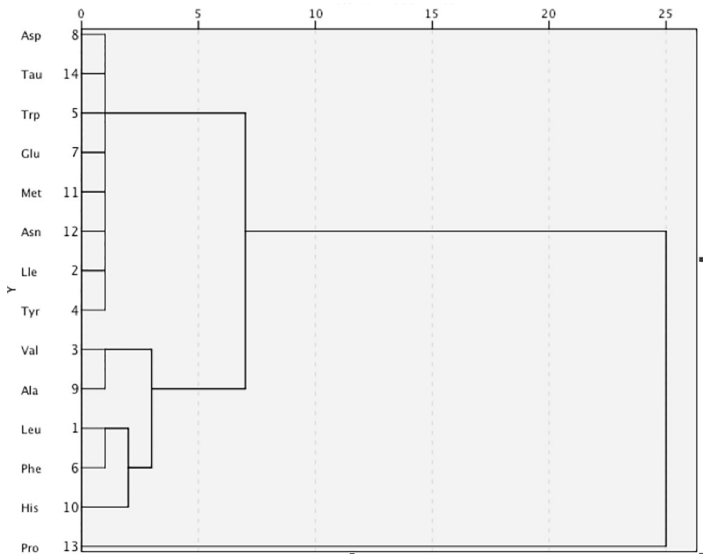


FIGURE 2 | The side dendrogram represents the hierarchical clustering of the 14 amino acids based on Pearson correlation, and we can see a strong correlation with the amino acids. Ile, isoleucine; Leu, leucine; Val, valine; Tyr, tyrosine; Trp, tryptophan; Phe, phenylalanine; Glu, glutamic; Asp, aspartic; Ala, alanine; His, histidine; Met, methionine; Asn, asparagine; Pro, proline; Tau, taurine.

As revealed in this study, we have stated that BCAAs, AAAs, Glu, and Ala were positively associated with BMI, WC, WHR, VFA, FINS, 2h-insulin, 2h-PG, SBP, DBP, TG, and UA and negatively associated with HDL-c. Our study also discovered that His was positively associated with WC and insulin resistance. Met and Pro were positively linked to BMI, WC, and VFA. Gly and Cys did not differ significantly in the Chinese Han population in this study. However, we discovered that Tau was significantly lower in MetS and was negatively correlated with metabolic-related variables. Numerous studies with rats, mice, and rabbits revealed that Tau effectively reduces TC, TG, blood glucose, and blood pressure (35–39).

TABLE 4 | The result from principal component analysis exploring amino acid profiles.

	PC		
	1	2	3
His	**0.545**	**–0.654**	0.027
Ile	**0.761**	0.359	–0.224
Leu	**0.854**	0.321	–0.103
Met	**0.688**	–0.369	–0.075
Phe	0.083	0.373	**0.746**
Trp	**0.815**	0.116	0.115
Val	**0.750**	0.248	–0.166
Tau	–0.066	**0.435**	0.264
Tyr	**0.722**	0.001	–0.105
Asn	**0.675**	**–0.541**	0.181
Asp	**0.616**	–0.124	**0.600**
Glu	**0.828**	0.071	0.036
Ala	**0.701**	0.36	–0.034
Pro	**0.459**	0.295	–0.307
% of variance	43.289	11.438	8.826

Extraction method: principal component analysis (PCA) with varimax rotation; items with a loading ≥ |0.4| were reported as composing a given factor (bold font type).

While interethnic differences in the pathophysiology of insulin dysregulation and visceral obesity between Asian and Western populations are well known, few studies have examined the relationship between amino acids and lifestyle-related disease risks in Chinese populations (40). Our research has extracted "PC1" using PCA. It was the dominant factor (accounting for 43.29% of the variance in PCA) and was mostly constituted of BCAAs, Tyr, Trp, Glu, Asp, Asn, Ala, His, Met, and Pro. After adjusting for age, gender, smoking, drinking, and moderate exercise, the amino acid profile of PC1 was linked to abdominal obesity, abnormal glucose, dyslipidemia, elevated blood pressure, and MetS. After a 5-year follow-up, this amino acid profile remained significantly correlated with the future incidence of MetS within 5 years. BCAAs and AAAs (except Phe) exhibited the highest loadings in PC1, consistent with previous research from other countries indicating that BCAA/AAA pattern was connected with MetS and its components. However, the correlation between Phe and MetS is not strong in Chinese Han populations. Yamakado et al. (11) used a method other than PCA to calculate a plasma free amino acid (PFAA) index but also discovered that indices that included BCAAs and AAAs were positively associated with VFA and circulating insulin levels in cross-sectional analyses, confirming that plasma free amino acid profiles could predict the future development of DM, MetS, and dyslipidemia in the Japanese population, even within a relatively short period (4 years). Other studies performed a systems metabolomics approach to predict MetS development, but these predictive models are specific to the T2DM component of MetS. Therefore, the prediction of MetS subjects without a T2DM component reveals a high rate of misclassified subjects (around 30%), implying poor prediction

TABLE 5 | The odds ratios for abdominal obesity, abnormal glucose, dyslipidemia, elevated blood pressure, and MetS adjusting for additional factors.

	PC1	*P*-value	PC2	*P*-value
	OR (95% CI)		OR (95% CI)	
Abdominal obesity	1.499 (1.283–1.752)	<0.001***	0.922 (0.776–1.094)	0.352
Abnormal glucose	1.264 (1.077–1.483)	0.004**	1.332 (1.082–1.641)	0.007**
Dyslipidemia	1.339 (1.168–1.535)	<0.001***	1.238 (1.052–1.457)	0.01*
Elevated blood pressure	1.154 (1.011–1.316)	0.033*	0.954 (0.813–1.12)	0.566
MetS	1.723 (1.424–2.085)	<0.001***	1.325 (1.043–1.684)	0.021*

*Values are odds ratios (95% confidence intervals) as a continuous variable per SD increment for developing from abdominal obesity, abnormal glucose, dyslipidemia, abnormal blood pressure, and MetS logistic regressions. Adjustment for age, gender, current smoking, alcohol drinking, and physical activity level. Statistical differences between disease and non-disease subjects are shown as *P < 0.05, **P < 0.01, and ***P < 0.001. PC1 (isoleucine, leucine, valine, tryptophan, tyrosine, histidine, methionine, aspartic, asparagine, glutamic, alanine, proline); PC2 (taurine, histidine, asparagine).*

MetS, metabolic syndrome; OR, odds ratio.

capacity of MetS hypertension and dyslipidemia components (41). Our amino acid profile was associated with MetS and its components (abdominal obesity, abnormal glucose, dyslipidemia, and elevated blood pressure). It can be employed as a potential biomarker for assessing MetS risk.

The amino acid profile, which mainly included Tau, His, and Asn, denoted "PC2," accounted for 11.44% of PCA variance. This amino acid profile was associated with abnormal glucose, dyslipidemia, and MetS after adjustment for age, gender, smoking, drinking, and moderate exercise. After a 5-year follow-up, PC2 exhibited no significant difference with future development of MetS. However, Tau was significantly negatively correlated with MetS after adjusting for age, gender, smoking, drinking, exercise, BMI, WC, Fat%, VFA, SBP, FINS, TG, HDL-c,

TABLE 6 | Comparison of the baseline of general clinical characteristics and biochemical indicators between 42 subsequent new MetS and 218 subsequent non-MetS.

Variables	Subsequent non-MetS *n* = 218	Subsequent new MetS *n* = 42	*P*-value
Female, *N* (%)	131 (60.09%)	20 (47.62%)	0.198
Current smoker, *N* (%)	48 (22.02%)	11 (26.19%)	0.531
Alcohol drinker, *N* (%)	88 (40.37%)	18 (42.86%)	0.749
Moderate exercise, *N* (%)	107 (49.08%)	17 (40.48%)	0.334
Age (years)	53.01 ± 6.34	53.49 ± 7.46	0.856
BMI (kg/m^2)	22.44 ± 2.47	25.07 ± 1.96	<0.001*
WC (cm)	75.35 ± 7.71	83.19 ± 6.04	<0.001*
WHR	0.85 ± 0.06	0.90 ± 0.05	<0.001*
Fat% (%)	27.58 ± 6.76	30.95 ± 5.06	0.007*
SFA (cm^2)	141.3 (110.3, 189.35)	155.3 (126.6, 203.9)	0.112
VFA (cm^2)	63.66 (36.42, 89.54)	104.5 (66.68, 128.75)	<0.001*
SBP (mmHg)	119.27 ± 14.90	125.50 ± 15.26	0.023*
DBP (mmHg)	80.65 ± 16.08	84.01 ± 9.35	0.268
FPG (mmol/L)	4.95 ± 0.84	5.09 ± 0.97	0.361
2h-PG (mmol/L)	5.91 ± 2.79	6.55 ± 3.39	0.225
FINS (mIU/L)	10.39 (6.87, 13.33)	13.67 (8.61, 18.64)	<0.001*
2 h insulin (mIU/L)	37.25 (24.60, 60.30)	56.29 (32.17, 87.59)	0.034*
HOMA-IR	2.13 (1.49, 3.07)	3.09 (1.99, 4.20)	<0.001*
HbA1c (%)	5.65 ± 0.63	5.61 ± 0.59	0.689
CHOL (mmol/L)	5.52 ± 1.09	5.58 ± 0.85	0.738
TG (mmol/L)	1.19 (0.80, 1.54)	1.62 (1.26, 2.47)	0.002*
LDL-c (mmol/L)	2.41 ± 0.56	2.40 ± 0.53	0.948
HDL-c (mmol/L)	1.51 ± 0.38	1.28 ± 0.34	0.001*
ALT (IU/L)	19 (14, 26)	21 (16, 27.75)	0.657
AST (IU/L)	20 (17, 23)	20 (17.25, 24.75)	0.688
CREA (mg/dl)	0.80 ± 0.16	0.84 ± 0.26	0.709
BUN (mmol/L)	16.76 ± 3.59	16.11 ± 3.55	0.318
UA (mg/dl)	4.58 ± 1.39	5.34 ± 1.84	0.002*
UACR (mg/mmol)	4.43 (2.79, 12.09)	4.59 (3.13, 7.03)	0.15

*After a 5-year follow-up, 260 non-MetS were followed up effectively, of which 42 developed new MetS. Statistical differences between subsequent new MetS and subsequent non-MetS are shown as *P < 0.05.*

MetS, metabolic syndrome; BMI, body mass index; WC, waist circumference; WHR, waist-to-hip ratio; Fat%, body fat percentage; SFA, subcutaneous fat area; VFA, visceral fat area; SBP, systolic blood pressure; DBP, diastolic blood pressure; FPG, fasting blood glucose; 2h-PG, 2-h postprandial glucose; FINS, fasting serum insulin; HOMA-IR, homeostasis model assessment for insulin resistance; HbA1c, glycosylated hemoglobin A1c; TC, total cholesterol; TG, triglyceride; LDL-c, low-density lipoprotein-cholesterol; HDL-c, high-density lipoprotein-cholesterol; ALT, alanine aminotransferase; AST, aspartate aminotransferase; CREA, serum creatinine; BUN, serum urea nitrogen; UA, uric acid; UACR, urine albumin-to-creatinine ratio.

TABLE 7 | The baseline of plasma amino acid concentrations in subsequent new MetS and subsequent non-MetS groups.

	Subsequent non-MetS (n=218)	Subsequent new MetS (n=42)	*P*-value	OR (95% CI)
Leu	130.46 ± 23.15	141.46 ± 21.83	0.009*	1.020 (1.005–1.036)
Ile	53.25 ± 12.83	59.29 ± 11.39	0.01*	1.036 (1.009–1.064)
Val	186.17 ± 35.74	195.97 ± 36.24	0.13	1.008 (0.998–1.017)
Tyr	70.18 ± 13.50	76.20 ± 14.14	0.017*	1.031 (1.015–1.046)
Trp	30.11 ± 5.01	30.80 ± 4.37	0.437	1.028 (0.958–1.104)
Phe	141.11 ± 41.19	149.74 ± 38.17	0.122	0.993 (0.983–1.002)
Glu	35.59 ± 6.06	37.03 ± 5.60	0.182	1.040 (0.982–1.101)
Asp	6.71 ± 1.58	7.15 ± 1.90	0.134	1.169 (0.953–1.433)
Ala	196.27 ± 40.33	211.39 ± 43.24	0.042*	1.009 (1.000–1.018)
His	97.19 ± 26.77	103.88 ± 24.99	0.162	1.009 (0.996–1.022)
Met	42.89 ± 9.76	46.25 ± 11.03	0.063	1.033 (0.998–1.069)
Asn	48.63 ± 11.02	50.74 ± 8.81	0.272	1.018 (0.986–1.051)
Pro	300.04 ± 94.72	309.83 ± 65.48	0.551	1.001 (0.998–1.004)
Tau	1.68 ± 0.59	1.37 ± 0.53	0.003*	0.325 (0.153–0.686)

*Statistical differences between subsequent new MetS and subsequent non-MetS are shown as *P < 0.05.*
Ile, isoleucine; Leu, leucine; Val, valine; Tyr, tyrosine; Trp, tryptophan; Phe, phenylalanine; Glu, glutamic; Asp, aspartic; Ala, alanine; His, histidine; Met, methionine; Asn, asparagine; Pro, proline; Tau, taurine.

and UA. Decreased Tau could be linked with the future development of MetS within 5 years. Other investigations have demonstrated that patients with obesity (42) and diabetes (43) have lower Tau levels in their bodies, consistent with our finding that Tau decreased in MetS. The decrease in blood Tau levels was associated with a decrease in cysteine dioxygenase expression, a rate-limiting enzyme in taurine synthesis (42). In contrast, Tau supplementation increases plasma Tau levels, reduces plasma levels of inflammatory and oxidative markers, and increases plasma adiponectin levels in humans. Research studies have indicated that Tau prevents obesity mainly due to increasing energy expenditure by upregulating relative factor expression involved in fatty acid oxidation. It also prevents hypercholesterolemia by promoting bioconversion of cholesterol to bile acids, promoting bile acid excretion in feces, and suppressing bile acid absorption from the ileum. It also prevents diabetes mellitus by exerting antioxidant and anti-inflammatory effects, protecting pancreatic β cells, promoting insulin secretion, improving insulin resistance by inhibiting JNK1 activation, improving insulin signaling in the liver, and preventing hypertension by suppressing RAAS. Tau may be beneficial for preventing MetS (30, 44, 45).

CONCLUSION

This cross-sectional study of a cohort investigated the correlation between amino acids and MetS in a Chinese Han population. In the present study, we extracted the amino acid profile using PCA, which can be used as biomarkers to assess and monitor MetS risk. Tau may be beneficial for preventing MetS. Decreased Tau levels were associated with the future development of MetS. However, this study has shortcomings, such as that it only involved participants aged 40–65 years old and the number of samples with clinical follow-up was small, implying that more samples are required to verify these results.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding authors.

ETHICS STATEMENT

Written informed consent was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

AUTHOR CONTRIBUTIONS

All authors listed have made a substantial, direct, and intellectual contribution to the work and approved it for publication.

FUNDING

This work was supported by the Program for Zhejiang Leading Team of Science and Technology Innovation (2012R10050-03).

REFERENCES

1. Ford ES. Risks for All-Cause Mortality, Cardiovascular Disease, and Diabetes Associated With the Metabolic Syndrome - A Summary of the Evidence. *Diabetes Care* (2005) 28:1769–78. doi: 10.2337/diacare.28.7.1769
2. Galassi A, Reynolds K, He J. Metabolic Syndrome and Risk of Cardiovascular Disease: A Meta-Analysis. *Am J Med* (2006) 119:812–9. doi: 10.1016/j.amjmed.2006.02.031
3. Chen W, Pan Y, Jing J, Zhao X, Liu L, Meng X, et al. Recurrent Stroke in Minor Ischemic Stroke or Transient Ischemic Attack With Metabolic

Syndrome and/or Diabetes Mellitus. *J Am Heart Assoc* (2017) 6:e005446. doi: 10.1161/JAHA.116.005446
4. Razay G, Vreugdenhil A, Wilcock G. The Metabolic Syndrome and Alzheimer Disease. *Arch Neurol* (2017) 64:93–6. doi: 10.1001/archneur.64.1.93
5. Samson SL, Garber AJ. Metabolic Syndrome. *Endocrinol Metab Clin North Am* (2014) 43:1–23. doi: 10.1016/j.ecl.2013.09.009
6. Nagao K, Yamakado M. The Role of Amino Acid Profiles in Diabetes Risk Assessment. *Curr Opin Clin Nutr Metab Care* (2016) 19:328–35. doi: 10.1097/MCO.0000000000000305
7. Chen T, Ni Y, Ma X, Bao Y, Liu J, Huang F, et al. Branched-Chain and Aromatic Amino Acid Profiles and Diabetes Risk in Chinese Populations. *Sci Rep* (2016) 6:20594. doi: 10.1038/srep20594
8. Wiklund P, Zhang X, Pekkala S, Autio R, Kong L, Yang Y, et al. Insulin Resistance Is Associated With Altered Amino Acid Metabolism and Adipose Tissue Dysfunction in Normoglycemic Women. *Sci Rep* (2016) 6:24540. doi: 10.1038/srep24540
9. Zhao X, Han Q, Liu Y, Sun C, Gang X, Wang G. The Relationship Between Branched-Chain Amino Acid Related Metabolomic Signature and Insulin Resistance: A Systematic Review. *J Diabetes Res* (2016) 2016:1–12. doi: 10.1155/2016/2794591
10. Zakaria NF, Hamid M, Khayat ME. Amino Acid-Induced Impairment of Insulin Signaling and Involvement of G-Protein Coupling Receptor. *Nutrients* (2021) 13:2229. doi: 10.3390/nu13072229
11. Yamakado M, Nagao K, Imaizumi A, Tani M, Toda A, Tanaka T, et al. Plasma Free Amino Acid Profiles Predict Four-Year Risk of Developing Diabetes, Metabolic Syndrome, Dyslipidemia, and Hypertension in Japanese Population. *Sci Rep* (2015) 5:11918. doi: 10.1038/srep11918
12. Cheng S, Rhee EP, Larson MG, Lewis GD, McCabe EL, Shen D, et al. Metabolite Profiling Identifies Pathways Associated With Metabolic Risk in Humans. *Circulation* (2012) 125:2222–31. doi: 10.1161/CIRCULATIONAHA.111.067827
13. Würtz P, Soininen P, Kangas AJ, Rönnemaa T, Lehtimäki T, Kähönen M, et al. Branched-Chain and Aromatic Amino Acids Are Predictors of Insulin Resistance in Young Adults. *Diabetes Care* (2013) 36:648–55. doi: 10.2337/dc12-0895
14. Ntzouvani A, Nomikos T, Panagiotakos D, Fragopoulou E, Pitsavos C, McCann A, et al. Amino Acid Profile and Metabolic Syndrome in a Male Mediterranean Population: A Cross-Sectional Study. *Nutr Metab Cardiovasc Dis* (2017) 27:1021–30. doi: 10.1016/j.numecd.2017.07.006
15. Pujos-Guillot E, Brandolini M, Pétéra M, Grissa D, Joly C, Lyan B, et al. Systems Metabolomics for Prediction of Metabolic Syndrome. *J Proteome Res* (2017) 16:2262–72. doi: 10.1021/acs.jproteome.7b00116
16. Newgard CB. Interplay Between Lipids and Branched-Chain Amino Acids in Development of Insulin Resistance. *Cell Metab* (2012) 15:606–14. doi: 10.1016/j.cmet. 2012.01.024
17. Carraro V, Maurin A-C, Lambert-Langlais S, Averous J, Chaveroux C, Parry L, et al. Amino Acid Availability Controls TRB3 Transcription in Liver Through the GCN2/eIF2alpha/ATF4 Pathway. *PLoS One* (2010) 5:e15716. doi: 10.1371/journal.pone.0015716
18. Lynch CJ. Role of Leucine in the Regulation of mTOR by Amino Acids: Revelations From Structure-Activity Studies. *J Nutr* (2001) 131:861S–5S. doi: 10.1093/jn/131.3.861S
19. Leclerc I, Rutter GA. AMP-Activated Protein Kinase: A New Beta-Cell Glucose Sensor?: Regulation by Amino Acids and Calcium Ions. *Diabetes* (2004) 53:S67–74. doi: 10.2337/diabetes.53.suppl_3.s67
20. Tremblay F, Marette A. Amino Acid and Insulin Signaling *via* the mTOR/P70 S6 Kinase Pathway. A Negative Feedback Mechanism Leading to Insulin Resistance in Skeletal Muscle Cells. *J Biol Chem* (2001) 276:38052–60. doi: 10.1074/jbc.M106703200
21. Adams SH. Emerging Perspectives on Essential Amino Acid Metabolism in Obesity and the Insulin-Resistant State. *Adv Nutr* (2011) 2:445–56. doi: 10.3945/an.111.000737
22. Newgard CB, An J, Bain JR, Muehlbauer MJ, Stevens RD, Lien LF, et al. A Branched-Chain Amino Acid-Related Metabolic Signature That Differentiates Obese and Lean Humans and Contributes to Insulin Resistance. *Cell Metab* (2009) 9:311–26. doi: 10.1016/j.cmet.2009.02.002
23. Ho JE, Larson MG, Ghorbani A, Cheng S, Chen MH, Keyes M, et al. 32 Metabolomic Profiles of Body Mass Index in the Framingham Heart Study Reveal Distinct Cardiometabolic Phenotypes. *PLoS One* (2016) 11:e0148361. doi: 10.1371/journal.pone.0148361
24. Yamada C, Kondo M, Kishimoto N, Shibata T, Nagai Y, Imanishi T, et al. Association Between Insulin Resistance and Plasma Amino Acid Profile in Non-Diabetic Japanese Subjects. *J Diabetes Investig* (2015) 6:408–15. doi: 10.1111/jdi.12323
25. Zhang M, Bi LF, Fang JH, Su XL, Da GL, Kuwamori T, et al. Beneficial Effects of Taurine on Serum Lipids in Overweight or Obese Non-Diabetic Subjects. *Amino Acids* (2004) 26:267–71. doi: 10.1007/s00726-003-0059-z
26. Yokogoshi H, Mochizuki H, Nanami K, Hida Y, Miyachi F, Oda H. Dietary Taurine Enhances Cholesterol Degradation and Reduces Serum and Liver Cholesterol Concentrations in Rats Fed a High-Cholesterol Diet. *J Nutr* (1999) 129:1705–12. doi: 10.1093/jn/129.9.1705
27. El Mesallamy HO, El-Demerdash E, Hammad LN, El Magdoub HM. Effect of Taurine Supplementation on Hyperhomocysteinemia and Markers of Oxidative Stress in High Fructose Diet Induced Insulin Resistance. *Diabetol Metab Syndr* (2010) 2:46–57. doi: 10.1186/1758-5996-2-46
28. Nandhini AT, Thirunavukkarasu V, Anuradha CV. Potential Role of Kinins in the Effects of Taurine in High-Fructose-Fed Rats. *Can J Physiol Pharm* (2004) 82:1–8. doi: 10.1139/y03-118
29. Xiao C, Giacca A, Lewis GF. Oral Taurine But Not N-Acetylcysteine Ameliorates NEFA-Induced Impairment in Insulin Sensitivity and Beta Cell Function in Obese and Overweight, non-Diabetic Men. *Diabetologia* (2008) 51:139–46. doi: 10.1007/s00125-007-0859-x
30. Wu N, Lu Y, He B, Zhang Y, Lin J, Zhao S. Taurine Prevents Free Fatty Acid-Induced Hepatic Insulin Resistance in Association With Inhibiting JNK1 Activation and Improving Insulin Signaling *In Vivo*. *Diabetes Res Clin Pract* (2010) 90:288–96. doi: 10.1016/j.diabres.2010.08.020
31. Alberti KGMM, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA, et al. Harmonizing the Metabolic Syndrome: A Joint Interim Statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity. *Circulation* (2009) 120:1640–5. doi: 10.1161/CIRCULATIONAHA.109.192644
32. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis Model Assessment: Insulin Resistance and Beta-Cell Function From Fasting Plasma Glucose and Insulin Concentrations in Man. *Diabetologia* (1985) 28:412–9. doi: 10.1007/BF00280883
33. Midttun Ø, McCann A, Aarseth O, Krokeide M, Kvalheim G, Meyer K, et al. Combined Measurement of 6 Fat-Soluble Vitamins and 26 Water-Soluble Functional Vitamin Markers and Amino Acids in 50 µl of Serum or Plasma by High-Throughput Mass Spectrometry. *Anal Chem* (2016) 88:10427–36. doi: 10.1021/acs.analchem.6b02325
34. Virág D, Király M, Drahos L, Édes AE, Gecse K, Bagdy G, et al. Development, Validation and Application of LC-MS/MS Method for Quantification of Amino Acids, Kynurenine and Serotonin in Human Plasma. *J Pharm BioMed Anal* (2020) 180:113018. doi: 10.1016/j.jpba.2019.113018
35. Chen W, Suruga K, Nishimura N, Gouda T. Comparative Regulation of Major Enzymes in Bile Acids Biosynthesis Pathways by Cholesterol, Cholic Acid and Taurine in Mice and Rats. *Life Sci* (2005) 77:746–57. doi: 10.1016/j.lfs.2004.11.036
36. El-Batch M, Hassan AM, Mahmoud HA. Taurine is More Effective Than Melatonin on Cytochrome P450 2E1 and Some Oxidative Stress Markers in Streptozotocin- Induced Diabetic Rats. *J Agr Food Chem* (2011) 59:4995–5000. doi: 10.1021/jf1049547
37. Gavrovskaya LK, Ryzhova OV, Safonova AF, Matveev AK, Sapronov NS. Protective Effect of Taurine on Rats With Experimental Insulin-Dependent Diabetes Mellitus. *B Exp Biol Med* (2008) 146:226–8. doi: 10.1186/1475-2840-11-129
38. Imae M, Asano T, Murakami S. Potential Role of Taurine in the Prevention of Diabetes and Metabolic Syndrome. *Amino Acids* (2014) 46:81–8. doi: 10.1007/s00726-012-1434-4
39. Murakami S. Taurine and Atherosclerosis. *Amino Acids* (2014) 46:73–80. doi: 10.1007/s00726-012-1432-6
40. Yamori Y, Taguchi T, Mori H, Mori M. Low Cardiovascular Risks in the Middle Aged Males and Females Excreting Greater 24-Hour Urinary Taurine and Magnesium in WHO-CARDIAC Study Populations in the World. *J BioMed Sci* (2010) 17:S21. doi: 10.1186/1423-0127-17-S1-S21

41. Yamaguchi N, Mahbub MH, Takahashi H, Hase R, Ishimaru Y, Sunagawa H, et al. Plasma Free Amino Acid Profiles Evaluate Risk of Metabolic Syndrome, Diabetes, Dyslipidemia, and Hypertension in a Large Asian Population. *Environ Health Prev Med* (2017) 22:35. doi: 10.1186/s12199-017-0642-7
42. Rosa FT, Freitas EC, Deminice R, Jordão AA, Marchini JS. Oxidative Stress and Inflammation in Obesity After Tau- Rine Supplementation: A Double-Blind, Placebo-Controlled Study. *Eur J Nutr* (2014) 53:823–30. doi: 10.1007/s00394-013-0586-7
43. Merheb M, Daher RT, Nasrallah M, Sabra R, Ziyadeh FN, Barada K. Taurine Intestinal Absorption and Renal Excretion Test in Diabetic Patients: A Pilot Study. *Diabetes Care* (2007) 30:2652–4. doi: 10.2337/dc07-0872
44. Chen W, Guo JX, Zhang YZ, Zhang J. The Beneficial Effects of Taurine in Preventing Metabolic Syndrome. *Food Funct* (2016) 7:1849–63. doi: 10.1039/c5fo01295c
45. Ma RC, Chan JC. Type 2 Diabetes in East Asians: Similarities and Differences With Populations in Europe and the United States. *Ann N Y Acad Sci* (2013) 1281:64–91. doi: 10.1111/nyas.12098

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 16 March 2022
doi: 10.3389/fendo.2022.850334

Perirenal Fat Thickness: A Surrogate Marker for Metabolic Syndrome in Chinese Newly Diagnosed Type 2 Diabetes

Xiu Li Guo[1], *Mei Tu*[2], *Yang Chen*[2] *and Wei Wang*[2]*

[1] Department of Radiology, Longyan First Affiliated Hospital of Fujian Medical University, Longyan, China, [2] Department of Endocrinology, Longyan First Affiliated Hospital of Fujian Medical University, Longyan, China

Objective: Increasing evidence suggested that perirenal fat thickness (PrFT) was associated with metabolic risk factors. This study aimed to assess the association between PrFT and metabolic syndrome (MetS) in Chinese newly diagnosed type 2 diabetes (T2DM), further evaluating the ability of PrFT in identifying MetS.

Method: A total of 445 Chinese newly diagnosed T2DM were enrolled in this study from January to June 2021. Demographic and anthropometric information were collected. PrFT was evaluated by CT scan on Revolution VCT 256. MetS was based on the Chinese Diabetes Society definition. Receiver operating characteristic (ROC) curve was conducted to assess the optimal cutoff value of PrFT in identifying MetS.

Results: Overall, the prevalence of MetS was 57.5% (95% CI: 54.0–64.0%) in men and 58.9% (95% CI: 52.3–65.5%) in women separately. The correlation analysis showed that PrFT was significantly correlated with metabolic risk factors like body mass index, waist circumference, triglycerides, high-density lipoprotein cholesterol, systolic blood pressure, diastolic blood pressure, uric acid, and insulin resistance. PrFT was also shown to be independently associated with MetS after adjustment for other confounders. The odds ratios (ORs, 95% CI) were 1.15 (1.03–1.38) in men and 1.31 (1.08–1.96) in women ($P < 0.05$). The ROC curves showed a good predictive value of PrFT for MetS. The areas under the curve of PrFT identifying MetS were 0.895 (95% CI: 0.852–0.939) in men and 0.910 (95% CI: 0.876–0.953) in women ($P < 0.001$). The optimal cutoff values of PrFT were 14.6 mm (sensitivity: 83.8%, specificity: 89.6%) for men and 13.1 mm (sensitivity: 87.6%, specificity: 91.1%) for women.

Conclusions: PrFT was significantly associated with MetS and showed a powerful predictive value for it, which suggested that PrFT can be an applicable surrogate marker for MetS in Chinese newly diagnosed T2DM.

Clinical Trial Registration: This study was registered in clinicaltrials.gov (ChiCTR2100052032).

Keywords: perirenal fat thickness, metabolic syndrome, newly diagnosed type 2 diabetes, optimal cut-off value, visceral adipose tissue

OPEN ACCESS

Edited by:
Ozra Tabatabaei-Malazy, Tehran University of Medical Sciences, Iran

Reviewed by:
Sahar Saeedi Moghaddam, Tehran University of Medical Sciences, Iran
Kazem Khalagi, Tehran University of Medical Sciences, Iran

***Correspondence:**
Wei Wang
591187650@qq.com

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *07 January 2022*
Accepted: *07 February 2022*
Published: *16 March 2022*

Citation:
Guo XL, Tu M, Chen Y and Wang W (2022) PerirenalFat Thickness: A Surrogate Marker for Metabolic Syndrome in Chinese Newly Diagnosed Type 2 Diabetes. Front. Endocrinol. 13:850334. doi: 10.3389/fendo.2022.850334

INTRODUCTION

Metabolic syndrome (MetS) is the common pathological basis and early stage of many major diseases that is characterized by the simultaneous presence of obesity, hypertension, dyslipidemia, and hyperglycemia in individuals, leading to increased prevalence of cardiovascular disease (CVD) and stroke and risk of diabetes (1). Type 2 diabetes (T2DM) is a kind of metabolic disease characterized by chronic hyperglycemia due to the failure of pancreatic islet β-cells to sustain the hyperinsulinemia required to compensate for insulin resistance and often accompanied with other metabolic disorders. The prevalence of MetS has rapidly increased in China. A cross-sectional survey reported the prevalence of MetS in diabetes which is in the range from 53 to 68.1% (2). Due to the great harm and high prevalence of MetS in newly diagnosed T2DM, early diagnosis is urgently needed, whereas the diagnosis and awareness rate are suboptimal in clinical diagnosis and treatment (3). The diagnostic process for MetS in patients with diabetes is cumbersome and thus may limit the early diagnosis of MetS (4). An effective surrogate marker for MetS can help clinicians in identifying MetS in newly diagnosed T2DM.

Visceral adipose tissue is considered to be a type of "ectopic fat" which has adverse influences on systemic inflammation, insulin resistance, and metabolic profiles and, finally, increases the risk of developing MetS and CVD (5–7). Among visceral adipose tissue deposits, perirenal fat is located around and enclosed from the inner side of the abdominal musculature to the surface of the kidney, which can be easily measured by ultrasound, CT, and MRI scanning (8). Anatomical studies demonstrated that perirenal fat may modulate the metabolism system through neural reflexes, adipokine secretion, and adipocyte interactions due to its unique structure compared with other connective tissues (9–11). Thus, these features may provide a basis for the involvement of perirenal fat in MetS regulation. Cross-sectional studies also observed that perirenal fat thickness (PrFT) is associated with the components of MetS, such as hypertension, obesity, and dyslipidemia (12, 13). Based on the above-mentioned anatomical and cross-sectional studies, it may indicate to us that PrFT can be a surrogate marker for MetS. Hence, we design a cross-sectional study to assess the association between PrFT and MetS in Chinese newly diagnosed T2DM, further evaluating the ability of PrFT in identifying MetS.

STUDY DESIGN AND METHODS

Study Design and Participants

This cross-sectional study consecutively enrolled individuals from the Department of Endocrinology Clinic who were screened for diabetes at the Longyan First Affiliated Hospital of Fujian Medical University and who fulfilled the study criteria between January 2021 and June 2021. The study inclusion criteria were as follows: (1) newly diagnosed T2DM using the World Health Organization (WHO) 2019 criteria (fasting plasma glucose ≥126 mg/dl or 2-h postprandial ≥200 mg/dl during oral glucose tolerance test (OGTT) or HbA1C ≥6.5% or a patient with classic symptoms of hyperglycemia or hyperglycemic crisis or with random plasma glucose ≥200 mg/dl and (2) autoimmune antibodies like glutamic acid decarboxylase antibody (GADA), insulin autoantibody (IAA), and islet cell autoantibody (ICA) negative. Participants were excluded if they were any of the following cases: (1) pregnancy at diagnosis or gestational diabetes mellitus, (2) secondary or special type of diabetes, (3) presence of acute diseases that could interfere with glucose metabolism, (4) with renal structure abnormalities (tumors and cysts or history of renal region surgery), and (5) currently receiving lipid-lowering therapies. In this study, we estimated the sample size according to the requirement of multiple binomial logistic regression model; 10–12 variables may be put into the logistic regression model according to the principle of 5–10 events per variable (14), and the prevalence of MeTS is about 53 to 68.1% (2). Thus, we planned a sampling size of 400–500 patients. The definition of newly diagnosed T2DM is as follows: previous unknown hyperglycemia status and diagnosed with T2DM for the first time (15). Metabolic and hormonal parameters were assessed, and demographic and anthropometric information were evaluated. Then, a CT scan was performed on all participants to measure PrFT. All procedures were conducted in accordance with the Declaration of Helsinki. This study was approved by the Ethical Committee of Longyan First Affiliated Hospital of Fujian Medical University (LY-2020–069) and registered in clinicaltrials.gov (ChiCTR2100052032). All participants enrolled in the study provided informed consent.

Definition of Metabolic Syndrome

MetS was defined according to the Chinese guideline for diabetes with MetS management (16). Patients who meet three or more of the following criteria are considered to have MetS: (1) abdominal obesity: waist circumference (WC) ≥90 cm in men or ≥85 cm in women, (2) hyperglycemia: fasting blood glucose (FBG) ≥6.1 mmol/L or (OGTT) 2-h blood glucose ≥7.8 mmol/L or previously diagnosed diabetes with treatment, (3) hypertension: blood pressure ≥130/85 mmHg or currently under anti-hypertension therapy, (4) fasting triglycerides (TGs) ≥1.70 mmol/L (without lipid-lowering therapies), (5) fasting high-density lipoprotein cholesterol (HDL-c) <1.04 mmol/L. All patients in this study fulfilled the criteria for hyperglycemia and were diagnosed as newly diagnosed T2DM.

Anthropometric Measurements and Metabolic Parameters

Demographic information was collected through a standard questionnaire *via* face-to-face interviews by a physician (mainly including gender, age, history of medication, disease, surgery, family diabetes, and current or ever-smoking). Information was also obtained by a review of medical records and laboratory data. Physical examination was conducted by the research nurses (including height, weight, WC, and blood pressure). Body mass index (BMI) was calculated as the weight (kg, rounded to the nearest kilogram) divided by the square of

height (m, rounded to the nearest centimeter). WC was measured at the anatomical waist (the natural depression between the iliac crest and the 10th rib), which should be the narrowest part of the abdomen. Systolic and diastolic blood pressure (SBP and DBP) were recorded on at least three different occasions by an electronic sphygmomanometer with an appropriate cuff size after the patients have rested for more than 5 min, and the three readings were calculated.

Serum FBG, insulin, glycosylated hemoglobin, autoimmune antibodies (GADA, IAA, and ICA), total cholesterol (TC), HDL-c, LDL-c, TGs, uric acid (UA), and high-sensitivity C-reactive protein were measured by standard methods using fasting venous blood samples that were taken between 8 and 9 a.m. after overnight fasting for at least 12 h. Homeostasis model assessment (HOMA-IR) was used to assess insulin resistance. The estimate of HOMA score was calculated with the formula: fasting serum insulin (μU/ml) fasting plasma glucose (mmol/l)/22.5 (17).

Measurement of Perirenal Fat Thickness

CT scan was performed on all patients using Revolution VCT 256 (General Electric, Milwaukee, WI, USA) while in a supine position to measure PrFT. Images were reconstructed with Advantage work station 4.7 software (GE, Milwaukee, WI, USA) to obtain 1.25-mm-thick consecutive slices. The CT-scanned area covered was between the pubic symphysis and the 10th thoracic vertebra. Perirenal fat was differentiated from other tissues by density (HU). The center of the window is set in -100 HU, and the window widths ranged from 50 to 200 HU for further analysis. Each compartment is drawn by using a manually controlled trackball cursor. PrFT was measured on both sides for each patient according to a recent study (8). The adipose area of the renal sinus was separated by a tangent line touching the outer limits of the kidney and crossing over the renal hilum. Moreover, perirenal fat was separated by tracing the boundaries of the kidney, the aforementioned tangent line, and the perirenal fascia. The average of the maximal distance between the posterior wall of the kidney and the inner limit of the abdominal wall across the renal venous plane on both kidneys was calculated as the PrFT (**Figure 1**). Two radiologists were involved in the measurement of PrFT to reduce the inter-operator variability. The inter-operator agreement between the two radiologists is 0.92.

Statistical Analysis

Data were analyzed by using SPSS 23.0 software (SPSS Inc., IBM). Descriptive data were expressed as means ± standard deviation (SD). Discrete variables were summarized in frequency tables (*N*, %). The patients enrolled in this study were divided into three groups based on the tertiles of PrFT. The Cohen k statistic was used to assess the agreement of the PrFT measurements between the two radiologists. Statistical differences among groups were established with one-way analysis of variance (ANOVA), followed by Tukey test for multiple comparisons. Chi-square (χ^2) test or Fisher's exact test was used for the comparison of categorical variables. Student's *t*-test was used to compare the mean PrFT between the two genders. The relationship between PrFT and the metabolic parameters was assessed using Pearson's or Spearman's correlation analysis. A multiple binomial logistic regression model was used to estimate the independent effect of PrFT on the MetS after adjusting for other covariates by gender. The ROC curve was used to evaluate the identifying value of PrFT for MetS in newly diagnosed T2DM. The optimal cutoff value was based on the greatest value of the Youden index. A two-tailed value of $P < 0.05$ was considered statistically significant.

RESULTS

Overall, a total of 470 patients were screened; 445 patients meeting the inclusion and exclusion criteria were enrolled in this study. The flow diagram of the excluded and included patients is presented in **Figure 2**. Among the 445 patients, 226 (50.8%) patients were men. The mean age was 53.3 ± 7.9 years, ranging from 32 to 70 years old. The mean PrFT was 12.8 ± 4.8 mm. The prevalence of MetS was 57.5% (95% CI: 54.0–64.0%) in men and 58.9% (95% CI: 52.3–65.5%) in women separately. There was a significant difference in the mean PrFT between men and women (13.3 ± 5.1 *vs.* 12.2 ± 4.3 mm, $P < 0.001$). The characteristics of the patients divided into three groups based on tertiles of PrFT in men and women are shown in **Tables 1** and **2**. There was a significant difference in BMI, WC, TG, HDL-c, LDL-apolipoprotein B, UA, SBP, DBP, and HOMA-IR among groups both in men and women ($P < 0.05$). Patients in the higher-PrFT groups showed a higher level of BMI, WC, TG, UA, SBP, DBP, and HOMA-IR and a lower level of HDL-c compared with the lower-PrFT groups ($P < 0.05$). Moreover, patients in the higher-PrFT groups showed more patients that had MetS and hypertension ($P < 0.05$).

The main correlations between metabolic parameters and PrFT in the subgroup divided by sex are presented in **Table 3**. The results showed that PrFT was significantly and positively correlated with WC, BMI, TG, LDL-c, UA, SBP, DBP, and HOMA-IR in men and women groups. Moreover, PrFT was significantly and negatively correlated with HDL-c in both groups.

The association between MetS and PrFT was further investigated by binomial logistic regression analysis divided by sex (**Table 4**). The PrFT was shown to be independently associated with MetS after adjustment for age (model 1). The ORs (95% CI) were 1.53 (1.38–1.70) in men and 1.66 (1.47–1.88) in women. After further adjustment for BMI, TC, LDL-c, UA, and HOMA-IR (model 2), the PrFT was shown to be independently associated with MetS. The ORs (95% CI) were 1.33 (1.16–1.53) in men and 1.50 (1.27–1.78) in women. After further additional adjustment for TG, WC, HDL-c, SBP, and DBP (model 3), the ORs remained significant. The ORs (95% CI) were 1.15 (1.03–1.38) in men and 1.31 (1.08–1.96) in women ($P < 0.05$).

The ROC curve analysis was used to further evaluate the ability of PrFT in identifying MetS divided by sex. From the ROC curve analysis, the results showed a good predictive value of PrFT for MetS. The areas under the curve of PrFT in identifying MetS were 0.895 (95% CI: 0.852–0.939, $P < 0.001$) in men and 0.910 (95% CI: 0.876–0.953, $P < 0.001$) in women (**Figure 3**). The optimal cutoff values of PrFT were 14.6 mm (sensitivity: 83.8%, specificity: 89.6%) for men and 13.1 mm (sensitivity: 87.6%, specificity: 91.1%) for women (**Table 5**).

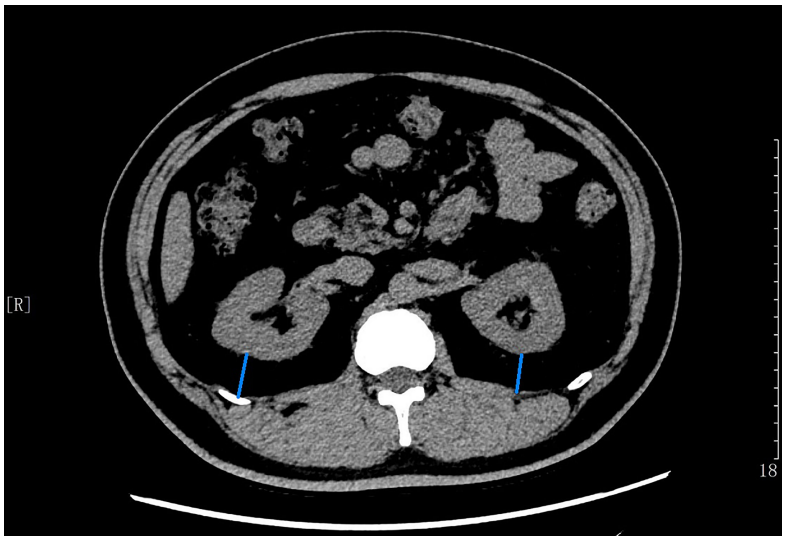


FIGURE 1 | The average of maximal thickness values (blue line) between the posterior wall of the kidney and the inner limit of the abdominal wall across the renal venous plane was calculated as the PrFT.

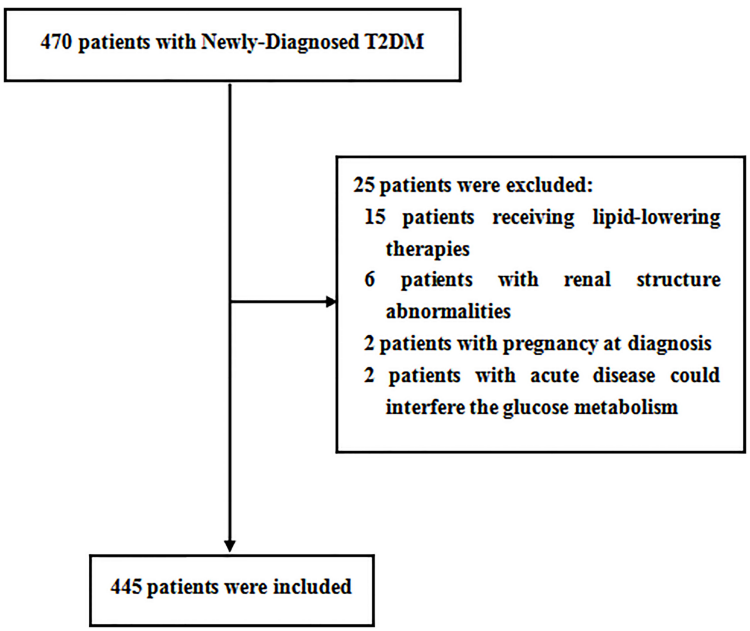


FIGURE 2 | Flow diagram of the patients excluded and included in this study.

DISCUSSION

Diabetes is a metabolic disease characterized by chronic hyperglycemia that is often accompanied with MetS at the first diagnosis. MetS is a cluster of conditions that can increase the risk of cardiovascular diseases, heart disease, and stroke, which may increase all-cause mortality. Due to the complexity of MetS diagnosis, leading clinical practice often overlooked it. In the present study, the results confirmed that PrFT shows a close correlation with metabolic risk factors. Moreover, PrFT was significantly associated with higher odds of MetS after adjustment for other confounders. The ROC curves also showed a good predictive value of PrFT for MetS. The optimal cutoff values of PrFT in identifying MetS for Chinese newly diagnosed T2DM was 14.6 mm for men and 13.1 mm for women.

TABLE 1 | The characteristics of newly diagnosed T2DM were divided into three groups based on tertiles of PrFT in men.

	Total	T1 (< 10.7 mm)	T2 (10.7–16.1 mm)	T3 (> 16.1 mm)	*P*
Age (year)	52.5 ± 8.2	52.4 ± 8.9	52.3 ± 7.7	52.8 ± 8.1	0.618
WC (cm)	86.6 ± 7.0	80.3 ± 3.3[ab]	86.9 ± 4.8[ac]	92.4 ± 6.4[bc]	< 0.001
BMI (kg/m^2)	24.8 ± 3.1	22.0 ± 1.9[ab]	25.0 ± 1.9[ac]	27.2 ± 2.9[bc]	< 0.001
HbA1c (%)	8.8 ± 0.9	8.8 ± 1.0	8.7 ± 0.8	8.9 ± 0.9	0.327
TG (mmol/L)	2.3 ± 1.5	1.5 ± 1.0[ab]	2.0 ± 0.7[ac]	3.3 ± 1.9[bc]	< 0.001
TC (mmol/L)	5.4 ± 1.3	5.1 ± 1.1[ab]	5.5 ± 1.4[a]	5.6 ± 1.2[b]	0.026
HDL-c (mmol/L)	1.1 ± 0.2	1.3 ± 0.2[ab]	1.1 ± 0.1[ac]	0.9 ± 0.1[bc]	< 0.001
LDL-c (mmol/L)	3.6 ± 1.0	3.4 ± 0.9[ab]	3.8 ± 1.1[a]	3.7 ± 0.9[b]	0.014
APOA (g/L)	1.3 ± 0.3	1.4 ± 0.2[ab]	1.3 ± 0.2[a]	1.2 ± 0.3[b]	0.003
APOB (g/L)	1.1 ± 0.3	1.0 ± 0.3[ab]	1.1 ± 0.3[a]	1.1 ± 0.3[b]	0.018
UA (umol/L)	360.4 ± 86.2	307.5 ± 60.1[ab]	372.4 ± 77.2[ac]	401.4 ± 90.5[bc]	< 0.001
SBP (mmHg)	134.3 ± 18.4	119.1 ± 11.4[ab]	137.2 ± 10.2[ac]	146.1 ± 19.4[bc]	< 0.001
DBP (mmHg)	82.5 ± 10.3	76.6 ± 6.1[ab]	83.4 ± 12.2[ac]	87.5 ± 7.2[bc]	< 0.001
HOMA-IR	11.5 ± 6.5	6.3 ± 3.7[ab]	12.4 ± 4.7[ac]	15.6 ± 6.8[bc]	< 0.001
hs-CRP (mg/L)	3.4 ± 1.1	3.3 ± 0.9	3.4 ± 1.0	3.5 ± 0.9	0.218
Hypertension, *n* (%)	84 (37.2)	9 (12.0)[ab]	27 (36.5)[ac]	48 (62.3)[bc]	< 0.001
Smoking, *n* (%)	120 (53.1)	38 (50.7)	40 (54.1)	42 (54.5)	0.683
MetS, *n* (%)	130 (57.5)	11 (14.7)[ab]	24 (32.4)[ac]	69 (89.6)[bc]	< 0.001

BMI, body mass index; WC, waist circumference; HbA1c, glycated hemoglobin; UA, uric acid; TG, triglyceride; TC, total cholesterol; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; SBP, systolic blood pressure; DBP, diastolic blood pressure; HOMR-IR, Homeostasis Model Assessment—Insulin Resistance; hs-CRP, high-sensitivity C-reactive protein; MetS, metabolic syndrome.

[a]*P < 0.05: T1 vs. T2.*

[b]*P < 0.05: T1 vs. T3.*

[c]*P < 0.05: T2 vs. T3.*

It indicated that PrFT can be a surrogate marker for MetS in Chinese newly diagnosed T2DM.

Visceral fat and subcutaneous fat are the most important and common categories in adipose biology based on the anatomical and physiological characteristics of fat depots. Clinical evidences demonstrated that Asians are more likely to have more obesity-related consequences in patients with lower WC and BMI due to more visceral fat mass deposition compared with Caucasians (18). CT scan is a reliable tool to quantify adipose tissue depots. The density of adipose tissue in Hounsfield unit (HU) can be used to distinguish visceral fat from other tissues. Among visceral adipose tissue deposits, perirenal fat located in the retroperitoneal space and surrounding the kidneys can be quantitatively measured by radiological diagnosis for renal positioning, and the posterolateral

TABLE 2 | The characteristics of newly diagnosed T2DM were divided into three groups based on tertiles of PrFT in women.

	Total	T1 (< 10.2 mm)	T2 (10.2–14.9 mm)	T3 (> 14.9 mm)	*P*
Age (year)	54.2 ± 7.5	53.5 ± 7.7	54.1 ± 7.5	54.8 ± 7.3	0.569
WC (cm)	85.1 ± 6.7	80.4 ± 3.8[ab]	84.6 ± 5.0[ac]	90.2 ± 6.8[bc]	< 0.001
BMI (kg/m^2)	24.2 ± 2.9	22.1 ± 2.1[ab]	24.1 ± 2.2[ac]	26.3 ± 2.6[bc]	< 0.001
HbA1c (%)	8.7 ± 1.0	8.7 ± 1.1	8.8 ± 0.9	8.7 ± 1.0	0.624
TG (mmol/L)	2.0 ± 1.1	1.3 ± 0.5[ab]	1.8 ± 0.6[ac]	2.9 ± 1.3[bc]	< 0.001
TC (mmol/L)	5.1 ± 1.1	4.9 ± 1.1[b]	5.2 ± 1.2	5.2 ± 1.0[b]	0.089
HDL-c (mmol/L)	1.1 ± 0.2	1.3 ± 0.2[ab]	1.1 ± 0.2[ac]	0.9 ± 0.2[bc]	< 0.001
LDL-c (mmol/L)	3.4 ± 0.9	3.1 ± 0.8[ab]	3.6 ± 0.9[a]	3.5 ± 1.0[b]	0.001
APOA (g/L)	1.3 ± 0.3	1.3 ± 0.3	1.3 ± 0.2	1.2 ± 0.3	0.519
APOB (g/L)	1.0 ± 0.3	0.9 ± 0.3[ab]	1.0 ± 0.3[a]	1.0 ± 0.3[b]	0.001
UA (umol/L)	348.9 ± 86.8	281.8 ± 62.3[ab]	352.5 ± 66.3[ac]	411.5 ± 76.8[bc]	< 0.001
SBP (mmHg)	132.3 ± 16.6	119.5 ± 12.2[ab]	132.6 ± 16.6[ac]	144.6 ± 9.7[bc]	< 0.001
DBP (mmHg)	81.0 ± 88.0	74.6 ± 5.7[ab]	82.0 ± 9.4[ac]	86.5 ± 6.5[bc]	< 0.001
HOMA-IR	10.7 ± 5.4	6.5 ± 3.7[ab]	11.4 ± 4.3[ac]	14.0 ± 5.1[bc]	< 0.001
hs-CRP (mg/L)	2.9 ± 0.8	3.0 ± 0.9	2.8 ± 1.2	2.8 ± 0.9	0.692
Hypertension, *n* (%)	81 (37.0)	11 (15.1)[ab]	24 (33.3)[ac]	46 (62.2)[bc]	< 0.001
Smoking, *n* (%)	6 (2.7)	3 (4.1)	3 (4.2)	0 (0)	0.207
MetS, *n* (%)	129 (58.9)	9 (12.3)[ab]	50 (69.4)[ac]	70 (94.6)[bc]	< 0.001

BMI, body mass index; WC, waist circumference; HbA1c, glycated hemoglobin; UA, uric acid; TG, triglyceride; TC, total cholesterol; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; SBP, systolic blood pressure; DBP, diastolic blood pressure; HOMR-IR, Homeostasis Model Assessment—Insulin Resistance; hs-CRP, high-sensitivity C-reactive protein; MetS, metabolic syndrome.

[a]*P < 0.05: T1 vs. T2.*

[b]*P < 0.05: T1 vs. T3.*

[c]*P < 0.05: T2 vs. T3.*

TABLE 3 | Main correlations between metabolic parameters and PrFT in newly diagnosed T2DM divided by sex.

Parameter	Men (*n* = 226)		Women (*n* = 219)	
	R	*P*	*R*	*P*
WC (cm)	0.770	< 0.001	0.674	< 0.001
BMI (kg/m^2)	0.779	< 0.001	0.690	< 0.001
HbA1c (%)	0.018	0.798	0.115	0.084
TG (mmol/L)	0.602	< 0.001	0.726	< 0.001
TC (mmol/L)	0.128	0.068	0.131	0.051
LDL-c (mmol/L)	0.156	0.019	0.190	0.005
HDL-c (mmol/L)	-0.592	< 0.001	-0.507	< 0.001
APOA (g/L)	-0.055	0.417	-0.046	0.528
APOB (g/L)	0.078	0.246	0.122	0.066
UA (umol/L)	0.494	< 0.001	0.665	< 0.001
SBP (mmHg)	0.695	< 0.001	0.713	< 0.001
DBP (mmHg)	0.538	< 0.001	0.611	< 0.001
hs-CRP (mg/L)	-0.067	0.364	-0.086	0.204
HOMA-IR	0.688	< 0.001	0.656	< 0.001

BMI, body mass index; HbA1c, glycated hemoglobin; WC, waist circumference; TG, triglyceride; TC, total cholesterol; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; UA, uric acid; SBP, systolic blood pressure; DBP, diastolic blood pressure; hs-CRP, high-sensitivity C-reactive protein; HOMR-IR: Homeostasis Model Assessment —Insulin Resistance.

TABLE 4 | Binomial logistic regression analysis adjusted odds ratios (95% CIs) of PrFT in newly diagnosed T2DM divided by sex.

Parameter	Men (*n* = 226)		Women (*n* = 219)	
	OR (95% CI)	*P*	OR (95% CI)	*P*
Model 1	1.53 (1.38–1.70)	< 0.001	1.66 (1.47–1.88)	< 0.001
Model 2	1.33 (1.16–1.53)	< 0.001	1.50 (1.27–1.78)	< 0.001
Model 3	1.15 (1.03–1.38)	0.046	1.31 (1.08–1.96)	0.034

Model 1 was adjusted for age. Model 2 was adjusted for BMI, TC, LDL-C, UA, and HOMA-IR. Model 3 was additionally adjusted for TG, WC, HDL-c, SBP, and DBP.
BMI, body mass index; WC, waist circumference; HbA1c, glycated hemoglobin; UA, uric acid; TG, triglyceride; TC, total cholesterol; HDL-c, high-density lipoprotein cholesterol; LDL-c, low-density lipoprotein cholesterol; SBP, systolic blood pressure; DBP, diastolic blood pressure; HOMR-IR, Homeostasis Model Assessment Insulin Resistance.

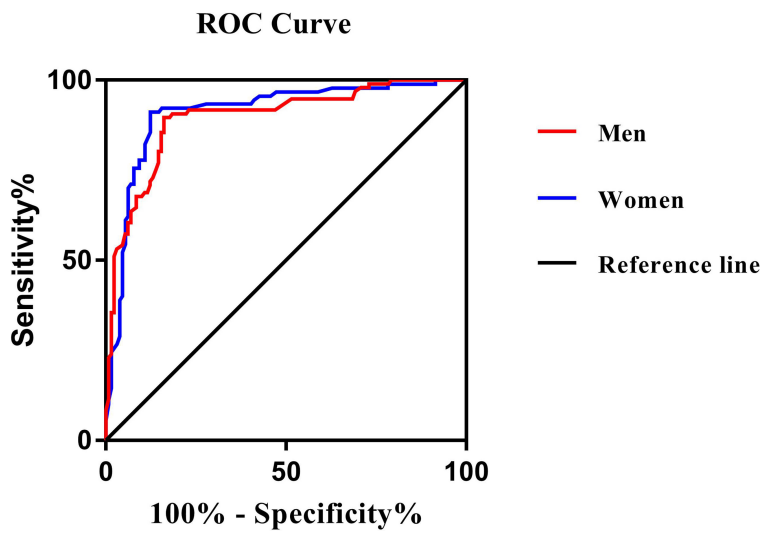


FIGURE 3 | Receiver operating characteristic curves for the cutoff value of PrFT to identify MetS. In men, the area under the curve was 0.895 (95% CI: 0.852–0.939). In women, the area under the curve was 0.910 (95% CI: 0.876–0.953). The optimal cutoff values of PrFT were 14.6 mm (sensitivity: 83.8%, specificity: 89.6%) for men and 13.1 mm (sensitivity: 87.6%, specificity: 91.1%) for women.

TABLE 5 | Receiver operating characteristic curve analysis of PrFT in identifying MetS in newly diagnosed T2DM divided by sex.

AUC (95% CI)	Cut-off value	Sensitivity (%) (95% CI)	Specificity (%) (95% CI)	PPV (%) (95% CI)	NPV (%) (95% CI)
Men (*n* = 226)					
0.895	14.6	83.8	89.6	91.6	80.4
(0.852–0.939)		(79.3–88.9)	(85.3–92.4)	(86.5–96.7)	(72.7–88.0)
Women (*n* = 219)					
0.910	13.1	87.6	91.1	93.4	83.7
(0.876–0.953)		(83.3–92.1)	(87.0–94.7)	(88.9–97.9)	(76.2–91.1)

MetS, metabolic syndrome; PrFT, perirenal fat thickness; PPV, positive predictive value; NPV, negative predictive value.

perirenal fat thickness measured by CT scanning had shown a positive correlation with perirenal fat mass (8). The anatomical structure and location of perirenal fat determined its specific biological characteristics. Compared with other adipose tissues classified as loose connective tissues, perirenal fat has a complete system of blood supply, lymph fluid drainage, innervation, and other special morphological features, which make it similar to other internal organs and different from traditionally classified connective tissues (9). These special anatomical structure ensured that perirenal fat can modulate the metabolic system through neural reflexes (19), adipokine secretion (20), adipocyte interactions (21), and paracrine substance (22). Among them, adipokines (leptin, adiponectin, apelin, and nesfatin) play important regulatory roles in endocrine metabolic systems, insulin sensitivity, and lipolysis *via* the autocrine, paracrine, and endocrine pathways (23, 24). In addition, other bioactive factors, such as leptin, adiponectin, tumor necrosis factor-α, interleukin-6, interleukin-8, and MCP-1, can also be released from perirenal fat, which is involved in the pathogenesis of CVD, metabolic disorders, and T2DM (25, 26). Thus, these specific biological and anatomical characteristics provided a basis for the involvement of perirenal fat in MetS regulation.

Clinical studies have also observed the association between PrFT and metabolic risk factors. A study that enrolled overweight and obese subjects showed that PrFT was independently associated with HDL-c and WC (13). Another study has also shown that PrFT was significantly correlated with metabolic risk factors such as UA, TG, and WC in patients with chronic kidney disease (27). Moreover, PrFT also showed a positive independent association between PrFT and mean 24-h diastolic blood pressure levels in overweight and obese subjects (28). The results in our study also showed a positive correlation between PrFT and HOMA-IR, which was confirmed to participate in the occurrence and development of MetS and T2DM (29). Meanwhile, PrFT is also reported to be associated with other metabolic diseases and T2DM complications. Satsuki K et al. have also demonstrated that PrFT can be a reliable method for the quantification offatty liver as well as for the quantification of visceral fat (30). Increasing evidence have suggested that the accumulation of perirenal fat increases the risk for the development of chronic kidney disease through decreasing the eGFR level and increasing the excretion rate of urinary protein (31–33). The results in our study were consistent with these previous studies. PrFT was correlated with metabolic risk factors like WC, TG, HDL-c, SBP, DBP, UA, and HOMA-IR. As expected, PrFT was significantly independent with higher odds (95% CI) of MetS after adjustment for other confounders. The ROC curve analysis results in our study showed a good predictive value of PrFT for MetS both in men and women, which indicated that PrFT can be a surrogate marker for MetS in newly diagnosed T2DM.

To our knowledge, this is the first study to have confirmed the predictive value of PrFT for MetS in Chinese newly diagnosed T2DM. There are some limitations in our study. Firstly, due to the fact that the prevalence of MetS may vary in geographic distribution and race (34), the optimal cutoff values of PrFT may not be applicable to other races. Secondly, although CT scanning can accurately measure the PrFT, the radiation may limit its use in clinical practice. In conclusion, in this cross-sectional study, a surrogate marker for MetS in Chinese newly diagnosed T2DM was found. PrFT was significantly independent with MetS and showed a powerful predictive value for MetS, which suggested that PrFT can be a surrogate marker for MetS in Chinese newly diagnosed T2DM.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding author.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Ethical Committee of Longyan First Affiliated Hospital of Fujian Medical University. The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

WW contributed to data curation and writing—review and editing. WW, XG, YC and MT conducted the investigation. XG took charge of the software and contributed to writing—original draft. All authors contributed to the article and approved the submitted version.

ACKNOWLEDGMENTS

We thank the Department of Radiology of our hospital for providing the CT scan equipment and technical support.

REFERENCES

1. Zimmet P, alberti G, Kaufman F, Tajima N, Silink M, Arslanian S, et al. The Metabolic Syndrome in Children and Adolescents. *Lancet (London England)* (2007) 369(9579):2059–61. doi: 10.1016/S0140-6736(07)60958-1
2. LI X, Cao C, Tang X, Yan X, Zhou H, Liu J, et al. Prevalence of Metabolic Syndrome and Its Determinants in Newly-Diagnosed Adult-Onset Diabetes in China: A Multi-Center, Cross-Sectional Survey. *Front Endocrinol* (2019) 10):661. doi: 10.3389/fendo.2019.00661
3. Chen W, Pan Y, Jing J, Zhao X, Liu L, Meng X, et al. Recurrent Stroke in Minor Ischemic Stroke or Transient Ischemic Attack With Metabolic Syndrome and/or Diabetes Mellitus. *J Am Heart Assoc* (2017) 6(6):e005446. doi: 10.1161/JAHA.116.005446
4. Fujiyoshi A, Murad MH, Luna M, Rosario A, Ali S, Paniagua D, et al. Metabolic Syndrome and Its Components are Underdiagnosed in Cardiology Clinics. *J Eval Clin Pract* (2011) 17:78–83. doi: 10.1111/j.1365-2753.2010.01371.x
5. Kuk J, Church T, Blair S, Ross R. Does Measurement Site for Visceral and Abdominal Subcutaneous Adipose Tissue Alter Associations With the Metabolic Syndrome? *Diabetes Care* (2006) 29(3):679–84. doi: 10.2337/diacare.29.03.06.dc05-1500
6. Fox C, Massaro J, Hoffmann U, Pou K, Maurovich P, Liu Y, et al. Abdominal Visceral and Subcutaneous Adipose Tissue Compartments: Association With Metabolic Risk Factors in the Framingham Heart Study. *Circulation* (2007) 116(1):39–48. doi: 10.1161/CIRCULATIONAHA.106.675355
7. Rotheny M, Catapano A, Xia J, Wacker W, Tidone C, Grigore L, et al. Abdominal Visceral Fat Measurement Using Dual-Energy X-Ray: Association With Cardiometabolic Risk Factors. *Obesity (Silver Spring Md)* (2013) 21 (9):1798–802. doi: 10.1002/oby.20223
8. Favre G, Grangeon-Chapon C, Raffaelli C, François F, Iannelli A, Esnault V, et al. Perirenal Fat Thickness Measured With Computed Tomography Is a Reliable Estimate of Perirenal Fat Mass. *PloS One* (2017) 12(4):e0175561. doi: 10.1371/journal.pone.0175561
9. Kim JH, Han EH, Jin ZW, Lee HK, Fujimiya M, Murakami G, et al. Fetal Topographical Anatomy of the Upper Abdominal Lymphatics: Its Specific Features in Comparison With Other Abdominopelvic Regions. *Anat Rec* (2011) 295(1):91–104. doi: 10.1002/ar.21527
10. Czaja K, Kraeling R, Klimczuk M, Franke-Radowiecka A, Sienkiewicz W, Lakomy M, et al. Distribution of Ganglionic Sympathetic Neurons Supplying the Subcutaneous, Perirenal and Mesentery Fat Tissue Depots in the Pig. *Acta Neurobiol Exp (Wars)* (2002) 62(4):227–34.
11. Wang QA, Tao C, Jiang L, Shao M, Ye R, Zhu Y, et al. Distinct Regulatory Mechanisms Governing Embryonic Versus Adult Adipocyte Maturation. *Nat Cell Biol* (2015) 17(9):1099–111. doi: 10.1038/ncb3217
12. Ricci M, Scavizzi M, Ministrini S, De Vuono S, Pucci G, Lupattelli G. Morbid Obesity and Hypertension: The Role of Perirenal Fat. *J Clin Hypertens (Greenwich Conn)* (2018) 20(10):1430–7. doi: 10.1111/jch.13370
13. Manno C, Campobasso N, Nardecchia A, Triggiani V, Zupo R, Gesualdo L, et al. Relationship of Para- and Perirenal Fat and Epicardial Fat With Metabolic Parameters in Overweight and Obese Subjects. *Eat Weight Disord* (2019) 24(1):67–72. doi: 10.1007/s40519-018-0532-z
14. Austin Peter C, Steyerberg Ewout W. Events Per Variable (EPV) and the Relative Performance of Different Strategies for Estimating the Out-of-Sample Validity of Logistic Regression Models. *Stat Methods Med Res* (2017) 26:796–808. doi: 10.1177/0962280214558972
15. American Diabetes Association. Standards of Medical Care in Diabetes-20202. Classification and Diagnosis of Diabetes. *Diabetes Care* (2020) 43: S14–31. doi: 10.2337/dc22-S002
16. Society CD. Guideline for the Prevention and Treatment of Type 2 Diabetes Mellitus in China (2020 Edition). *Chin J Diabetes Mellitus* (2021) 13(4):378–9. doi: 10.3760/cma.j.cn112138-20211027-00751
17. Bonora E, Targher G, Alberiche M, Bonadonna RC, Saggiani F, Zenere MB, et al. Homeostasis Model Assessment Closely Mirrors the Glucose Clamp Technique in the Assessment of Insulin Sensitivity: Studies in Subjects With Various Degrees of Glucose Tolerance and Insulin Sensitivity. *Diabetes Care* (2000) 23(1):57–63. doi: 10.2337/diacare.23.1.57
18. Deurenberg YM, Chew SK, Deurenberg P. Elevated Body Fat Percentage and Cardiovascular Risks at Low Body Mass Index Levels Among Singaporean Chinese, Malays and Indians. *Obes Rev* (2002) 3:209–15. doi: 10.1046/j.1467-789X.2002.00069.x
19. Xiong XQ, Chen W, Han Y, et al. Enhanced Adipose Afferent Reflex Contributes to Sympathetic Activation in Diet-Induced Obesity Hypertension. *Hypertension* (2012) 60:1280–6. doi: 10.1161/HYPERTENSIONAHA.112.198002
20. Kelesidis T, Kelesidis I, Chou S, et al. Narrative Review: The Role of Leptin in Human Physiology: Emerging Clinical Applications. *Ann Intern Med* (2010) 152:93–100. doi: 10.7326/0003-4819-152-2-201001190-00008
21. Hall JE, do Carmo JM, da Silva A, et al. Obesity-Induced Hypertension: Interaction of Neurohumoral and Renal Mechanisms. *Circ Res* (2015) 116:991–1006. doi: 10.1161/CIRCRESAHA.116.305697
22. Ma ST, Zhu XY, Eirin A, et al. Perirenal Fat Promotes Renal Arterial Endothelial Dysfunction in Obese Swine Through Tumor Necrosis Factor-α. *J Urol* (2016) 195:1152–9. doi: 10.1016/j.juro.2015.08.105
23. Simonds SE, Pryor JT, Ravussin E, et al. Leptin Mediates the Increase in Blood Pressure Associated With Obesity. *Cell* (2014) 159:1404–16. doi: 10.1016/j.cell.2014.10.058
24. Díez J, Iglesias P. The Role of the Novel Adipocyte-Derived Protein Adiponectin in Human Disease: An Update. *Mini Rev Med Chem* (2010) 10:856–69. doi: 10.2174/138955710791608325
25. Lira FS, Rosa JC, Dos SR, Venancio DP, Carnier J, Sanches PL, et al. Visceral Fat Decreased by Long-Term Interdisciplinary Lifestyle Therapy Correlated Positively With Interleukin-6 and Tumor Necrosis Factor-αand Negatively With Adiponectin Levels in Obese Adolescents. *Metabolism* (2011) 60:359–65. doi: 10.1016/j.metabol.2010.02.017
26. Ohman MK, Wright AP, Wickenheiser KJ, Luo W, Russo HM, Eitzman DT, et al. Monocyte Chemoattractant Protein-1 Deficiency Protects Against Visceral Fat-Induced Atherosclerosis. *Arterioscler Thromb Vasc Biol* (2010) 30:1151–8. doi: 10.1161/ATVBAHA.110.205914
27. D'Marco L, Salazar J, Cortez M, Salazar M, Wettel M, Lima-Martínez M, et al. Perirenal Fat Thickness Is Associated With Metabolic Risk Factors in Patients With Chronic Kidney Disease. *Kidney Res Clin Pract* (2019) 38:365–72. doi: 10.23876/j.krcp.18.0155
28. De Pergola G, Campobasso N, Nardecchia A, Triggiani V, Caccavo D, Gesualdo L, et al. Para- and Perirenal Ultrasonographic Fat Thickness is Associated With 24-Hours Mean Diastolic Blood Pressure Levels in Overweight and Obese Subjects. *BMC Cardiovasc Disord* (2015) 15:108. doi: 10.1186/s12872-015-0101-6
29. Antonio-Villa NE, Bello-Chavolla OY, Vargas-Vázquez A, Mehta R, Aguilar-Salinas CA. The Combination of Insulin Resistance and Visceral Adipose Tissue Estimation Improves the Performance of Metabolic Syndrome as a Predictor of Type 2 Diabetes. *Diabetes Med* (2020) 37:1192–201. doi: 10.1111/dme.14274
30. Kawasaki S, Hasegawa O, Satoh S, Numata K, Terauchi Y. Sonographic Assessment of Fatty Liver Infiltration Using the Measurement of Para- and Perirenal Fat Thickness. *J Clin Ultrasound* (2010) 38:470–4. doi: 10.1002/jcu.20736
31. Fang Y, Xu YC, Yang YX, Liu C, Zhao , Ke J, et al. The Relationship Between Perirenal Fat Thickness and Reduced Glomerular Filtration Rate in Patients With Type 2 Diabetes. *J Diabetes Res* (2020) 2020:6076145. doi: 10.1155/2020/6076145
32. Shen FC, Cheng BC, Chen JF. Peri-Renal Fat Thickness Is Positively Associated With the Urine Albumin Excretion Rate in Patients With Type 2 Diabetes. *Obes Res Clin Pract* (2020) 14:345–9. doi: 10.1016/j.orcp.2020.06.006
33. Chen XJ, Mao Y, Hu JB, Han S, Gong L, Luo T, et al. Perirenal Fat Thickness Is Significantly Associated With the Risk for Development of Chronic Kidney Disease in Patients With Diabetes. *Diabetes* (2021) 70:2322–32. doi: 10.2337/db20-1031
34. Zhou H, Cheng LY, Shan ZY, Jia WP, Yang WY, Lu JM, et al. Effectiveness of Different Waist Circumference Cut-Off Values in Predicting Metabolic Syndrome Prevalence and Risk Factors in Adults in China. *BioMed Environ Sci* (2014) 27:325–34. doi: 10.3967/bes2014.057

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers in Endocrinology

SYSTEMATIC REVIEW
published: 24 March 2022
doi: 10.3389/fendo.2022.857930

Normal Weight Obesity and Cardiometabolic Risk Factors: A Systematic Review and Meta-Analysis

Nami Mohammadian Khonsari[1], Patricia Khashayar[2], Ehsan Shahrestanaki[3], Roya Kelishadi[4], Sahar Mohammadpoor Nami[1], Motahar Heidari-Beni[5], Zahra Esmaeili Abdar[1], Ozra Tabatabaei-Malazy[6] and Mostafa Qorbani[1]**

[1] Non-Communicable Diseases Research Center, Alborz University of Medical Sciences, Karaj, Iran, [2] Center for Microsystems Technology, Imec & Ghent University, Zwijnaarde-Gent, Belgium, [3] Social Determinants of Health Research Center, Alborz University of Medical Sciences, Karaj, Iran, [4] Department of Pediatrics, Child Growth and Development Research Center, Research Institute for Primordial Prevention of Non-Communicable Disease, Isfahan University of Medical Sciences, Isfahan, Iran, [5] Department of Nutrition, Child Growth and Development Research Center, Research Institute for Primordial Prevention of Non-Communicable Disease, Isfahan University of Medical Sciences, Isfahan, Iran, [6] Non-Communicable Diseases Research Center, Endocrinology and Metabolism Population Sciences Institute, Tehran University of Medical Sciences, Tehran, Iran

OPEN ACCESS

Edited by:
Luca Busetto,
Università degli Studi di Padova, Italy

Reviewed by:
Valeria Guglielmi,
University of Rome Tor Vergata, Italy
Alexis Elias Malavazos,
IRCCS San Donato Polyclinic, Italy

***Correspondence:**
Mostafa Qorbani
mqorbani1379@yahoo.com
Ozra Tabatabaei-Malazy
tabatabaeiml@sina.tums.ac.ir

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: 19 January 2022
Accepted: 14 February 2022
Published: 24 March 2022

Citation:
Mohammadian Khonsari N, Khashayar P, Shahrestanaki E, Kelishadi R, Mohammadpoor Nami S, Heidari-Beni M, Esmaeili Abdar Z, Tabatabaei-Malazy O and Qorbani M (2022) Normal Weight Obesity and Cardiometabolic Risk Factors: A Systematic Review and Meta-Analysis. Front. Endocrinol. 13:857930. doi: 10.3389/fendo.2022.857930

Background: Obesity is one of the most significant causes of morbidity and mortality worldwide. Current studies suggest a new type of obesity, normal weight obesity (NWO), which is defined as having a normal body mass index (BMI), but a high-fat percentage increases the risk of cardiometabolic risk factors (CMRFs). This systematic review and meta-analysis aimed to pool the association between NWO with CMRFs.

Methods: A systematic search of the literature in all available electronic databases, including Scopus, Web of Science, EMBASE, and PubMed, was performed until October 2021. All English studies that assessed the association of NWOs [compared to normal weight non-obese (NWNO)] and the CMRFs were included. Two investigators extracted data and performed a quality assessment. The heterogeneity between studies was assessed with I-squared and Cochran's Q tests. Odds ratio (OR) was used as an effect size to pool the association of NWO with CMRFs.

Results: Twenty-five articles that met the inclusion criteria entered the study. The total number of participants was 177,792, with an age range of 13 to 75 years. Most studies were conducted on the general population (adults) and were from China. The result of fixed-effect model meta-analysis indicated an increased odds of hyperglycemia (OR:1.50, 95%:1.23, 1.76), high TG (OR:1.90, 95% CH:1.44, 2.35), low HDL (OR: 1.28, 95% CI:1.06, 1.49) and diabetes (OR:1.39, 95% CI:1.30, 1.49). Moreover, the random effect meta-analysis showed that NWO increased the odds of dyslipidemia (OR:1.83, 95% CI:1.61, 20.4), HTN (OR:1.40, 95% CI:1.28, 1.51) and metabolic syndrome (OR:1.92, 95% CI:1.58, 2.26). Moreover, the mean of all CMRFs except plasma glucose in NWO subjects was statistically higher than NWNO subjects (p-value<0.05).

Conclusion: The present study showed that NWO increased the odds of CMRFs. These findings indicate the inadequacy of the BMI measurement and the need for body fat assessment for a better obesity risk assessment.

Keywords: normal weight obesity, central obesity, obesity, cardiometabolic, metabolic syndrome

BACKGROUND

Obesity is one of the most significant causes of morbidity and mortality worldwide (1, 2). In literature, obesity is usually defined as a body mass index (BMI) above 30 Kg/m^2 (2). The prevalence of obesity is increasing throughout the globe. This disease imposes a significant burden on the affected population and the health system. It is also considered a fulcrum of other conditions, such as cardiometabolic conditions, that arise from obesity and are the leading cause of death worldwide (1–3). Although the prevalence of these supposedly obesity-related complications (e.g., diabetes, hypertension, dyslipidemia, etc.) and cardiometabolic diseases is exceptionally higher among obese individuals, their prevalence has been increased in the past few decades, among the non-obese population (BMI under 30 Kg/m^2) and even in those considered healthy based on their BMI levels (BMI between 18.5 to 24.9 Kg/m^2) (4–6). This shows that BMI, long known as a great assessment tool, cannot determine an individual's body composition, and fat percentage, lacking the adequate properties to identify those with a high body fat percentage or disproportionate body fat distribution (e.g. abdominal obesity) (7) Recent studies suggest the percentage of body fat is directly related to cardiometabolic and obesity-related conditions. This is a new type of obesity in which an individual with normal BMI levels is considered as obese based on their body composition and fat percentage (8, 9). Normal weight obesity (NWO) has different definitions based on the studies, population, and gender; however, it is usually defined as a body fat percentage above 30% (10). Due to the lifestyle changes, lack of proper physical activity and the use of processed food, the numbers of obese individuals are on the rise (11, 12); accordingly, the number of the normal weigh obese might be increasing; however, due to their normal BMI they will remain undiagnosed, and no proper preventive measure is taken until it is too late (13). Since there has been no new individual data or aggregated systematic reviews and meta-analyses on this relatively novel subject, we conducted this study to assess the cardiometabolic risk factors (CMRFs) and anthropometric measurements in the NWO individuals and compare them with the normal population. This study aims to give a realistic overview of the emerging obesity-related conditions so that health authorities can take proper action and implement appropriate preventive measures.

Abbreviations: NWO, Normal weight obesity; CMRFs, cardio-metabolic risk factors; NWNO, normal weight non obese; M, male; F, female; TC, total cholesterol; HOMA, Homeostatic Model Assessment for Insulin Resistance; LDL, low-density lipoproteins; HDL, high-density lipoproteins; TG, triglyceride; SBP, systolic blood pressure; DBP, diastolic blood pressure; BMI, body mass index; WC, waist circumference; MetS, metabolic syndrome; DM, diabetes mellitus; HTN, hypertension; CM, centimeters; IDF, International Diabetes Federation; SMD, Standardized Mean Difference; OR, odds ratio; SD, Standard Deviation; CI, Confidence Interval; Q.A, quality assessment.

METHODS

This study was conducted according to the Preferred Reporting Items for Systematic Review and Meta-Analysis (PRISMA) guidelines.

Search Strategy

We conducted a systematic search of the literature in all available electronic databases, including Scopus, Web of Science, EMBASE, and PubMed, until October 2021. The terms used for the search was "NWO", "central obesity"," high-fat percentage", and their equivalent terms based on MesH terms. The search strategy is presented in the **Supplementary Table 1**. Moreover, one investigator conducted the search, and another investigator reviewed the search results.

Eligibility Criteria and Selection Study

All English studies that assessed anthropometric measurements and the CMRFs [BMI, lean body mass, body fat mass, waist, hip, plasma glucose level, total cholesterol, Homeostatic Model Assessment for Insulin Resistance (HOMA), low-density lipoprotein (LDL), High-density lipoprotein (HDL), Triglyceride (TG), Total cholesterol (TC), systolic blood pressure (SBP), diastolic blood pressure (DBP), hypertension (HTN)] among NWO individuals were included. All studies had to represent the target population and compare them with the normal-weight non-obese (NWNOs) individuals and adjust for possible confounders to be included in our study. Only the most recent studies were included in our research if multiple studies used the same data source. All definitions of NWO, Regardless of their variety (e.g. normal BMI with high body fat percentage, normal BMI with high waist circumference (central obesity), high waist to hip/height ratio and etc.) were included in this study.

After removing the duplicates using EndNote X7, two investigators independently assessed the titles, abstracts, and finally, the full texts of the remaining articles. In addition, hand searching was performed to find relevant studies from the reference list of the included articles. Any discrepancies were referred to the third investigator for resolution.

Data Extraction Strategy

Two investigators independently extracted the data using an electronic data extraction sheet. The extracted data included the

name of the first author, the year of the study, sample size, sex, age (mean or range), NWO definition, CMRFs, Odds ratios (OR) or standard mean difference (SMD), and 95% confidence interval (CI) as an effect size of dichotomous and continuous data respectively. Two other investigators helped resolve any discrepancy.

Quality Assessment (QA)

The Newcastle-Ottawa Scale was used for the quality assessment of the included articles. This seven-item scale scores the selection, exposure (case-control study) and outcome (cohort study), and comparability of the studies. The total score, which is the sum of each item score, ranges from 0 to 9, with greater scores indicating lower bias risk. The scores were categorized as 0 to 4, 5 and 6, 7 to 9, meaning unsatisfactory, satisfactory, and good quality, respectively. All of the above steps were assessed independently by two investigators. Finally, any discrepancies were referred to the third investigator for resolution.

Statistical Analysis

The heterogeneity between the studies was assessed using the I-squared and Cochran's Q tests if heterogeneity was statistically significant (P-value<0.1) (14). a random effect model was used; otherwise, a fixed model was applied. Odds Ratio (OR) and 95% confidence interval (CI) were used as an effect size of meta-analysis to pool the association of NWO with CMRFs as a dichotomous variable. We also calculated and pooled the standardized mean difference (SMD) as an effect size for NWO association with the means of CMRFs. Meta-analysis was done for outcomes that were reported in more than three studies. Sub-group analysis was performed for the CMRFs. Publication bias was assessed using Egger's test for each CMRF; if publication bias was seen, sensitivity analysis was performed. STATA version 11 (Stata Corporation, College Station, Texas, USA) was used for the analysis.

RESULTS

Search Results

From the 523 studies of the initial search, 270 were duplicates; thus, 253 articles were evaluated, and 201 were considered irrelevant based on the title and abstract. The remaining 52 articles' full text was then assessed and evaluated for eligibility criteria, and 27 articles were excluded. Twenty-three articles met the inclusion criteria. Two studies with unadjusted data for potential confounders were included due to their exceptionally high, quality assessment score; however, these two studies were not included in the Quantitative synthesis. This process is illustrated in **Figure 1**.

General Characteristics

These studies were conducted worldwide (United States of America, Sweden, Korea, Colombia, West Indies, China, India, Iran, Japan, Iceland, Malaysia, Switzerland, Brazil, and Finland). The total number of participants was 177,792, with an age range of 13 to 75 years. These data, alongside other study characteristics, are presented in **Table 1**. Most of these studies were conducted on the general population (adults) and were from china (5 studies) and Korea (4 studies). With one study, Sweden, West Indies, India, Iran, Japan, Iceland, Malaysia, Switzerland, and Finland had the lowest number of studies. The largest sample size was from Japan with 117163 participants, and the smallest was from Iceland with 182 participants (4, 6, 8, 15–36).. These general characteristics of included studies for NWO association with CMRFs are shown in **Table 1**.

Qualitative Synthesis

The association of NWO with the means of CMRFs compared to NWNO in included studies is shown in **Table 2** alongside their effect size. As illustrated, most anthropometric components among NWOs significantly differed from that of NWNOs with the most significant, regarding the fat mass [effect size: -1.9 95%.CI (-2.01_-1.8)]. The reported mean range of the associated cardiometabolic parameters are as follows, plasma glucose 81.96 to 95.7 mg/dL for NWNOs and 80.47 to 99.1 mg/dL for NWOs. Total cholesterol, 181.61 to 204.97 mg/dL for NWNOs and 189.61 to 216.55 mg/dL for NWOs, HOMA, 0.6 to 3.32 for NWNOs and 0.8 to 3.18 for NWOs, LDL, 105.3 to 121.8 mg/dL for NWNOs and 117.67 to 132.64 mg/dL for NWOs, HDL mg/dL, 42.09 to 72.7 mg/dL for NWNOs and 45.72 to 69.5 mg/dL for NWOs, TG, 76.72 to 116.03 mg/dL for NWNOs and 85.32 to 136.4 mg/dL for NWOs, SBP, 107.81 to 129 mmHg for NWNOs and 110.58 to 134 mmHg for NWOs, DBP, 71 to 80 mmHg for NWNOs and 72.1 to 85 mmHg for NWOs. As for anthropometrics, the mean ranges of lean mass were 40.21 to 57.5 kg in NWNOs and 39.9 to 57.4 kg in NWOs, waist, 59.12 to 84.4 cm for NWNOs and 63.94 to 89.9 in NWOs, hip 93.2 to 96.1 cm in NWNOs and 94.6 to 98.9 cm NWOs. Association of NWO with CMRFs as categorical data in included studies are shown in **Table 3**. As illustrated, most reported ORs are statistically significant. The greatest reported OR was of elevated waist circumference (WC) among NWOs [OR: 26.61 95%.CI (4.75-149.14)] and the odds of metabolic syndrome (MetS) among NWOs [OR:8.89 95%.CI (3.32-4.47)].

Quantitative Synthesis

The overall and sex-stratified pooled ORs of the relationship between NWO and CMRFs are shown in **Table 4**. The result of the meta-analysis showed that the overall odds ratio of hyperglycemia increased by 50% (OR:1.50, 95%:1.23, 1.76), of high TG by 90% (OR:1.90, 95% CH:1.44, 2.35), of low HDL by 28% (OR: 1.28, 95% CI:1.06, 1.49) and of diabetes by 39% (OR:1.39, 95% CI:1.30, 1.49) among NWO individuals. Also, the random effect meta-analysis showed increased odds of dyslipidemia by 83% (OR:1.83, 95% CI:1.61, 20.4), of HTN by 40% (OR:1.40, 95% CI:1.28, 1.51) and of metabolic syndrome by 92% (OR:1.92, 95% CI:1.58, 2.26) in the same population (**Figure 2**).

The overall and sex-stratified association between NWO and the mean of CMRFs are shown in **Table 5**. A low to high

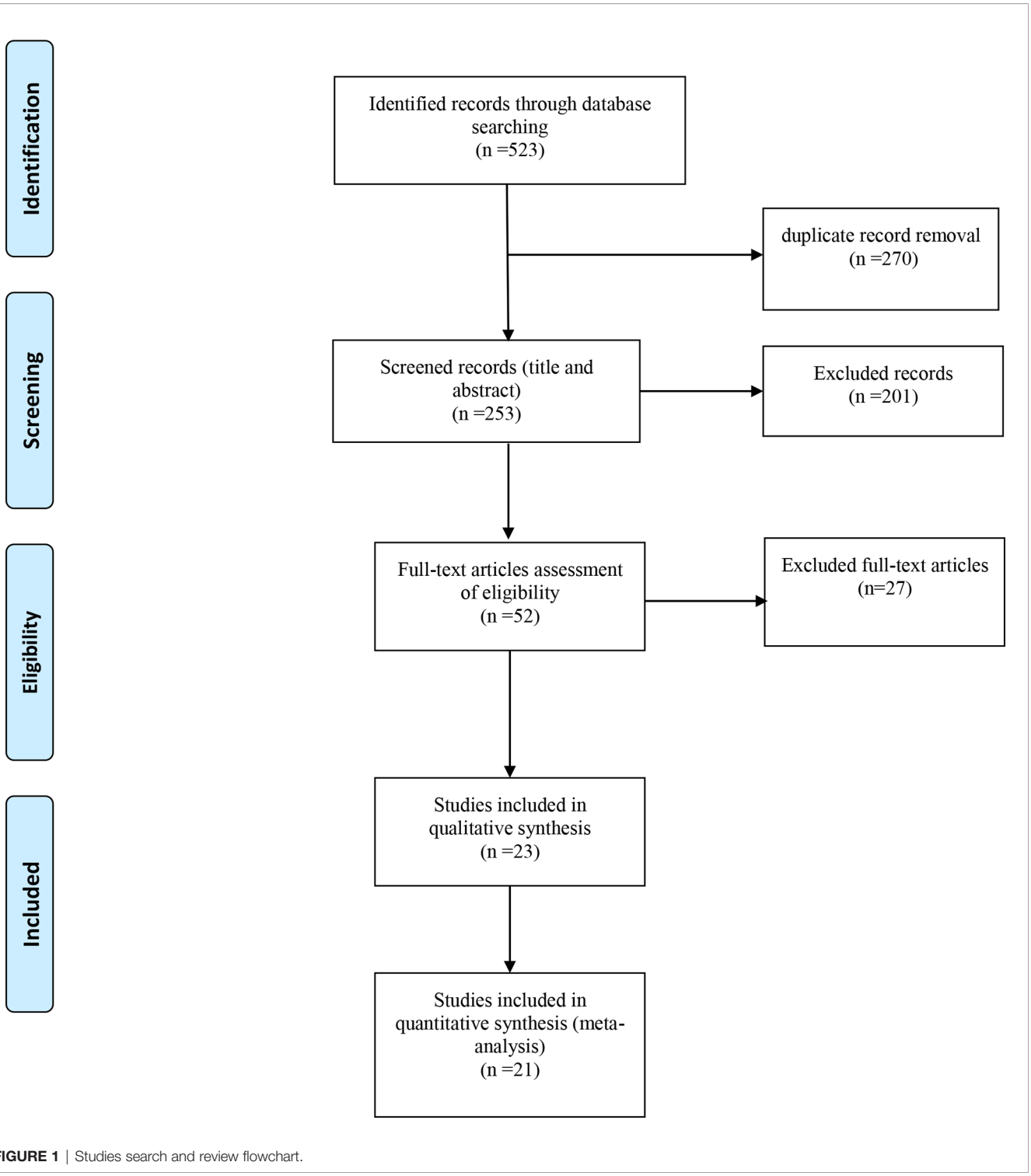


FIGURE 1 | Studies search and review flowchart.

heterogeneity was seen among included studies based on the CMRFs. Based on the fixed-effect model meta-analysis, the overall mean of TC (SMD: 0.22, 95% CI: 0.16, 0.28) and LDL (SMD: 17, 95% CI: 0.13, 0.12) was higher in NWO individuals compared to the normal weight none obese (NWNO) individuals. Based on the random effect meta-analysis, being NWO statistically increased the mean of HOMA (SMD: 0.12, 95% CI: 0.09, 0.32), TG (SMD: 0.13, 95% CI: 0.05, 0.20), SBP (SMD: 0.15, 95% CI: 0.07, 0.23), DBP (SMD: 0.16, 95% CI: 0.03, 0.29). However, the relationship between plasma glucose, HDL and lean mass was not statistically significant (**Figure 3**).

TABLE 1 | General Characteristics of included studies for association of NWO with CMRFs.

Author Year	Country	sample size					Mean Age/ Age Range	Study Population	Definition of NWO*	Q.A
		Total	M	F	NWNO	NWO				
Bellissimo 2019 (15)	USA	289	63	116	26	43	47	Adults/general population	BF: 23% men, 30% women	6
Berg 2015 (16)	Sweden	1471	581	890	1080	266	25-74	Adults /general population	BF: 25% men , 38% women	8
W. K. Cho 2015 (17)	Korea	1700	888	812	1266	144	13-18	Adolescents	upper highest quartile (Q4) of age and sex specific Waist-to-height ratio	8
M. Correa 2020 (18)	Colombia	1354	528	826	961	393	18-32	Adults /general population	BF: 25.5% men , 38.9% women	9
Ramsaran 2017 (19)	West Indies	236	76	160	189	74	18-32	Adults /general population	BF: 23% men , 33% women	6
A. García 2020 (20)	Colombia	1919	955	964	1035	884	13	Children and Adolescents	BF: boys > 23.4%–28.3% and girls > 31.0%–34.1%	7
H. He 2019 (21)	China	2654	(–)	(-)	1916	729	46.9 ± 1380	Adults /general population	BF: 21.4% men , 31.4% women	9
A. Jia 2018 (22)	China	15291	1492	13799	9988	1771	under 75	Adults / general population	BF: ≥24% for men and ≥33% for women	10
Kapoor 2020 (23)	India	1147	619	528	200	364	47.3 ± 7.5	Adults /general population	BF: , ≥20.6% men, ≥33.4% women,	9
Kim 2014 (24)	Korea	12217	5313	6904	3382	1575	44	Adults /general population	BF: ≥20.6% men, ≥33.4% women	8
Kim 2018 (25)	Korea	3949	(–)	(-)	2213	868	(-)	Adults /general population	BF: ≥23.1% men, ≥33.1% women	6
Sohee Kim 2015 (26)	Korea	2078	1141	937	1795	283	53.4	Adults /general population	BF: ≥25.4 % men, ≥31.4 % women	8
H. Zhao 2012 (27)	China	407	(-)	(-)	(-)	(-)	(-)	(-)	BF: ≥25 % men, ≥35 % women	5
Tayefi 2019 (28)	Iran	2439	(–)	(-)	1311	1128	47	Adults /general population	BF: > 25%men, >30% women	9
T. Shirasawa 2019 (8)	Japan	117163	82487	34676	43055	12877	40-64	Adults /general population	Waist-to-height ratio ≥ 0.5	10
A. Romero 2010 (6)	USA	4116	2089	2027	2054	2062	41.3	Adults /general population	BF: ≥23.1 % men, ≥33.3 % women	8
A. S. Olafsdottir 2016 (29)	Iceland	182	96	86	106	76	17.7-18.9	High-school students/ adolescents	BF: > 17.6% men , >31.6% women	5
F. M. Moy 2015 (30)	Malaysia	858	0	858	511	237	40.47 ± 8.9	Adults /teachers	BF: >28.52%	6
K. E. Martinez 2017 (31)	USA	3600	(-)	(-)	1624	288	adults	Adults /general population	BF: 27.8 ± 0.2% men, 40.5 ± 0.2 % women	5
P. Marques-Vidal 2010 (32)	Switzerland	2301	0	2301	1667	173	35-75	Adults/general population whites	BF: ≥38%	7
F. B. Madeira 2013 2013 (4)	Brazil	1222	546	676	1111	111	23-25	young adults	BF: ≥23% men, ≥30% women	10
P. J. Liu 2017 (33)	China	412	0	412	214	198	55.72	Post menopause general population women	third tertile of normal weight body fat	6
C. C. N. Da Silva 2021 (34)	Brazil	787	346	441	553	47	23-25	Young adults	>90th percentile body fat	10

*CMRFs, cardio-metabolic risk factors; M,male; F, female; NWNO, normal weight none obese; NWO, normal weight obese; BF, body fat (based on percentage); USA, United State; Q.A, quality assessment based on the Newcastle Ottawa scale (out of 10 points); * all NWOs had BMIs within normal range.*

Publication Bias

Except for HTN (coefficient =1.70, p-value=0.003), no publication bias was observed in articles studying the association between NWO and CMRFs for dichotomous and continuous data.

Sensitivity Analysis

The sensitivity analysis result indicated that the pooled OR of the relationship between NWO and HTN was not substantially affected by each study (OR:1.23, 95% CI:1.19, 1.27).

DISCUSSION

To our knowledge, this is the first systematic review and meta-analysis that compared CMRFs among NWO and NWNO individuals across the entire population. We found 50% and 42% increased odds of hyperglycemia and diabetes among NWO individuals compared to the NWNOs, respectively. NWO individuals also have 40%, 83%, and 32% increased odds of HTN, dyslipidemia, and reduced HDL levels, respectively. Interestingly, NWO individuals also had an increased odds of hypertriglyceridemia

TABLE 2 | Association of NWO with mean of CMRFs in included studies.

Author, year	Outcome	NWNO		NWO		Effect size		Adjustment
		Mean	SD	Mean	SD	SMD	95% CI	
Bellissimo, 2019 (15)	Plasma glucose	95.7	24	93.1	26.2	0.1	-0.39_0.59	Sex, age and ethnicity
	Plasma insulin	2.7	2.5	3.6	3.2	-0.3	-0.79_0.19	
	TC	193.9	47	201.4	52.4	-0.15	-0.63_0.34	
	HOMA	0.6	0.5	0.8	0.65	-0.33	-0.82_0.16	
	LDL	105.3	41	117.8	45.85	-0.28	-0.77_0.21	
	HDL	72.7	19	63.5	21.61	0.44	-0.05_0.93	
	TG	81.1	57	101.2	62.22	-0.33	-0.82_0.16	
	SBP	119.4	18	118.1	20.3	0.07	-0.42_0.55	
	DBP	74.7	12.5	75.5	13.75	-0.06	-0.55_0.43	
	BMI	23.9	4.59	24.3	4.55	-0.09	-0.57_0.40	
	Lean mass*	50.3	7.65	44.4	16.25	0.43	-0.07_0.92	
	Fat mass*	16.5	9.18	21.5	9.75	-0.52	-1.01_-0.02	
berg, 2015 (16) (male)	Plasma glucose	91.8	17.83	93.6	10.82	-0.11	-0.31_0.08	Age
	TC*	201.08	38.33	216.55	46.52	-0.38	-0.58_-0.18	
	LDL*	119.69	33.28	131.27	50.07	-0.3	-0.5_-0.1	
	HDL*	61.78	0	57.92	23.24	0.32	0.12_0.52	
	TG*	88.57	307.09	106.28	53.29	-0.07	-0.26_0.13	
	SBP*	129	19.81	134	18.04	-0.26	-0.45_-0.06	
	DBP*	80	9.9	85	9.02	-0.52	-0.71_-0.32	
	BMI*	22.8	2.47	24.1	2.7	-0.51	-0.71_-0.32	
	Lean mass	57.5	5.94	57.4	5.41	0.02	-0.18_0.21	
	Fat mass*	15	2.97	20.6	4.51	-1.62	-1.84_-1.4	
	Waist*	84.4	7.92	89.8	7.51	-0.69	-0.89_-0.49	
	Hip*	96.1	5.44	98.9	5.71	-0.51	-0.7_-0.31	
Berg, 2015 (16) (female)	Plasma glucose	88.2	12.17	88.2	10.34	0	-0.19_0.19	
	TC*	204.97	52.28	216.55	33.34	-0.23	-0.42_-0.04	
	LDL*	111.97	52.21	119.69	33.29	-0.16	-0.34_0.03	
	HDL	69.5	26.1	69.5	22.19	0	-0.19_0.19	
	TG*	88.57	59.92	97.42	50.91	-0.15	-0.34_0.04	
	SBP	125	13.52	125	17.24	0	-0.19_0.19	
	DBP	80	13.52	80	8.62	0	-0.19_0.19	
	BMI*	22.1	2.7	24.1	2.87	-0.73	-0.93_-0.54	
	Lean mass	42.8	4.05	42.9	4.02	-0.02	-0.21_0.16	
	Fat mass*	17.6	4.05	23.3	4.88	-1.36	-1.56_-1.16	
	WC*	60.6	8.11	66	7.76	-0.67	-0.86_-0.48	
	Hip*	94.8	7.44	98.9	6.61	-0.56	-0.75_-0.37	
A. García, 2020 (20) (male)	Plasma glucose	83.36	14.95	81.91	17.04	0.09	-0.04_0.22	Age, BMI and pubertal stage
	HDL	49.6	12.99	45.72	11.75	0.31	0.18_0.44	
	TG*	76.72	30.8	85.32	38.13	-0.25	-0.38_-0.12	
	SBP	111.44	14.98	111.46	13.34	0	-0.13_0.13	
	WC*	61.78	5.12	65.85	6.13	-0.73	-0.86_-0.59	
A. García, 2020 (20) (female)	Plasma glucose	81.96	15.16	80.47	16.86	0.09	-0.03_0.22	
	HDL	49.63	12.51	47.15	12.21	0.2	0.08_0.32	
	TG*	87.16	36.61	97.86	62.59	-0.21	-0.34_-0.09	
	SBP	107.81	12.31	110.58	12.36	-0.22	-0.35_-0.1	
	WC*	59.12	5.07	63.94	5.89	-0.88	-1.01_-0.75	

(Continued)

TABLE 2 | Continued

Author, year	Outcome	NWNO		NWO		Effect size		Adjustment
		Mean	SD	Mean	SD	SMD	95% CI	
A. García, 2020 (20)	BMI*	17.86	1.85	19.67	1.91	-0.96	-1.06_-0.87	
	Fat mass*	7.24	3.09	9.89	3.56	-0.8	-0.89_-0.71	
Sohee kim, 2015 (26)	Plasma glucose *	95.7	17.5	99.1	15.9	-0.2	-0.32_-0.07	Age, sex, and smoking status
	TC*	192.5	34.6	199.6	35.5	-0.2	-0.33_-0.08	
	LDL*	117.1	30.6	123.9	32.2	-0.22	-0.35_-0.1	
	HDL*	51	12.8	49.6	11.3	0.11	-0.01_0.24	
	TG*	106.4	65.1	116.6	63.3	-0.16	-0.28_-0.03	
	SBP*	122.5	15.5	128.6	15.7	-0.39	-0.52_-0.27	
	DBP*	76.6	10.1	79.6	9.3	-0.3	-0.43_-0.17	
	BMI*	22.53	4.3	23.9	0.8	-0.34	-0.47_-0.22	
Tayefi, 2019 (28)	Plasma glucose	86.38	35.66	91.3	40.4	-0.13	-0.21_-0.05	Age and sex
	TC	181.93	38.16	189.61	39.14	-0.2	-0.28_-0.12	
	LDL	113.19	33.54	117.67	35.06	-0.13	-0.21_-0.05	
	HDL*	42.09	9.8	45.74	9.43	-0.38	-0.46_-0.3	
	TG*	105	738.93	126	753.96	-0.03	-0.11_0.05	
	SBP*	116.26	16.39	118.1	19.9	-0.1	-0.18_-0.02	
	DBP*	76.35	11.84	75.75	11.85	0.05	-0.03_0.13	
	BMI*	22.62	3.38	23.39	4.18	-0.2	-0.28_-0.12	
	WC*	84.52	9.89	86.01	11.53	-0.14	-0.22_-0.06	
	Hip*	94.22	6.91	96.88	8.86	-0.34	-0.42_-0.26	
A. Romero, 2010 (6) (male)	Plasma glucose	95.6	15.92	96.8	22.32	-0.06	-0.15_0.03	Age and race
	HOMA*	0.84	0.31	1	0.63	-0.32	-0.41_-0.23	
	LDL*	121.8	49.03	132.64	49.11	-0.22	-0.31_-0.13	
	HDL*	49.11	12.1	47.56	12.11	0.13	0.04_0.22	
	TG*	116.03	84.38	113.75	84.51	0.03	-0.06_0.11	
	SBP	122	15.92	125	15.94	-0.19	-0.28_-0.1	
	DBP	74	12.73	76	9.57	-0.18	-0.26_-0.09	
	BMI*	22.7	1.27	23.5	1.27	-0.63	-0.72_-0.54	
	WC*	84.8	6.05	88.9	6.37	-0.66	-0.75_-0.57	
	Lean mass*	55.4	5.73	53	5.74	0.42	0.33_0.51	
	Fat mass*	14.6	1.59	18.5	2.55	-1.83	-1.94_-1.73	
	Hip*	93.2	4.13	94.6	4.14	-0.34	-0.43_-0.25	
A. Romero, W2010 (6) (female)	Plasma glucose	92	22.57	91.1	22.62	0.04	-0.05_0.13	
	HOMA*	0.87	0.32	0.98	0.64	-0.22	-0.3_-0.13	
	LDL*	116.4	49.66	124.13	62.39	-0.14	-0.22_-0.05	
	HDL	58	12.25	57.62	12.28	0.03	-0.05_0.12	
	TG*	100.97	85.46	136.4	228.87	-0.2	-0.29_-0.12	
	SBP	117	19.34	119	20.04	-0.1	-0.19_-0.02	
	DBP	71	9.67	72.1	10.34	-0.11	-0.2_-0.02	
	BMI*	22.1	1.29	23.5	0.97	-1.23	-1.32_-1.13	
	WC*	78.3	6.45	83.3	6.46	-0.77	-0.86_-0.69	
	Lean mass*	40.21	4.19	39.9	3.55	0.08	-0.01_0.17	
	Fat mass*	18.1	1.93	22.1	2.26	-1.9	-2.01_-1.8	
	Hip*	94.4	4.51	97.7	4.85	-0.7	-0.79_-0.62	
K. E. Martinez, 2017 (31)	HOMA*	1.1	2.01	1.6	1.52	-0.26	-0.38_-0.13	Age, sex, race, and year of assessment. moderate physical

(Continued)

TABLE 2 | Continued

Author, year	Outcome	NWNO		NWO		Effect size		Adjustment
		Mean	SD	Mean	SD	SMD	95% CI	
								activity, vigorous physical activity, and smoking
P. Marques-Vidal, 2010 (32)	Plasma insulin	8.56	13.88	8.17	6.97	0.03	-0.13_0.19	Age
	HOMA	3.32	5.71	3.18	2.89	0.03	-0.13_0.18	
	BMI*	21.8	4.08	23	2.63	-0.3	-0.46_-0.15	
	WC*	75.9	8.16	79.9	7.89	-0.49	-0.65_-0.33	
	Hip*	94.4	8.16	96.1	6.57	-0.21	-0.37_-0.06	

CMRFs, cardio-metabolic risk factors; NWNO, normal weight non obese; NWO, Normal weight obesity; M, male; F, female; TC, total cholesterol; HOMA, Homeostatic Model Assessment for Insulin Resistance; LDL, low-density lipoproteins; HDL, high-density lipoproteins; TG, triglyceride; SBP, systolic blood pressure; DBP, diastolic blood pressure; BMI, body mass index; WC, waist circumference; SMD, Standardized Mean Difference; SD, Standard Deviation; CI, Confidence Interval. Plasma glucose, TC, LDL, HDL, TG values are reported in the mg/dL unit. SBP and DBP in mmHg. BMI In kilograms by height (in meters) squared, lean and fat masses in kilograms, Hip and WC in centimeters.

**Statistically significant (p-value < 0.05).*

The SMDs were calculated based on Hedges' g formula.

as high as 90%. The results of other studies, such as Yu et al. (37) on CMRFs across various types of obesity, are comparable to that of ours. In the aforementioned study, the odds of hyperglycemia and diabetes were 40% and 103% in those with central obesity, 78% odds of HTN, and 142% hypertriglyceridemia (37). Although it seems that NWO imposes less a CMRF, some of its complications are comparable to that of obesity (central, general, and combined) (34); furthermore, despite being in a relatively better status than the obese, NWO individuals have a significantly greater CMRFs in comparison to the NWNOs. Moreover, the assessment of NWO-related comorbidities is of particular importance since the prevalence of NWO is exceptionally high (ranging from 5 to 45% based on sex, age, and the definition of NWO) (29, 38, 39). With a prevalence of 45% even among adolescents, NWO acts as a potent risk factor for future comorbidities; Hence, preventing and treating NWO can drastically reduce these comorbidities as well as obesity in adulthood; nonetheless, seemingly, due to lifestyle changes, unhealthy diets and lack of adequate physical activity, and sedentary lifestyle, the prevalence of NWO is increasing in a worrisome manner (40). However, these NWO individuals will go unnoticed and undiagnosed due to the inadequacies of BMI measurement, and despite their high body fat, due to their normal BMI levels, no treatment and preventive measure will be taken until it is too late. Furthermore, although obesity is a well-known associate of metabolic dysregulation, and there have been numerous studies on conditions that can result from obesity, yet the new concept of obesity (in which the weight itself is not as important as the body's fat percent) is not well known nor studied. There have been studies on normal weight obesity regarding the conditions that can arise from it; however, compared to obesity, the number of studies are preliminary and more studies need to be done so that normal weight obesity gets the recognition that it needs as it is imperative for individuals to be well aware of their condition in order to take preventive measures. The public must be educated on the subject of NWO and must know that a normal BMI does not necessarily mean that they are not obese; in fact, they might have normal weight obesity, and regardless of their normal BMI, they are at increased risk of cardiometabolic conditions.

Limitations and Strength

To the best of our knowledge, this is the first systematic review and meta-analysis comparing CMRFs among NWO and NWNO individuals in the entire population (age range 13 to 75), with a sample size of 177,792 proper research methods, it gives a realistic status of NWO globally. Our limitations were the use of manuscripts with an English full text. Furthermore, the unequal number of studies in different countries and differences in measurement and methodological aspects of the included studies resulting in high heterogeneity were among our limitations.

CONCLUSION

The present study illustrated the significant odds of CMRFs among NWO individuals compared to subjects with NWNO. Indicating the inadequacy of the BMI measurement and the need

TABLE 3 | Association of NWO with CMRFs in included studies.

Author Year	Outcome	Definition of outcome	OR (95% CI)**	Adjustment
W. K. Cho 2015 (17) (Male)	HOMA	Fasting glucose (in millimoles per liter) × fasting insulin (in milliunits per liter)/22.5	2.46 (1.21-4.99)	Age, weight, and ALT
W. K. Cho 2015 (17) (Female)	HOMA		1.51 (0.83-2.75)	
M. Correa 2020 (18)	hyperglycemia	FBS ≥5.6 mmol/L [100 mg/dL]	1.31 (0.73-2.33)	Age and sex
	HTN	≥130 mm Hg SBP and/ or DBP 85 mm Hg	1.42 (0.89-2.27)	
	Elevated TG	≥1.7 mmol/L [151 mg/dL]	1.31 (0.62-2.76)	
	Elevated LDL	≥2.6 mmol/L [100 mg/dL]	1.27 (0.85-1.90)	
	Reduced HDL	low HDL: < 1 mmol/L [38.7 mg/dL] in men and 1.3 mmol/L [50.3 mg/dL] in women	2.34 (1.61-3.93)*	
	Cardiometabolic risk Z-score	+ 1 SD above the mean	3.10 (2.06-4.67)*	
	obesity	Waist to hip ratio > 0.49 in men and > 0.50 in women	2.61 (0.69-9.87)	
	Abdominal Obesity	WC ≥ 90 cm in men, and ≥ 80 cm in women	7.27 (1.09-48.60)*	
Ramsaran 2017 (19)	Elevate DBP	high SBP and DBP systolic ≥120 mm Hg	0.98 (0.39-2.48)	Not adjusted
	Elevate SBP	and the diastolic ≥80 mm Hg	1.85 (0.52-5.52)	
	Elevated WC	men ≥94 cm and women ≥80 cm	26.61 (4.75-149.14)*	
H. He 2019 (21)	HTN	SBP≥ 140 and,or DBP ≥90	1.82 (1.43-2.30)*	Age, sex, social economic profiles, lifestyle factors, family history of HTN and other disease status, etc.
A. Jia 2018 (22)	DM	FBS 7.0 ≥ mmol/L; blood glucose 2 h after an OGTT ≥ 11.1 mmol/L; a previous diagnosis of diabetes; or current use of hypoglycemic agents	1.44 (1.10–1.88)*	Age , sex, ethnicity, smoking, alcohol use, exercise, education, yearly family income, family history of disease, and WC
	HTN	SBP≥ 130 and,or DBP ≥85	1.53 (1.27–1.84)*	
	MetS	IDF[1]	1.48 (1.22–1.79)*	
	Elevated Framingham risk	Score ≥ 10%	2.36 (1.76–3.17)*	
Kapoor 2020 (23)	DM	FBS ≥ 126 mg/dl and/or 2-h plasma glucose value of ≥ 200 mg/dl were diagnosed to have diabetes/	2.72 (1.46–5.08)*	Age, sex, tobacco use and alcohol intake
	HTN	SBP≥ 140 and, or DBP ≥90	1.89 (0.92–3.86)	
	Dyslipidemia	taking lipid-lowering medications and/or TC >200 mg/dl and/or LDL >100 mg/dl and/or HDL <40 mg/dl in men and <50 mg/dl in women and/ or TG >200 mg/dl.	2.37 (1.55–3.64)*	
Kim, 2014 (24) (Male)	DM	fasting blood glucose ≥ 126 mg/dl or treatment of the disease	1.38 (1.04 -1.83)*	Age and lifestyle factors
	HTN	SBP≥ 140 and, or DBP ≥90	1.70 (1.42 - 2.02)*	
	dyslipidemia	total cholesterol ≥ 240 mg/dl and/or high-density lipoprotein (HDL) cholesterol <40 mg/dl and/or triglyceride ≥ 150 mg/dl or treatment of dyslipidemia**	2.69 (2.29 - 3.17)*	
	MetS		2.50 (2.10 - 2.97)*	
	Mets risk factor above 1	plus 1 metabolic risk odds	3.54 (2.89 - 4.34)*	
Kim, 2014 (24) (Female)	DM	fasting blood glucose ≥ 126 mg/dl or treatment of the disease	1.72 (1.30 - 2.29)*	
	HTN	SBP≥ 130 mmHg and, or DBP ≥85 mmHg	1.52 (1.25 - 1.86)*	
	dyslipidemia	total cholesterol ≥ 240 mg/dl and/or high-density lipoprotein (HDL) cholesterol <40 mg/dl and/or triglyceride ≥ 150 mg/dl or treatment of dyslipidemia**	1.70 (1.40 - 2.06)*	
	MetS		1.80 (1.48 - 2.20)*	
	Mets risk factor above 1	plus 1 metabolik risk odds	2.47 (2.01 - 3.03)*	
Kim 2018 (25)	MetS	IDF	1.83 (1.21 - 2.76)*	Potential confounders
	Plaque formation risk	(–)	1.46 (1.027 - 2.07)*	

(Continued)

TABLE 3 | Continued

Author Year	Outcome	Definition of outcome	OR (95% CI)**	Adjustment
H. Zhao 2012 (27)	HTN risk	(-)	2.18*	Age and sex
	Hyperglycemia		2.12*	
	dyslipidemia		2.08*	
	Hyperuricemia		3.49*	
Tayefi 2019 (28)	Risk of metabolically abnormal phenotype	(-)	2.02 (1.68-2.42)*	Age and sex
	Cardiac risk (Q)	QRISK calculated online by using the Framingham risk equation	6 (4.45-8.08)*	
T. Shirasawa 2019 (8) (Male)	DM	FBS ≥ 126 mg/dl, random plasma glucose ≥ 200 mg/dl, HbA1c (National Glycohemoglobin Standardization Program) ≥ 6.5%, or receiving medical treatment for DM	1.35 (1.25-1.46)*	Age, weight, smoking status, alcohol intake, and physical activity
	HTN	SBP≥ 140 and, or DBP ≥90 or taking medication for HTN	1.22 (1.17-1.27)*	
	Dyslipidemia	as LDL-C ≥ 140 mg/dl, HDL-C < 40 mg/dl, TG ≥ 150 mg/dl, or taking medication for dyslipidemia	1.84 (1.74-1.89)*	
T. Shirasawa 2019 (8) (Female)	DM	FBS ≥ 126 mg/dl, random plasma glucose ≥ 200 mg/dl, HbA1c ≥ 6.5%, or receiving medical treatment for DM	1.60 (1.35-1.90)*	
	HTN	SBP≥ 140 and,or DBP ≥90 or taking medication for hypertension	1.23 (1.16-1.31)*	
	dyslipidemia	as LDL-C ≥ 140 mg/dl, HDL-C < 40 mg/dl, triglycerides ≥ 150 mg/dl, or taking medication for dyslipidemia	1.60 (1.52-1.69)*	
A. S. Olafsdottir (29)	Mets	IDF	2.2 (1.2-3.9)	not adjusted
F. M. Moy 2015 (30)	Elevated TG	TG ≥ 1.7 mmol/L	2.51 (1.47–4.29)*	age and ethnicity
	Reduced HDL	HDL-C ≤ 1.3 mmol/L in women	1.09 (0.75–1.58)	
	Hypertension	systolic ≥130 mmHg and/or diastolic ≥85 mmHg or on antihypertensive treatment	1.63 (1.15–2.31)*	
	Hyperglycemia	FBG ≥ 5.6 mmol/L	1.67 (0.90–3.08	
	Mets	IDF	1.70 (0.87–3.32)	
	Hypercholesterolemia	(–)	2.22 (0.21–23.20)	
	Diabetes	Hyperglycemia: FBG ≥ 5.6 mmol/L.	1.28 (0.34–4.92)	
P. Marques-Vidal 2010 (32)	Elevated TG	TG ≥ 1.7 mmol/L [151 mg/dL] and/or LDL ≥ 2.6 mmol/L [100 mg/dL] (in the presence of myocardial infarction, stroke, coronary artery disease or diabetes) and ≥ 4.2 mmol/L [163 mg/dL] in other cases and/or hypolipidaemic drug treatment	2.21 (1.43-3.42)*	
	Low HDL	1 mmol/L in men and 1.3 mmol/L in women	2.10 (1.23-3.57)*	
	HTN	(–)	1.38 (0.97-1.98)*	
	Hyperglycemia	fasting hyperglycemia; HOMA > 4.88 (90th percentile in men) or >3.57 (90th percentile in women)	1.63 (1.10-2.42)*	
	dyslipidemia	HDL < 1 mmol/L in men and 1.3 mmol/L in women and/or TG ≥ 1.7 mmol/L [151 mg/dL] and/or LDL ≥ 2.6 mmol/L [100 mg/dL] (in the presence of myocardial infarction, stroke, coronary artery disease or diabetes) and ≥ 4.2 mmol/L [163 mg/dL] in other cases and/or hypolipidaemic drug treatment	1.90 (1.34-2.68)*	
	CMRF	the presence of at least two of the following: HTN ; TG ≥ 1.7 mmol/L; HDL cholesterol < 1 mmol/L [38.7 mg/dL] in men and 1.3 mmol/L [50.3 mg/dL] in women; fasting hyperglycemia; HOMA > 4.88 (90th percentile in men) or >3.57 (90th percentile in women) and CRP > 5.2 mg/L (90th percentile in men) or >6.1 mg/L (90th percentile in women) , definition 1.	1.37 (0.97-1.95)*	

(Continued)

TABLE 3 | Continued

Author Year	Outcome	Definition of outcome	OR (95% CI)**	Adjustment
		A second definition of metabolic risk was also applied, using the same criteria but with HOMA > 5.0 and CRP > 4.0 mg/L		
	Abdominal obesity	(–)	2.64 (1.73–4.04)*	
F. B. Madeira 2013 (4)	Elevated TG	TG ≥ 150 mg/dL, use of lipid medications or self-reported diagnosis of hypertriglyceridemia	1.89 (0.97-3.70)	age, sex, skin colour, early and adult life variables (alcohol consumption, family income, schooling, marital
	Reduced HDL	HDL < 40 mg/dL for men and ,50 mg/dL for women	1.53 (1.00-2.34)	status, smoking, percentage of fat in the diet and physical activity
	HTN	SBP ≥ 130 mmHg and/ DBP ≥ 85 mmHg, current usage of antihypertensive drugs or previous diagnosis of hypertension	1.17 (0.65-2.13)	
	Hyperglycemia	high fasting blood glucose (≥100 mg/dL), current use of anti-diabetic medication or previously diagnosed diabetes	2.68 (1.01-7.12)*	
	HOMA	(–)	4.91 (1.85-13.04)*	
	Mets	IDF	8.89 (3.32-4.47)*	
	Elevated WC	central obesity (WC ≥ 90 cm for men and ≥ 80 cm for women	9.27 (5.32-16.15)*	
P. J. Liu 2017 (33)	Elevated TG	TGs ≥1.7 mmol/L	2.13 (1.10-4.12)*	age, smoking status ,drinking status,
	Reduced HDL	HDL <1.30 mmol	1.04 (0.61-1.75)	total cholesterol, LDL-c, high
	HTN	blood pressure ≥130/85 mmHg or current antihypertensive medication use	2.06 (1.09-3.90)*	sensitivity C-reactive protein, and the remaining non-adipose MetS
	Hyperglycemia	FBS ≥5.6 mmol/L, type 2 diabetes mellitus previously diagnosed by a physician, or current antidiabetic medication use	1.44 (0.77-2.68)	components, body fat percentage
	Mets risk factor above 2	IDF	2.00 (1.19-3.33)*	
C. C. N. Da Silva 2021 (34)	Elevated TG	TG levels above 150 mg/dL or use of lipid-lowering drugs	1.77 (1.12-2.79)*	total calories, family income, added sugar intake, total lipids intake, and
	Reduced HDL	<40 mg/dL for men and <50 mg/dL for women or use of lipid-lowering drugs	1.27 (0.98-1.65)	physical activity
	HTN	SBP > 130 mmHg, DBP > 85 mmHg, or use of antihypertensive drugs	1.44 (0.94-2.21)	
	Hyperglycemia	FBS > 100 mg/dL or use of glucose-lowering drugs	1.48 (0.96-1.65)	
	Mets	IDF	1.87 (1.36-2.57)*	
	Elevated WC	WC 90 cm for men and 80 cm for women	9.27 (5.32-16.15)*	

NWO, Normal weight obesity, M, male, F, female, TC, total cholesterol, HOMA, Homeostatic Model Assessment for Insulin Resistance, LDL, low-density lipoproteins, HDL, high-density lipoproteins, TG, triglyceride, SBP, systolic blood pressure, DBP, diastolic blood pressure, BMI, body mass index, WC, waist circumference, Mets, metabolic syndrome, DM, diabetes mellitus, HTN, hypertension, CM, centimeters, IDF, International Diabetes Federation, OR, odds ratio, CI, Confidence Interval.

**Statistically significant (p-value < 0.05).*

***OR calculated for NWO compare to NWNO.*

[1]MetS is defined based on the criteria of IDF.

TABLE 4 | Stratified meta-analysis of association between NWO with CMRFs according to sex.

variables	No study	Sample size	Pooled odds ratio (95% CI)	Heterogeneity assessment		
				I-squared %	Model	P-value
Hyperglycemia						
Overall	6	6,934	1.50 (1.23, 1.76)*	0.00	Fixed	0.958
Both sexes	3	3,363	1.46 (1.15, 1.78)	0.00	Fixed	0.684
Female	3	3,571	1.58 (1.10, 2.07)*	0.00	Fixed	0.937
HTN						
Overall	13	155,397	1.40 (1.28, 1.51)*	57.30	Random	0.005
Both sexes	5	21,659	1.56 (1.35, 1.78)*	0.00	Fixed	0.587
Male	2	87,800	1.43 (0.96, 1.90)	89.50	Random	0.002
Female	6	45,938	1.25 (1.18, 1.33)*	25.80	Fixed	0.241
High TG						
Overall	6	6,934	1.90 (1.44, 2.35)*	0.00	Fixed	0.785

(Continued)

TABLE 4 | Continued

variables	No study	Sample size	Pooled odds ratio (95% CI)	Heterogeneity assessment		
				I-squared %	Model	P-value
Both sexes	3	3,363	1.65 (1.05, 2.24)*	0.00	Fixed	0.746
Female	3	3,571	2.26 (1.55, 2.98)*	0.00	Fixed	0.924
Low HDL						
Overall	6	6,934	1.28 (1.06, 1.49)*	29.80	Fixed	0.212
Both sexes	3	3,363	1.38 (1.09, 1.67)*	38.20	Fixed	0.198
Female	3	3,571	1.15 (0.82, 1.47)	27.40	Fixed	0.252
Diabetes						
Overall	7	146,676	1.39 (1.30, 1.49)*	9.30	Fixed	0.358
Both sexes	2	16,438	1.49 (1.11, 1.87)*	45.5	Fixed	0.175
Male	2	87,800	1.35 (1.25, 1.45)*	0.00	Fixed	0.886
Female	3	42,438	1.62 (1.38, 1.86)*	0.00	Fixed	0.878
Metabolic syndrome						
Overall	6	36,854	1.92 (1.58, 2.26)*	68.40	Random	0.002
Both sexes	5	23,688	1.82 (1.38, 2.26)*	66.40	Random	0.018
Female	2	7,762	1.79 (1.44, 2.13)*	0.00	Fixed	0.878
Dyslipidemia						
Overall	7	135,276	1.83 (1.61, 2.04)*	80.00	Random	<0.001
Both sexes	3	2,737	1.73 (0.86, 2.60)*	45.70	Random	0.159
Male	2	87,800	2.23 (1.40, 3.06)*	92.80	Random	<0.001
Female	4	44,739	1.61 (1.52, 1.69)*	80.00	Random	<0.001

**Statistically significant (P-value < 0.05)*

HDL, high-density lipoproteins; TG, triglyceride; HTN, hypertension; No, number; CI, confidence interval; CMRFs, cardio-metabolic risk factors; NWO, Normal weight obesity; HDL, high-density lipoproteins; TG, triglyceride; HTN, hypertension; No, number; CI, confidence interval; CMRFs, cardio-metabolic risk factors; NWO, Normal weight obesity.

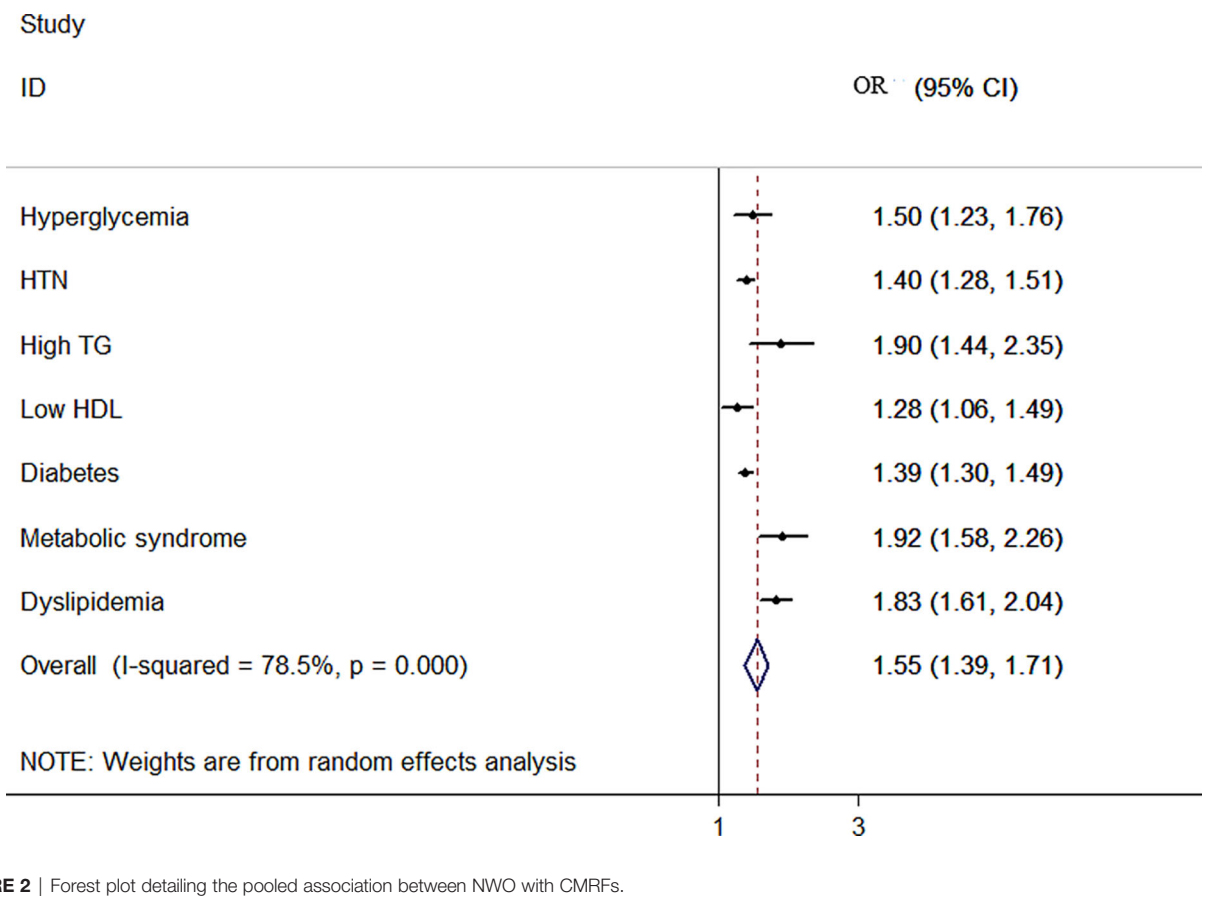


FIGURE 2 | Forest plot detailing the pooled association between NWO with CMRFs.

TABLE 5 | Stratified meta-analysis of association between NWO with mean of CMRFs according to sex.

Variables	No study	Sample size	SMD (95% CI)	Heterogeneity assessment		
				I-squared %	Model	P-value
Plasma glucose						
Overall	9	12,312	0.03 (-0.04, 0.10)	66.30	Random	0.003
Both sexes	3	4,806	0.14 (0.07, 0.21)*	0.00	Fixed	0.410
Male	3	3,510	0.02 (-0.04, 0.09)	54.90	Fixed	0.109
Female	3	3,996	-0.05 (-0.11, 0.01)	0.00	Random	0.672
Total cholesterol						
Overall	5	6,277	0.22 (0.16, 0.28)*	0.00	Fixed	0.559
Both sexes	3	3,132	0.20 (0.13, 0.26)*	0.00	Fixed	0.976
HOMA						
Overall	5	10,306	0.21 (0.09, 0.32)*	73.10	Random	0.005
Both sexes	2	3,889	0.26 (0.14, 0.38)*	0.00	Fixed	0.765
Female	2	4,390	0.10 (-0.13, 0.34)	85.9	Random	0.008
LDL						
Overall	7	10,393	0.17 (0.13, 0.21)*	0.00	Fixed	0.503
Both	3	4,806	0.15 (0.09, 0.22)*	0.00	Fixed	0.437
Male	2	2,608	0.23 (0.15, 0.34)*	0.00	Fixed	0.465
Female	2	2,979	0.14 (0.06, 0.21)*	0.00	Fixed	0.865
HDL						
Overall	8	12,312	-0.08 (-0.26, 0.10)	94.70	Random	<0.001
Both sexes	3	4,806	-0.01 (-0.44, 0.41)	95.90	Random	<0.001
Male	2	3,510	-0.21 (-0.39, -0.03)	80.70	Random	0.023
Female	3	3,996	-0.08 (-0.20, 0.04)	94.70	Random	<0.001
TG						
Overall	9	12,312	0.13 (0.05, 0.20)*	70.30	Random	0.001
Both sexes	3	4,806	0.09 (-0.02, 0.21)	50.30	Fixed	0.133
Male	3	3,510	0.05 (-0.09, 0.12)	83.20	Random	0.003
Female	3	3,996	0.20 (0.13, 0.26)*	0.00	Fixed	0.858
SBP						
Overall	6	12,312	0.15 (0.07, 0.23)*	71.80	Random	<0.001
Both sexes	3	4,806	0.19 (-0.06, 0.44)	87.20	Random	<0.001
Male	3	3,510	0.14 (0.01, 0.28)*	70.80	Random	0.033
Female	3	3,996	0.12 (0.05, 0.19)*	55.20	Fixed	0.107
DBP						
Overall	5	10,393	0.16 (0.03, 0.29)*	86.50	Random	<0.001
Both sexes	3	4,806	0.10 (-0.18, 0.40)	90.60	Random	<0.001
Male	2	2,608	0.33 (0.004, 0.66)*	89.50	Random	0.002
Female	2	2,979	0.09 (0.01, 0.17)*	7.10	Fixed	0.300
Lean mass						
Overall	5	5,876	-0.16 (-0.37, 0.04)	90.20	Random	<0.001
Male	2	2,608	-0.22 (-0.62, 0.16)	92.60	Random	<0.001
Female	2	2,979	-0.06 (-0.14, 0.01)	0.00	Fixed	0.323
Fat mass						
Overall	6	7,795	1.36 (0.89, 1.82)*	98.50	Random	<0.001
Both sexes	2	2,208	0.79 (0.69, 0.88)*	12.00	Fixed	0.284
Male	2	2,608	1.75 (1.54, 1.95)*	66.30	Random	0.085
Female	2	2,979	1.63 (1.10, 2.17)*	95.50	Random	<0.001
Waist circumference						
Overall	8	12,246	0.62 (0.42, 0.83)*	95.80	Random	<0.001
Male	3	N.R	0.68 (0.61, 0.75)	0.716	Fixed	0.716
Female	4	N.R	0.71 (0.56, 86.50)	80.30	Random	0.002
Hip circumference						
Overall	5	12,246	0.44 (0.28, 0.60)*	91.00	Random	<0.001
Male	2	3,510	0.36 (0.24, 0.55)	57.80	Fixed	0.124
Female	2	6,297	0.49 (0.18, 0.80)	93.10	Random	<0.001

**Statistically significant (P-value < 0.05).*

HOMA, Homeostatic Model Assessment for Insulin Resistance; LDL, low-density lipoproteins; HDL, high-density lipoproteins; TG, triglyceride; SBP, systolic blood pressure; DBP, diastolic blood pressure; HTN, hypertension; No, number; CI, confidence interval; CMRFs, cardio-metabolic risk factors; NWO, Normal weight obesity; HDL, high-density lipoproteins; TG, triglyceride; HTN, hypertension; No, number; CI, confidence interval; CMRFs, cardio-metabolic risk factors; NWO, Normal weight obesity; SMD, Standardized Mean Difference; N.R, Not Reported.

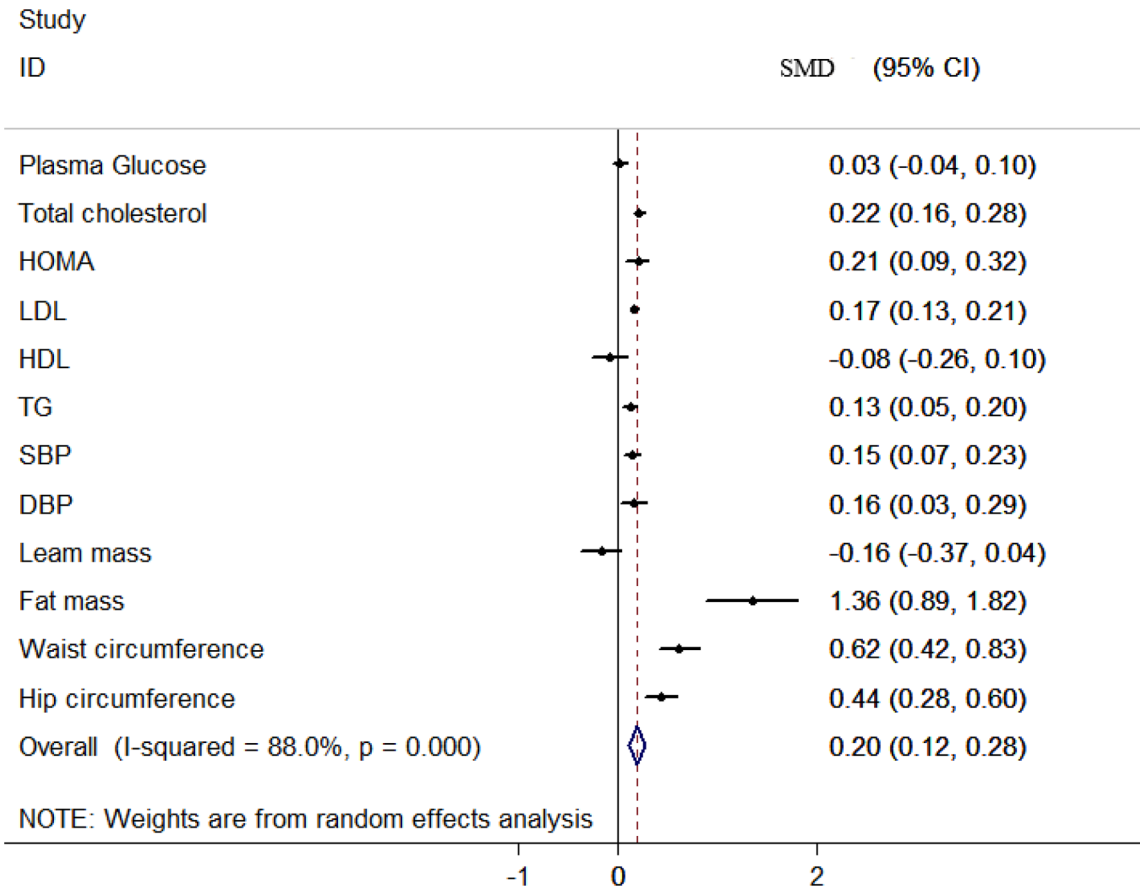


FIGURE 3 | Forest plot detailing the pooled association between NWO with mean CMRFs.

for body fat assessment instead, for a better risk assessment. Furthermore, the necessity of preventive measures and interventions to significantly reduce the burden of the aforementioned condition is essential to avoid the upcoming obesity pandemic.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

AUTHOR CONTRIBUTIONS

NK, MQ, and OT-M designed the study. NK and SN searched the databases. NK and SN screened and extracted the data. ES screened and analyzed the data. MQ, NK, OTM, RK, and MHB prepared the results. NK, MQ, and ZA wrote the paper. All other authors read and approved the final manuscript.

FUNDING

This study was funded by Alborz University of Medical Sciences.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2022.857930/full#supplementary-material

REFERENCES

1. Kelly T, Yang W, Chen C-S, Reynolds K, He J. Global Burden of Obesity in 2005 and Projections to 2030. *Int J Obes* (2008) 32(9):1431–7. doi: 10.1038/ijo.2008.102
2. Apovian CM. Obesity: Definition, Comorbidities, Causes, and Burden. *Am J Manag Care* (2016) 22(7 Suppl):s176–85.
3. Tremmel M, Gerdtham U-G, Nilsson PM, Saha S. Economic Burden of Obesity: A Systematic Literature Review. *Int J Environ Res Public Health* (2017) 14(4):435. doi: 10.3390/ijerph14040435
4. Madeira FB, Silva AA, Veloso HF, Goldani MZ, Kac G, Cardoso VC, et al. Normal Weight Obesity Is Associated With Metabolic Syndrome and Insulin Resistance in Young Adults From a Middle-Income Country. *PloS One* (2013) 8(3):e60673. doi: 10.1371/journal.pone.0060673

5. Jean N, Somers VK, Sochor O, Medina-Inojosa J, Llano EM, Lopez-Jimenez F. Normal-Weight Obesity: Implications for Cardiovascular Health. *Curr Atheroscl Rep* (2014) 16(12):1–8. doi: 10.1007/s11883-014-0464-7
6. Romero-Corral A, Somers VK, Sierra-Johnson J, Korenfeld Y, Boarin S, Korinek J, et al. Normal Weight Obesity: A Risk Factor for Cardiometabolic Dysregulation and Cardiovascular Mortality. *Eur Heart J* (2010) 31(6):737–46. doi: 10.1093/eurheartj/ehp487
7. Rothman KJ. BMI-Related Errors in the Measurement of Obesity. *Int J Obes* (2008) 32(3):S56–S9. doi: 10.1038/ijo.2008.87
8. Shirasawa T, Ochiai H, Yoshimoto T, Nagahama S, Kobayashi M, Ohtsu I, et al. Associations Between Normal Weight Central Obesity and Cardiovascular Disease Risk Factors in Japanese Middle-Aged Adults: A Cross-Sectional Study. *J Health Population Nutr* (2019) 38(1):1–7. doi: 10.1186/s41043-019-0201-5
9. Suliga E, Kozieł D, Głuszek S. Prevalence of Metabolic Syndrome in Normal Weight Individuals. *Ann Agric Environ Med* (2016) 23(4):631–5. doi: 10.5604/12321966.1226858
10. Oliveros E, Somers VK, Sochor O, Goel K, Lopez-Jimenez F. The Concept of Normal Weight Obesity. *Prog Cardiovasc Dis* (2014) 56(4):426–33. doi: 10.1016/j.pcad.2013.10.003
11. Finkelstein EA, Khavjou OA, Thompson H, Trogdon JG, Pan L, Sherry B, et al. Obesity and Severe Obesity Forecasts Through 2030. *Am J Prev Med* (2012) 42(6):563–70. doi: 10.1016/j.amepre.2011.10.026
12. Luhar S, Timæus IM, Jones R, Cunningham S, Patel SA, Kinra S, et al. Forecasting the Prevalence of Overweight and Obesity in India to 2040. *PloS One* (2020) 15(2):e0229438. doi: 10.1371/journal.pone.0229438
13. Robinson E. Overweight But Unseen: A Review of the Underestimation of Weight Status and a Visual Normalization Theory. *Obes Rev* (2017) 18 (10):1200–9. doi: 10.1111/obr.12570
14. Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring Inconsistency in Meta-Analyses. *Bmj* (2003) 327(7414):557–60. doi: 10.1136/bmj.327.7414.557
15. Bellissimo MP, Cai Q, Ziegler TR, Liu KH, Tran PH, Vos MB, et al. Plasma High-Resolution Metabolomics Differentiates Adults With Normal Weight Obesity From Lean Individuals. *Obesity* (2019) 27(11):1729–37. doi: 10.1002/oby.22654
16. Berg C, Strandhagen E, Mehlig K, Subramoney S, Lissner L, Björck L. Normal Weight Adiposity in a Swedish Population: How Well is Cardiovascular Risk Associated With Excess Body Fat Captured by BMI? *Obes Sci Pract* (2015) 1:50–8. doi: 10.1002/osp4.4
17. Cho WK, Kim H, Lee HY, Han KD, Jeon YJ, Jung IA, et al. Insulin Resistance of Normal Weight Central Obese Adolescents in Korea Stratified by Waist to Height Ratio: Results From the Korea National Health and Nutrition Examination Surveys 2008–2010. *Int J Endocrinol* (2015) 2015:158758. doi: 10.1155/2015/158758
18. Correa-Rodríguez M, González-Ruíz K, Rincón-Pabón D, Izquierdo M, García-Hermoso A, Agostinis-Sobrinho C, et al. Normal-Weight Obesity Is Associated With Increased Cardiometabolic Risk in Young Adults. *Nutrients* (2020) 12(4):1106. doi: 10.3390/nu12041106
19. Ramsaran C, Maharaj RG. Normal Weight Obesity Among Young Adults in Trinidad and Tobago: Prevalence and Associated Factors. *Int J Adolesc Med Health* (2017) 29. doi: 10.1515/ijamh-2015-0042
20. García-Hermoso A, Agostinis-Sobrinho C, Camargo-Villalba GE, González-Jiménez NM, Izquierdo M, Correa-Bautista JE. Normal-Weight Obesity Is Associated With Poorer Cardiometabolic Profile and Lower Physical Fitness Levels in Children and Adolescents. *Nutrients* (2020) 12:1171. doi: 10.3390/nu12041171
21. He H, Pan L, Liu F, Ma J, Hu Z, Wang L. Expanded Normal Weight Obesity and Blood Pressure in Chinese Adults: A Community-Based Crosss-Ectional Study. *Austral J Prim Health* (2019) 25. doi: 10.1071/PY18166
22. Jia A, Xu S, Xing Y, Zhang W, Yu X, Zhao Y. Prevalence and Cardiometabolic Risks of Normal Weight Obesity in Chinese Population: A Nationwide Study. *Nutr Metab Cardiovasc Dis* (2018) 28:1045–53. doi: 10.1016/j.numecd.2018.06.015
23. Kapoor N, Lotfaliany M, Sathish T, Thankappan KR, Thomas N, Furler J, et al. Prevalence of Normal Weight Obesity and its Associated Cardio-Metabolic Risk Factors–Results From the Baseline Data of the Kerala Diabetes Prevention Program (KDPP). *PloS One* (2020) 15(8):e0237974. doi: 10.1371/journal.pone.0237974
24. Kim MK, Han K, Kwon HS, Song KH, Yim HW, Lee WC, et al. Normal Weight Obesity in K Orean Adults. *Clin Endocrinol* (2014) 80(2):214–20. doi: 10.1111/cen.12162
25. Kim S, Joo HJ, Shim W-J, Lee J. Normal Weight Obesity and Metabolic Syndrome Risk in Korean Adults: 5-Year Longitudinal Health Checkup Study. *Circulation* (2018) 138(Suppl_1):A13448–A.
26. Kim S, Kyung C, Park JS, Lee S-P, Kim HK, Ahn CW, et al. Normal-Weight Obesity is Associated With Increased Risk of Subclinical Atherosclerosis. *Cardiovasc Diabetol* (2015) 14(1):1–9. doi: 10.1186/s12933-015-0220-5
27. Zhao H, Leng S, Liu Y, Sun G, Shujun Y. Relationship Between Normal Weight Obesity and Cardiovascular Risk Factors. *Chin J Health Manage* (2012) 6(4):255–8. doi: 10.3760/cma.j.issn.1674-0815.2012.04.011
28. Tayefi M, Tayefi B, Darroudi S, Mohammadi-Bajgiran M, Mouhebati M, Heidari-Bakavoli A, et al. There Is an Association Between Body Fat Percentage and Metabolic Abnormality in Normal Weight Subjects: Iranian Large Population. *Trans Metab Syndrome Res* (2019) 2(1):11–6. doi: 10.1016/j.tmsr.2019.08.001
29. Olafsdottir AS, Torfadottir JE, Arngrimsson SA. Health Behavior and Metabolic Risk Factors Associated With Normal Weight Obesity in Adolescents. *PloS One* (2016) 11(8):e0161451. doi: 10.1371/journal.pone.0161451
30. Moy FM, Loh DA. Cardiometabolic Risks Profile of Normal Weight Obese and Multi-Ethnic Women in a Developing Country. *Maturitas* (2015) 81 (3):389–93. doi: 10.1016/j.maturitas.2015.04.011
31. Martinez KE, Tucker LA, Bailey BW, LeCheminant JD. Expanded Normal Weight Obesity and Insulin Resistance in US Adults of the National Health and Nutrition Examination Survey. *Maturitas* (2017) 81(3):389–93. doi: 10.1016/j.maturitas.2015.04.011
32. Marques-Vidal P, Pécoud A, Hayoz D, Paccaud F, Mooser V, Waeber G, et al. Normal Weight Obesity: Relationship With Lipids, Glycaemic Status, Liver Enzymes and Inflammation. *Nutrition Metab Cardiovasc Dis* (2010) 20 (9):669–75. doi: 10.1016/j.numecd.2009.06.001
33. Liu PJ, Ma F, Lou HP, Zhu YN. Normal-Weight Central Obesity Is Associated With Metabolic Disorders in Chinese Postmenopausal Women. *Asia Pac J Clin Nutr* (2017) 26(4):692–7. doi: 10.1016/j.numecd.2009.06.001
34. da Silva Coelho CCN, Bragança MLBM, de Oliveira BR, Bettiol H, Barbieri MA, Cardoso VC, et al. Incidence of Metabolic Syndrome in Adults With Healthy Weight, Normal Weight Obesity, and Overweight/Obesity. *Nutrition* (2021) 85:111134. doi: 10.1016/j.nut.2020.111134
35. Xu S, Ming J, Jia A, Yu X, Cai J, Jing C, et al. Normal Weight Obesity and the Risk of Diabetes in Chinese People: A 9-Year Population-Based Cohort Study. *Sci Rep* (2021) 11(1):1–8. doi: 10.1038/s41598-021-85573-z
36. Wiklund P, Törmäkangas T, Shi Y, Wu N, Vainionpää A, Alen M, et al. Normal-Weight Obesity and Cardiometabolic Risk: A 7-Year Longitudinal Study in Girls From Prepuberty to Early Adulthood. *Obesity* (2017) 25 (6):1077–82. doi: 10.1002/oby.21838
37. Yu S, Xing L, Du Z, Tian Y, Jing L, Yan H, et al. Prevalence of Obesity and Associated Risk Factors and Cardiometabolic Comorbidities in Rural Northeast China. *BioMed Res Int* (2019) 2019:6509083. doi: 10.1155/2019/6509083
38. Marques-Vidal P, Chiolero A, Paccaud F. Large Differences in the Prevalence of Normal Weight Obesity Using Various Cut-Offs for Excess Body Fat. *Eur e-J Clin Nutr Metab* (2008) 3:e159–62. doi: 10.1016/j.eclnm.2008.05.003
39. Conus F, Rabasa-Lhoret R, Peronnet F. Characteristics of Metabolically Obese Normal-Weight (MONW) Subjects. *Appl Physiol Nutrition Metab* (2007) 32 (1):4–12. doi: 10.1139/h06-092
40. Cota BC, Suhett LG, Leite NN, Pereira PF, Ribeiro SAV. Franceschini SdCC. Cardiometabolic Risk and Health Behaviours in Adolescents With Normal-Weight Obesity: A Systematic Review. *Public Health Nutr* (2020) 24(5):870–81. doi: 10.1017/S1368980020004863

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers | Frontiers in Endocrinology

ORIGINAL RESEARCH
published: 12 April 2022
doi: 10.3389/fendo.2022.851369

A Preliminary Study on Infrared Thermograph of Metabolic Syndrome

Meng-jiao Gao[1†], Hui-zhong Xue[1†], Rui Cai[2], Bi-yao Jiang[1], Bao-hong Mi[3], Zong-jun Chen[1], Yin-chun Shi[1], Yong-hua Xiao[4] and Wen-zheng Zhang[4*]*

[1] The First Clinical Medical School, Beijing University of Chinese Medicine, Beijing, China, [2] Beijing Wholelife Medical Science Co., Ltd., Beijing, China, [3] School of Acupuncture-Moxibustion and Tuina, Beijing University of Chinese Medicine, Beijing, China, [4] Dongzhimen Hospital, Beijing University of Chinese Medicine, Beijing, China

OPEN ACCESS

Edited by:
Ozra Tabatabaei-Malazy, Tehran University of Medical Sciences, Iran

Reviewed by:
Solaleh Emamgholipour, Tehran University of Medical Sciences, Iran
Noushin Fahimfar, Tehran University of Medical Sciences, Iran
Seyed Pezhman Madani, Iran University of Medical Sciences, Iran

***Correspondence:**
Yong-hua Xiao
water_aqua@sina.com
Wen-zheng Zhang
zwz7857@sina.com

[†]These authors have contributed equally to this work and share first authorship

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *09 January 2022*
Accepted: *24 February 2022*
Published: *12 April 2022*

Citation:
Gao M-j, Xue H-z, Cai R, Jiang B-y, Mi B-h, Chen Z-j, Shi Y-c, Xiao Y-h and Zhang W-z (2022) A Preliminary Study on Infrared Thermograph of Metabolic Syndrome. Front. Endocrinol. 13:851369. doi: 10.3389/fendo.2022.851369

Objective: To explore the temperature distribution characteristics of the face, palms, feet and the trunk area of metabolic syndrome (MS) through infrared thermography (IRT) and provide evidence for the application of IRT in the assistant evaluation of MS population.

Methods: We collected thermographs of 184 participants (91 males, 93 females) and further divided participants of each gender into 4 groups according to the number of abnormal metabolic indexes. Mean temperatures of 6 Region of Interests (ROIs) (face, anterior trunk, bilateral palms and dorsum of feet) were calculated. Comparisons of the mean temperatures between genders, among groups and ROIs were carried out.

Results: Male participants had higher mean temperature in their face, palms ($P<0.01$) and dorsum of feet ($P<0.05$), and lower mean temperature in the anterior trunk ($P<0.01$). Female participants with MS had higher mean temperature in their palms and dorsum of feet ($P<0.01$) and lower mean temperature in the anterior trunk ($P<0.01$) than normal participants. Similar tendencies were shown in the mean temperature of the left palms and trunk of MS males. With the increase of the number of abnormal metabolic indexes, it seems that the mean temperature gradually increased in palms and dorsum of feet, and decreased in the anterior trunk.

Conclusion: The thermograph of MS exhibits certain characteristics. This may help reveal the correlations between Infrared thermography and metabolic disorders.

Keywords: metabolic syndrome, infrared thermography, temperature, evaluation method, hypertension, hyperglycemia, abdominal obesity

1 INTRODUCTION

Metabolic Syndrome (MS) is a clinical syndrome characterized by the presence of a group of metabolic disorders, including hyperglycemia (Diabetes Mellitus, DM, or Impaired Glucose Regulation, IGR), dyslipidemia (hypertriglyceridemia and/or low high density lipoprotein cholesterol), hypertension and central obesity (1). Evidence indicates that MS significantly promotes the onset and progression of type 2 diabetes mellitus (T2DM), cardiovascular and cerebrovascular diseases. Compared with patients without MS, MS patients had a higher risk and mortality of cardiovascular disease (CVD) (with a relative risk of 2.35 and 2.40, respectively) (2). In 2013-2014, the prevalence of MS adults was 31.5% in the US (3). In Mainland China, the prevalence

of MS among the population aged 15 years and older has reached 24.5% in 2016 (4). According to the Yearbook issued by China's National Health and Family Planning Commission, the total cost of hospitalization for diabetes, acute myocardial infarction and cerebral infarction in China has reached RMB 49.1 billion in 2016 (5), more than three times higher than that in 2012 (6). The increasing prevalence of MS implicates a heavy global health burden and socioeconomic cost, making it of great significance for early diagnosis and intervention of MS.

Nevertheless, in China, 2015, the awareness rate, treatment rate, and control rate of hypertension among adults were 51.6%, 45.8%, and 16.8%, respectively (7). In the 2013 national survey, undiagnosed diabetics accounted for 63% of the total (8). In 2010, the awareness rate, treatment rate, and control rate of dyslipidemia in Chinese adults were 10.93%, 6.84%, and 3.53%, respectively (9). The unsatisfying control station of MS might be the results due to asymptomatic MS patients of early-stage and complicated diagnostic procedure, which involves multiple assessments of body indexes such as blood glucose, blood lipid, blood pressure, waist circumference, etc., some of which are invasive examination and inconvenient to patients. Hence, it's meaningful to explore a method that can effectively and quickly pick out MS patients as well as the high-risk groups of MS from so-called "healthy people", assisting early detections and evaluation of the MS population.

Infrared thermography (IRT) is a technique capable of capturing infrared radiation emanating from the human body and converting it to temperature with the output of thermal maps (10). This accurate (with a thermal resolution of 0.03-0.1°C), non-contact and non-invasive technique has been used to early screening and efficacy evaluation of numerous diseases such as breast cancer, diabetic neuropathy, liver metastases, cardiovascular disease and so on (11, 12). The body surface temperature is mainly affected by local blood perfusion, but the influence of muscle activity and metabolic activity cannot be excluded (13). It might serve as an indicator of the function of corresponding parts of the body. Therefore, it's possible to detect diseases before the noticeable occurrence of structural changes.

According to recent studies, the metabolic indexes which are used in the diagnosis of MS do have correlations with skin temperature: temperature of palms of DM patients tend to be lower than that of normal people (14). While overweight females have lower mean abdominal temperature and higher hand temperature than the lean ones (15), and average skin surface temperature (°C) waveform of hypertension participants varies from normal participants (16). Besides, skin temperature of the anterior supraclavicular shows the ability to detect brown adipose tissue (BAT) activation, which is involved in the body weight, glucose and lipid regulation, reflecting the metabolic changes (17, 18). Therefore, IRT exhibits the potential of indicating metabolic disorders, and may have some advantages and application prospects in MS screening.

Based on the current findings, we conducted this study which sought to explore the temperature distribution characteristics of the face, trunk, palms and feet of MS, hope to establish the correlation between specific thermograph patterns and metabolic disorder, and provide reference to further studies and the application of IRT in early assistant diagnosis and evaluation of MS population.

2 MATERIALS AND METHODS

2.1 Participants

The data of this retrospective study were collected from patients who visited the Health Management Center of the International Department of Dongzhimen Hospital and the endocrinology department of Dongzhimen Hospital, Beijing University of Chinese Medicine from June 2016 to April 2019. After reviewing the inclusion, exclusion and rejection criteria, 184 Asian participants aged between 18 and 70 were brought into the study, including 91 males and 93 females. This study has been waived the requirement for informed consent by the institutional Ethics Committee of Dongzhimen Hospital because of its retrospective nature (No. DZMEC-KY-2020-12).

The MS diagnostic criteria used in the study was published in *Guidelines for the prevention and control of type 2 diabetes in China (2017 Edition)* (19) by Chinese Diabetes Society: 1) Abdominal obesity: waist circumference ≥ 90 cm in males and 85 cm in females; 2) Hyperglycemia: fasting plasma glucose ≥6.1 mmol/L or 2-hour plasma glucose ≥ 7.8mmol/L following a 75 g oral glucose load and (or) diabetes diagnosis has been confirmed; 3) Hypertension: blood pressure≥130/85 mmHg and (or) hypertension diagnosis has been confirmed; 4) Fasting TG≥1.70 mmol/L; 5) Fasting HDL-C<1.04 mmol/L; 3 out of 5 factors above are required.

Exclusion criteria: 1) Coronary heart disease, cerebral vascular disease, hypertensive nephropathy, diabetic retinopathy, diabetic peripheral neuropathy, diabetic peripheral vascular disease, diabetic nephropathy or other severe chronic complications caused by MS constituents (hypertension, hyperlipidemia, diabetes); 2) Menstruation, pregnancy or lactation; 3) Diseases that may affect body temperatures, such as cold and thyroid disease; 4) Medical histories of hepatic and renal insufficiency, hematopathy, tumor, severe trauma or major surgery; 5) Critical illness such as diabetic ketoacidosis, hypertensive crisis and hypertensive encephalopathy.

Rejection criteria: 1) Data with obvious error, low creditability or omissions; 2) Thermal maps taken in non-standard positions, blurring or interfered by sweat or medical ultrasonic couplant.

2.2 Groups

The participants were grouped by their gender (group M for males and group F for females) and further divided into four groups according to the number of abnormal metabolic indexes.

1) Two normal groups (M0, F0) with no abnormal metabolic index, consist of 26 participants in group M0 and 50 participants in group F0;

2) Two groups with 1 abnormal index (M1, F1), consist of 18 participants in group M1 and 21 participants in group F1;

3) Two groups with 2 abnormal indexes (M2, F2), consist of 16 participants in group M2 and 10 participants in group F2;

4) Two MS groups (M3, F3) with 3 or more abnormal indexes, consist of 31 participants in group M3 and 12 participants in group F3.

2.3 Methods

The following data of all participants were collected: demographics (gender, age), medical history, body measurement indexes (height, body mass, BMI, waist circumference, hip circumference), systolic blood pressure (SBP) and diastolic blood pressure (DBP), fasting plasma glucose (FPG), total cholesterol (TC), triglyceride (TG), high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C), serum uric acid (UA), etc.

Thermography and all those data above of each participant were obtained on the same day. Thermography was captured by an HIR-2000A (FLIR detector) medical infrared camera produced by Beijing Yuetian Optoelectronics Technology Co., Ltd, with the emissivity of 0.98, spectral range of 8-14μm, frame rate of 9 frames/second, pixels/frame of 256×324×14 Bits, thermal resolution of 0.05°C, and spatial resolution of 0.95 mrad. The detector had gone through blackbody correction and temperature calibration before leaving the factory. The ambient temperature was controlled at 22.0°C ± 2.0°C, and the relative humidity was 60% - 70%. There were no other electronic devices, no air convection nor direct illumination of strong light in the cabin. The participants were informed to avoid taking in foods that were too cold or too hot one hour before the conduction of thermography, loosen the clothing, take off hats and glasses, avoid pressing or scratching the body, and rest for 15 minutes before the examination. The participants were unclothed and kept 2 meters away from the detector. Both anterior and posterior views were captured in the anatomical position (the gesture of standing erect with palms facing forward).

The analysis of thermography was carried out by TMI-W Infrared Medical Imaging Workstation V1.0 produced by Beijing Wholelife Medical Science Co., Ltd. Mean temperature of the anterior trunk was generated automatically by the workstation. The other five regions of interest (ROIs) (face, bilateral palms and bilateral dorsum of feet) were manually delimited (**Figure 1**). Then mean temperature (T_{mean} ± SD) of each ROI were calculated (represented as T_f for the face, T_t for the anterior

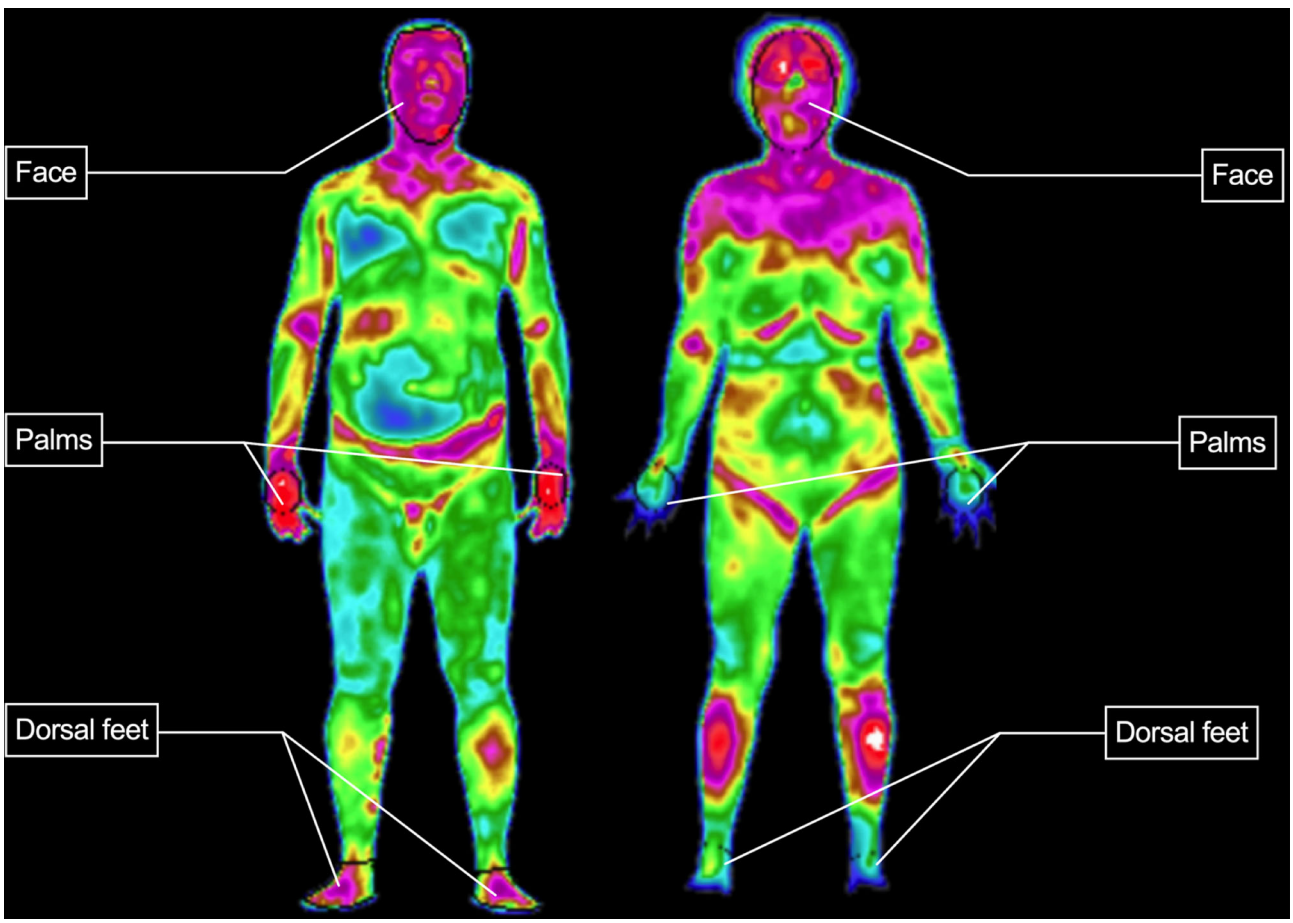


FIGURE 1 | Delimitation of ROI.

trunk, T_{rp}/T_{lp} for the right/left palm, T_{rf}/T_{rf} for the right/left dorsum of the foot).

2.4 Outcomes

The primary objectives of the analysis were to compare the mean temperature of six ROIs within each group to find out the temperature distribution characteristics of each group that conform to different numbers of indexes in MS diagnostic criteria, and to compare the mean temperature of four groups in each gender to explore the fluctuation trends of temperature along with the progress of metabolic disorders. And the secondary objective of the analysis was to probe into the correlation of the mean temperature of the ROIs with the measurement and laboratory data that relevant to metabolic disorders.

2.5 Statistical Analysis

Statistical analysis was carried out using IBM®SPSS®Statistics (version 24). Data were tested for normality using the Shapiro – Wilk test (α=0.1). Variable transformation was performed for data not conforming to normal distribution. With regard to the quantitative data with normal distribution, differences between two groups were tested using the two-sample t-test for data with homogeneity and t'-test for data with heterogeneity of variance. One-way analysis of variance (ANOVA) followed by Bonferroni's *post-hoc* test was used to compare multiple groups. For the data which did not conform to the normal distribution or homogeneity of variance, differences between two groups or multiple groups were tested using Wilcoxon rank-sum test and Kruskal-Wallis test, respectively. The measurement data were expressed as mean ± standard deviation ($\bar{x} \pm SD$). Correlation analysis was performed using the Pearson correlation test. Then perform unary linear regression or multiple stepwise regression to analyze the correlations between T_{mean} of ROI and correlated measurement and laboratory data. In regression analysis, T_{mean} of each ROI was set as the dependent variable, and independent variables were corresponding data. For all statistical analyses, a p-value lower than 0.05 was assumed significant. The complete study process is shown in **Figure 2**.

3 RESULTS

3.1 Baseline Characteristics

Demographic and clinical data are summarized in **Table 1**. The age distribution was similar in male groups while having significant differences (P<0.05) in female groups. With the increase of the number of abnormal metabolic indexes, except the age of women increased significantly, some of the body measurement and laboratory data also showed increasing trend among four subgroups in both genders (**Table 1**).

3.2 Characteristics of T_{mean} of 6 ROI in Different Gender Groups

Compared with females, males had higher mean temperatures in the areas of face, palms and dorsum of feet, and lower mean temperature in anterior trunk (P<0.05). These diversities indicated that gender difference significantly influences T_{mean} of each ROI. Therefore, it is necessary to discuss characteristics of each ROI grouping by gender (**Figure 3** and **Supplementary Table 1**).

3.2.1 Characteristics of T_{mean} of ROI in Male Groups

Within each male group, mean temperature exhibited a decreasing trend in the order of the face, anterior trunk, bilateral palms, bilateral dorsum of feet, and the differences among these temperature values were significant (P<0.01) (**Table 2** and **Figure 4**). With the increase of the number of abnormal metabolic indexes, Tt decreased successively from group M0 to M3. In T_{mean} of other ROIs, slightly increasing trends with no significant difference in the order of M0, M1 & M2, M3 can be recognized (**Table 2** and **Figure 5**). When focusing on the differences between group M0 and M3, MS males had higher mean temperature in their left palms and lower mean temperature in their anterior trunks (P<0.05) (**Supplementary Table 2**).

3.2.2 Characteristics of T_{mean} of 6 ROI in Female Groups

In female groups, characteristics of the temperature distribution of each ROI differ between groups with the different numbers of abnormal indexes. Along with the increase of abnormal metabolic indexes, the T_{mean} of bilateral palms and dorsum of feet increased, while the T_{mean} of anterior trunk decreased. In the normal group, the anterior trunk had the highest mean temperature, followed by the face, then palms and feet; in groups with 1 or 2 abnormal indexes, T_t became lower than T_f; in the MS group, T_t continuously decreased, and T_{mean} of the palms increased to a level slightly higher than T_t (**Table 2** and **Figure 6**). T_f was the only ROI that showed no significant differences among the four female groups (**Table 2** and **Figure 7**). Compared with the normal female group, female MS patients had higher mean temperature in their bilateral palms and dorsum of feet, lower mean temperature in their anterior trunks (P<0.01) (**Supplementary Table 2**).

3.3 Correlation Analysis and Linear Regression Between Mean Temperature of ROI and Measurement Data & Laboratory Data

3.3.1 Male Participants

For male participants, the results of correlation analysis showed positive correlations between T_f and DBP (r=0.256, P=0.014), T_{rp} & T_{lp} and BMI (with r-values of 0.239 and 0.234, respectively, P<0.05) and negative correlations between T_t and SBP, DBP, UA, TG, hip circumference, body mass, BMI and waist circumference (**Supplementary Table 3**). Unary linear regression was done between T_f & DBP, T_{rp} & BMI and T_{lp} & BMI (**Supplementary Table 4** and **Supplementary Figure 1**), details on the regression equations were listed in **Supplementary Data**.

3.3.2 Female Participants

Correlation analysis on T_{mean} of each ROI and measurement & laboratory data of female participants are shown in **Table 3**.

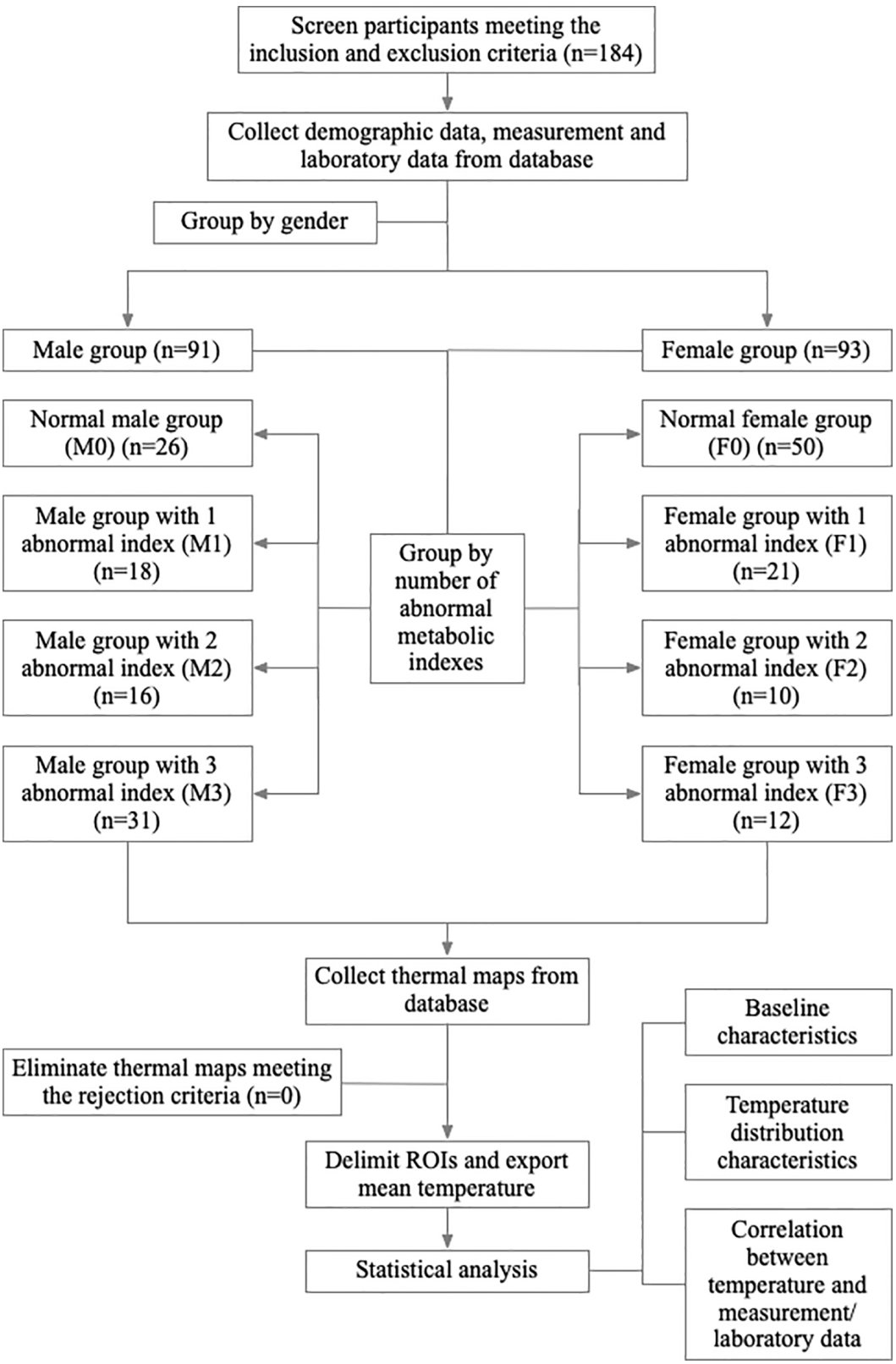


FIGURE 2 | Study flow chart.

TABLE 1 | Baselines of age, measurement and laboratory data in groups (mean, minimum-maximum).

Groups n	M0 26	M1 18	M2 16	M3 31	χ^2/F^4	*P*	F0 50	F1 21	F2 10	F3 12	χ^2/F^4	*P*
Age (years)	36.46 (23-58)	36.60 (25-63)	41.00 (24-61)	43.16 (28-66)	2.788[1]	0.045	33.50 (21-55)	41.76 (27-60)	44.10 (28-61)	55.58 (38-69)	16.027[1]	0.000
Height (cm)	172.55 (165.5-181.8)	174.13 (167.0-185.4)	173.24 (160.0-184.1)	173.64 (155.0-187.0)	0.305[2]	0.822	161.82 (150.5-177.8)	161.88 (147.7-170.3)	163.40 (155.8-171.0)	160.07 (155.2-166.4)	2.708[3]	0.439
Body mass (kg)	68.20 (60.6-79.75)	71.18 (59.5-82.1)	79.03 (64.0-99)	88.31 (65.0-110.0)	46.586[3]	0.000**	54.74 (43.6-72.6)	57.79 (43.8-74.6)	66.84 (56.1-78.8)	64.62 (52.0-82.2)	19.548[3]	0.000**
BMI (kg/m^2)	22.85 (20.84-27.15)	23.49 (21.04-25.89)	26.28 (21.94-32.18)	29.31 (23.34-38.51)	51.894[3]	0.000**	20.90 (16.52-26.63)	22.03 (15.97-25.90)	25.10 (20.52-31.39)	25.30 (20.06-33.78)	20.243[3]	0.000**
Waist circumference (cm)	81.24 (72.0-87.0)	86.22 (73.0-93.0)	93.06 (78.0-111.0)	100.20 (88.0-124.0)	58.219[3]	0.000**	72.11 (60.0-84.0)	77.50 (68.0-91.0)	82.40 (70.0-101.0)	85.75 (72.0-99.0)	30.965[3]	0.000**
Hip circumference (cm)	94.87 (87.0-103.0)	97.26 (92.0-103.0)	101.68 (92.0-112.0)	103.73 (93.3-116.0)	37.062[3]	0.000**	91.69 (81.0-104.0)	94.13 (84.0-107.0)	98.90 (91.0-110.0)	95.00 (87.0-105.0)	5.700[2]	0.001**
SBP (mmHg)	115.89 (101-129)	119.78 (107-137)	123.88 (100-151)	134.84 (99-172)	19.902[3]	0.000**	110.00 (97-127)	119.05 (94-145)	126.80 (102-151)	124.08 (107-140)	17.729[3]	0.001**
DBP (mmHg)	68.85 (61-78)	72.00 (60-87)	77.56 (59-97)	85.48 (53-112)	27.852[3]	0.000**	66.86 (53-85)	73.19 (57-96)	78.40 (65-94)	77.25 (55-110)	7.743[2]	0.000**
FPG (mmol/L)	5.09 (4.38-5.74)	5.57 (4.58-10.74)	5.73 (4.37-8.54)	7.27 (4.53-17.06)	21.156[3]	0.000**	4.96 (4.28-5.86)	5.14 (4.34-7.11)	5.56 (5.07-6.23)	7.58 (4.94-11.19)	30.419[3]	0.000**
UA (μmol/L)	352.0 (328.1-375.9)	364.7 (281.5-485.9)	402.8 (309.5-488.6)	414.7 (271.4-552.5)	4.831[2]	0.004**	253.9 (119.4-459.1)	252.3 (137.7-414.5)	289.8 (231.1-365.5)	306.1 (206.7-434.33)	10.683[3]	0.014*
TC (mmol/L)	4.95 (3.71-6.39)	4.98 (3.82-7.57)	5.06 (3.56-7.00)	5.08 (3.16-8.84)	0.719[3]	0.869	4.87 (3.22-7.62)	4.83 (3.52-6.01)	5.44 (3.15-7.02)	5.18 (4.20-7.25)	6.483[3]	0.090
TG (mmol/L)	0.91 (0.32-1.69)	1.25 (0.65-2.58)	1.65 (0.73-4.17)	2.88 (0.71-9.64)	46.253[3]	0.000**	0.78 (0.27-1.33)	1.22 (0.46-2.59)	1.40 (0.84-2.23)	2.20 (0.80-4.83)	33.767[3]	0.000**
HDL-C (mmol/L)	1.41 (1.07-2.57)	1.29 (0.88-3.14)	1.18 (0.71-1.85)	1.07 (0.63-2.18)	23.975[3]	0.000**	1.57 (1.16-2.54)	1.45 (0.95-1.90)	1.38 (0.79-2.40)	1.17 (0.84-1.52)	18.806[3]	0.000**
LDL-C (mmol/L)	2.79 (1.84-4.07)	2.91 (1.13-5.00)	3.02 (1.47-4.24)	2.82 (0.78-5.61)	0.372[2]	0.773	2.60 (1.40-4.75)	2.56 (1.81-3.87)	3.12 (1.82-4.31)	3.17 (0.79-4.84)	8.063[3]	0.045*

[1]*ANOVA performed on the age of both gender groups (with variable transformation), Post-Hoc tests (Bonferroni method) showed no significance in ages between four male groups (P > 0.05), female group F1, F2 (P = 1.000), and F2, F3 (P = 0.106).*

[2]*For data that conformed to the normal distribution and homogeneity of variance, ANOVA was implemented.*

[3]*For data that did not conform to the homogeneity of variance, Kruskal-Wallis test was used.*

[4]χ^2/F *refers to the* χ^2 *value in Kruskal-Wallis test/F value in ANOVA.*

**P < 0.05, ** P < 0.01.*

Multiple stepwise regression was carried out between T_{mean} and correlated indexes, details on the regression equations were listed in **Supplementary Data**.

4 DISCUSSION

Our study demonstrated that: With the increasing number of abnormal metabolic indexes, the mean temperature gradually increased in the face, bilateral palms and dorsum of feet, and decreased in the anterior trunk. And for the MS patients, the temperature of the anterior trunk and bilateral palms became approximated. This specific tendency may indicate the severity of metabolic disorders.

The gender difference in temperature distribution was the basis of our subsequent analysis. In this study, there were obvious gender differences that the mean temperatures of face, palms and dorsum of feet were significantly higher in males than in females, while the mean temperature of anterior trunk showed the contrary characteristic. Several studies have also reported gender differences in temperature distribution. A study on healthy participants showed that the mean temperature of the chest was significantly higher in women than in men, while in the other analyzed body surface areas, including back, abdomen, and four limbs (anterior and posterior), the mean temperatures were significantly lower in women, which may be related to less skeletal muscle and thicker subcutaneous fat in females (20). Since we did not separate the anterior trunk region into chest and abdomen, the temperature characteristics of the anterior trunk of these two studies are not comparable, while the results that males have higher temperature in palms and dorsum of feet are consistent with this study. Another study found lower mean temperatures in the trunk and four limbs in the female group, which may have a bearing on higher body fat rates in females (21). The result of the lower mean temperature of the anterior trunk in females did not conform to our study. Consequently, further researches refer to the gender difference in body surface temperature distribution with a more detailed delimitation of ROI remain to be done.

The features of the thermal maps of the MS population were the key points of our study. And we did observe specific characteristic changes in the thermal maps of the MS

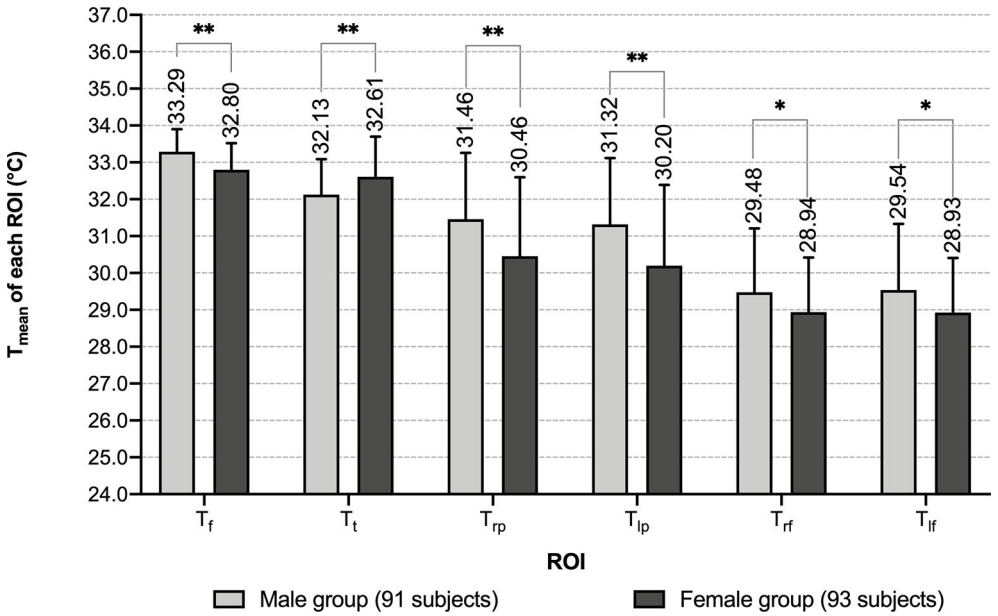


FIGURE 3 | Comparison of T_{mean} of each ROI between different genders. *: $P < 0.05$ **: $P < 0.01$.

population. There were few articles in allusion to the characteristic of infrared thermography of the MS population. Still, several studies on thermal characteristics of obesity, hyperlipidemia, hyperglycemia and hypertension have been done. Eduardo Borba Neves et al. (21) have discovered that, in males, the temperature in hands (anterior and posterior) had a tendency of increasing along with the body fat rate (BF%). Besides, BF% had a negative correlation with temperature in the anterior trunk, which was basically consistent with the results of our study. Eun-Mo Song et al. (22) found that the visceral fat area (VFA) was negatively correlated with the temperature of the acupoint CV4, which is located in the lower abdomen. This also corroborated the correlation between obesity and the decrease of temperature in the abdomen. Thiruvengadam J et al. (12) demonstrated that HDL was negatively correlated with the surface temperature of the left hand and anterior feet, which coincide with the conclusion of this study that severer metabolic disorder leads to higher mean temperature in palms and dorsum of feet. Aleck Ovechkin et al. (23) found the body surface temperature of the "Yin-Tang" acupuncture point which is located in the forehead negatively correlates with the severity of intracranial hypertension syndrome. This observation suggests varied temperature distribution characteristics of the face in hypertension patients compared with normal ones. While in this study, no change in the mean temperature of the face was found in MS participants, suggesting changes in body surface temperature of certain diseases might take place in some particular small areas, which could be covered when chosen

TABLE 2 | Comparison of T_{mean} of each ROI among four groups in each gender (C; $\bar{x} \pm s$).

Groups	M0	M1	M2	M3	χ^2/F^4	*P*	F0	F1	F2	F3	χ^2/F^4	*P*
n	26	18	16	31			50	21	10	12		
T_f	33.14 ± 0.72	33.41 ± 0.69	33.20 ± 0.46	33.40 ± 0.53	1.157[2]	0.331	32.76 ± 0.84	32.88 ± 0.71	32.73 ± 0.47	32.93 ± 0.36	1.851[3]	0.604
T_t	33.12 ± 0.50	32.76 ± 0.63	32.31 ± 0.50	32.00 ± 0.73	17.405[2]	0.000**	33.14 ± 0.67	32.74 ± 0.69	32.56 ± 0.88	32.33 ± 0.79	13.426[3]	0.004**
T_{rp}	30.95 ± 2.16	31.48 ± 1.60	31.07 ± 1.78	32.06 ± 1.46	5.155[3]	0.161	29.88 ± 1.98	30.59 ± 1.93	30.64 ± 2.60	32.49 ± 1.56	14.628[3]	0.002**
T_{lp}	30.81 ± 2.18	31.34 ± 1.67	31.03 ± 1.64	31.87 ± 1.51	5.430[3]	0.143	29.61 ± 1.94	30.19 ± 2.04	30.37 ± 2.71	32.54 ± 1.53	17.660[3]	0.001**
T_{rf}	29.26 ± 1.95	29.41 ± 1.76	29.42 ± 1.49	29.74 ± 1.70	2.330[3]	0.507	28.43 ± 1.32	29.02 ± 0.87	28.95 ± 1.76	30.89 ± 1.14	23.742[3]	0.000**
T_{lf}	29.33 ± 2.07	29.41 ± 1.82	29.59 ± 1.51	29.77 ± 1.71	2.128[3]	0.546	28.39 ± 1.28	29.18 ± 0.95	28.84 ± 1.87	30.82 ± 1.10	23.378[3]	0.000**
χ^2	82.062[1]	60.080[1]	56.496[1]	97.698[1]			200.326[1]	67.455[1]	23.618[1]	29.322[1]		
P^1	0.000**	0.000**	0.000**	0.000**			0.000**	0.000**	0.000**	0.000**		

[1]*Kruskal-Wallis test was used for comparing T_{mean} of six ROI within each gender group.*

[2]*ANOVA followed by Bonferroni's post-hoc test was implemented for comparing T_{mean} of each ROI between four male groups. In post-hoc test, significance was found between T_t of group M0/M2, M0/M3, M1/M3 (adjusted P<0.05).*

[3]*When comparing T_{mean} of each ROI between four groups in each gender, for data that did not conform to the normal distribution, Kruskal-Wallis test was used. Bonferroni correction showed significance between T_t and T_{rp} of group F0/F3, T_{lp} of group F0/F3, F1/F3, T_{rf} and T_{lf} of group F0/F3, F1/F3, F2/F3 (adjusted P < 0.05).*

[4]*χ^2/F refers to the χ^2 value in Kruskal-Wallis test/F value in ANOVA. ** P < 0.01.*

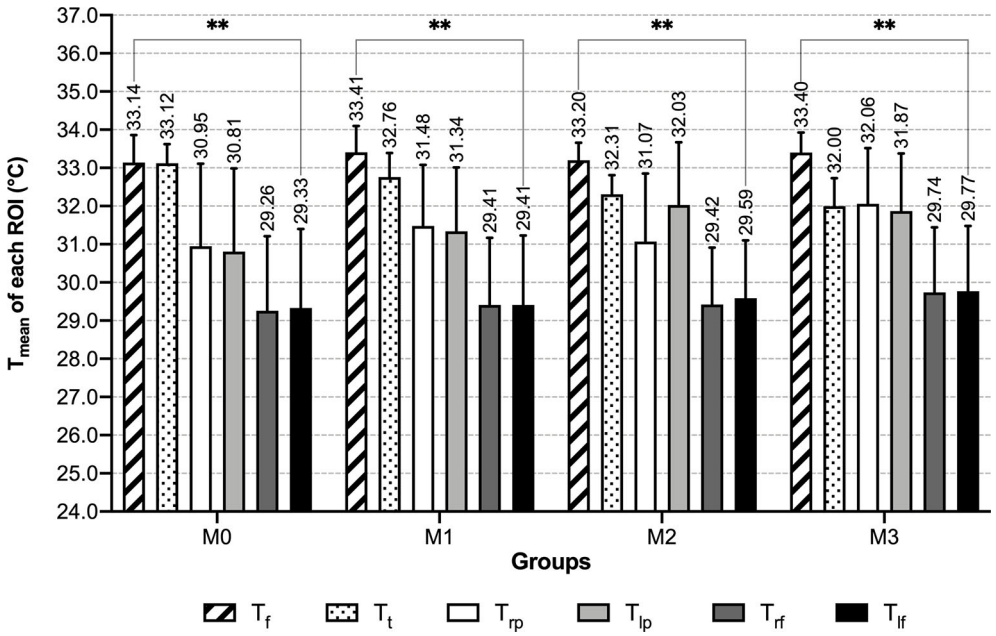


FIGURE 4 | T_{mean} of each ROI of male groups - grouped by number of abnormal indexes. **: P < 0.01.

ROIs are quite bigger. In summary, the characteristics of higher mean temperatures in palms and dorsum of feet, and lower mean temperature in the anterior trunk might be results of multiple pathological mechanisms as above. Studies on one particular metabolic disorder like hyperlipidemia, hypertension, and hyperglycemia may help in understanding the underlying mechanism of characteristic changes. However, clinical trials on large scales are still in lack, which conduces to a more accurate interpretation of the thermography characteristic of MS.

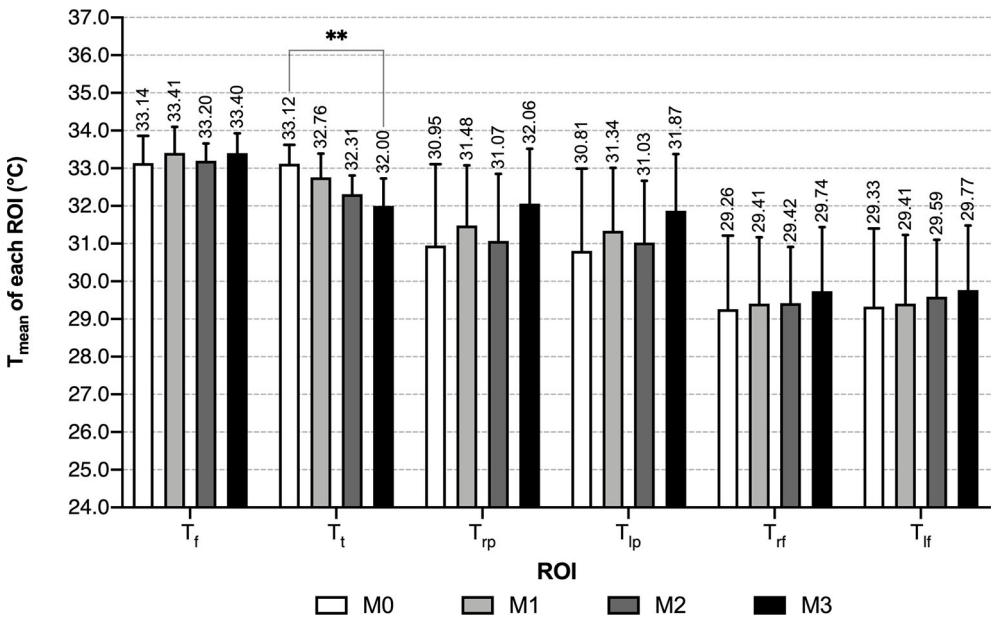


FIGURE 5 | T_{mean} of each ROI of male groups - grouped by ROI. **: *P* < 0.01.

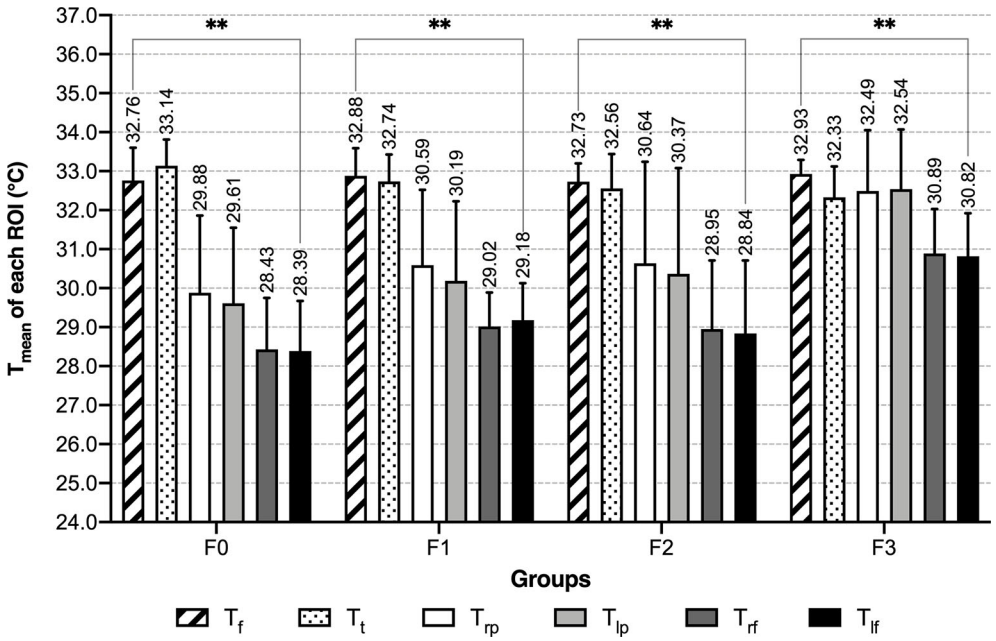


FIGURE 6 | T_{mean} of each ROI of female groups - grouped by number of abnormal indexes. **: $P < 0.01$.

Through correlation analysis and linear regression, in both male and female groups, we found a positive correlation between the mean temperature of the palms and BMI, and negative correlations between mean temperature of the trunk and DBP, UA, TG, waist circumference, hip circumference, weight and BMI. These patterns can be consistent with the above-mentioned law of increasing palm temperature and decreasing trunk temperature with the aggravation of metabolic abnormalities. The unique results in male groups are: a positive correlation between T_f and DBP, and a negative correlation between T_t and SBP. In females,

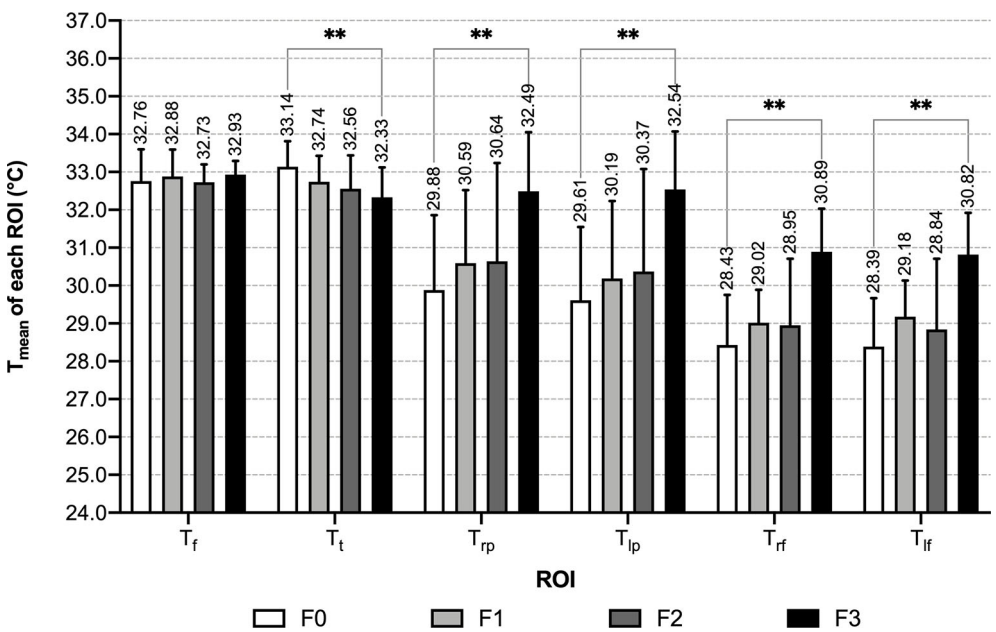


FIGURE 7 | T_{mean} of each ROI of female groups - grouped by ROI. **: $P < 0.01$.

TABLE 3 | Pearson correlation analysis between T_{mean} of each ROI and measurement data & laboratory data in female groups.

	Pearson Correlation Coefficient (r)					
	T_f	T_t	T_{rp}	T_{lp}	T_{rf}	T_{lf}
Age (years)	-0.157	**-0.392****	**0.410****	**0.421****	**0.526****	**0.481****
Height (cm)	**0.229***	**0.344****	0.014	-0.007	-0.013	0.018
Body mass (kg)	**0.207***	**-0.378****	**0.340****	**0.369****	**0.319****	**0.315****
BMI (kg/m2)	0.127	**-0.514****	**0.343****	**0.381****	**0.334****	**0.317****
Waist circumference (cm)	0.069	**-0.494****	**0.308****	**0.342****	**0.297****	**0.298****
Hip circumference (cm)	0.185	**-0.346****	**0.239***	**0.253***	0.190	0.196
SBP (mmHg)	-0.001	-0.177	0.023	0.025	0.116	0.112
DBP (mmHg)	-0.071	**-0.231***	0.043	0.049	0.081	0.071
FPG (mmol/L)	0.028	**-0.219***	**0.261***	**0.274****	**0.388****	**0.384****
UA (μmol/L)	0.107	**-0.224***	0.201	**0.223***	**0.236***	0.188
TC (mmol/L)	-0.124	-0.157	0.044	0.024	0.036	0.026
TG (mmol/L)	0.104	**-0.374****	**0.365****	**0.377****	**0.342****	**0.356****
HDL-C (mmol/L)	-0.161	**0.274****	**-0.372****	**-0.376****	**-0.425****	**-0.371****
LDL-C (mmol/L)	-0.101	**-0.218***	0.145	0.132	0.187	0.141

Bold r-values mean that the correlations between the corresponding T_{mean} and measurement data and laboratory data are statistically significant.
**P < 0.05, ** P < 0.01.*

the unique results are: T_f was positively correlated with height and weight, the mean temperature of palms and dorsum of feet was positively correlated with hip circumference, UA, weight, BMI, waist circumference, FPG, TG and age, negatively correlated with HDL-C, and the average temperature of the trunk was negatively correlated with FPG, LDL-C, age and height. In general, the correlation between T_t and multiple metabolic indexes is obvious regardless of gender. While the mean temperature of palms and dorsum of feet showed correlations with a sizable amount of indexes only in the female group, and correlated with age at the same time. Given the uneven age baseline of female subjects, the relationship between palms and feet temperature and metabolic abnormalities still needs to be verified based on more homogeneous sample studies.

5 LIMITATIONS

It is also of note that heterogeneity was found among the age of four female groups. The average age of the female MS group was 55.58 ± 10.431 years old, mostly in perimenopausal and postmenopausal stages (start at 47.5 years old on average (24)). The particular age of this group may be due to the prevalence increase of MS in perimenopausal and postmenopausal women. A meta-analysis carried out by Hallajzadeh et al. (25) found the prevalence of MS among postmenopausal women was 37.17% and the pooled OR for MS in postmenopausal women in comparison with the premenopausal was 3.54. In perimenopausal and postmenopausal women, decreased estradiol level attenuates the protective effect of estradiol, which may account for the inclination of carbohydrate and lipid metabolism disorder and contribute to the development of MS (26, 27). However, due to the small sample size of this group, there remains the possibility that the sampling error led to the elder age of women in this group. Accordingly, further study with an enlarged sample size is necessary.

Owing to the retrospective nature of this study, the results could be influenced by time, temperature, humidity and state of equipment, etc. Due to the lack of research on MS using IRT, the sample size was not calculated in advance. While we calculated the sample size afterward based on the results of this study. Except for the comparison of T_{lp} between M0 and M3, T_t between F0 and F3, T_{rf} and T_{lf} between F2 and F3, the statistical powers of the other comparisons were higher than 0.80. Moreover, the sample size and age of each group were not evenly distributed, which may also have some bearing on the outcome. A methodological problem is that we did not capture the thermograph of the dorsum of feet in a vertical angle, for the retrospective nature, also. This could affect the output temperatures with camera (28), while since all of the thermographs were captured in approximate angles, the tendency of temperature changes we observed is still advisable.

6 CONCLUSION

This work was a pilot study of temperature characteristics in the MS population, and it proved the feasibility of screening and evaluating metabolic disorders through IRT. The key findings of this work were the gender difference in temperature distribution, the sequences of the mean temperature of the face, anterior trunk, palms and dorsum of feet in subjects of different metabolic conditions, the trends of the temperature changes in the above body parts with increasing number of abnormal metabolic indexes, and the certain differences in mean temperature of these body parts between the normal group and MS group. And the secondary findings are the correlations between measurement or laboratory data related to metabolic disorders and the mean temperature of different body parts. With the popularization of IRT and the deepening of the research, this correlation may help screen patients with metabolic disorders, further postponing the development of diseases.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by Ethics Committee of Dongzhimen Hospital. The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

All authors developed the concept for the study. Y-hX and W-zZ offered the design of this manuscript. RC provided suggestions. M-jG and Z-jC conducted the experiment. H-zX drafted the manuscript. B-yJ did literature search. B-hM provided technical support on the software in this study and guided the revision process of this manuscript. Y-cS reviewed the manuscript and made corrections. All authors contributed to the article and approved the submitted version.

FUNDING

This work was supported by State Administration of Traditional Chinese Medicine JB063 programme and the Fundamental Research Funds for the Central Universities 2020-JYB-ZDGG-117 programme.

ACKNOWLEDGMENTS

We would like to thank Prof. Wen-xue Hong from College of Electrical Engineering, Yanshan University, for his support in medical infrared imaging technology during this study.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2022.851369/full#supplementary-material

REFERENCES

1. Kassi E, Pervanidou P, Kaltsas G, Chrousos G. Metabolic Syndrome: Definitions and Controversies. *BMC Med* (2011) 9:48. doi: 10.1186/1741-7015-9-48
2. Mottillo S, Filion KB, Genest J, Joseph L, Pilote L, Poirier P, et al. The Metabolic Syndrome and Cardiovascular Risk a Systematic Review and Meta-Analysis. *J Am Coll Cardiol* (2010) 56(14):1113–32. doi: 10.1016/j.jacc.2010.05.034
3. Marcotte-Chénard A, Deshayes TA, Ghachem A, Brochu M. Prevalence of the Metabolic Syndrome Between 1999 and 2014 in the United States Adult Population and the Impact of the 2007-2008 Recession: An NHANES Study. *Appl Physiol Nutr Metab = Physiol Appliquee Nutr Metabolisme* (2019) 44 (8):861–8. doi: 10.1139/apnm-2018-0648
4. Li R, Li W, Lun Z, Zhang H, Sun Z, Kanu JS, et al. Prevalence of Metabolic Syndrome in Mainland China: A Meta-Analysis of Published Studies. *BMC Public Health* (2016) 16:296. doi: 10.1186/s12889-016-2870-y
5. National Health Commission of the People's Republic of China. *Statistical Yearbook of Health and Family Planning of China 2017 Edition*. Beijing: Peking Union Medical College Press (2017).
6. National Health Commission of the People's Republic of China. *Statistical Yearbook of Health and Family Planning of China 2013 Edition*. Beijing: Peking Union Medical College Press (2013).
7. 2018 Chinese Guidelines for the Management of Hypertension. *Chin J Cardiovasc Med* (2019) 2401(1):002. doi: 10.3969/j.issn.1007-5410
8. Chinese Diabetes Society. Guidelines for Prevention and Treatment of Type 2 Diabetes in China (2017 Edition). *Chin J Diabetes Mellitus* (2018) 10(1):4–67. doi: 10.3760/cma.j.issn.1674-5809.2018.01.002
9. Li JH, Wang LM, Mi SQ, Zhang M, Wang LH, et al. Awareness Rate, Treatment Rate and Control Rate of Dyslipidemia in Chinese Adults, 2010. *Chin J Prev Med* (2012) 46(8):687–91. doi: 10.3760/cma.j.issn.0253-9624.2012.08.004
10. Tattersall GJ. Infrared Thermography: A Non-Invasive Window Into Thermal Physiology. *Comp Biochem Physiol A Mol Integr Physiol* (2016) 202:78–98. doi: 10.1016/j.cbpa.2016.02.022
11. Lahiri BB, Bagavathiappan S, Jayakumar T, Philip J. Medical Applications of Infrared Thermography: A Review. *Infrared Phys Technol* (2012) 55(4):221–35. doi: 10.1016/j.infrared.2012.03.007
12. Thiruvengadam J, Anburajan M, Menaka M, Venkatraman B. Potential of Thermal Imaging as a Tool for Prediction of Cardiovascular Disease. *J Med Phys* (2014) 39(2):98–105. doi: 10.4103/0971-6203.131283
13. Hinds T, McEwan I, Perkes J, Dawson E, Ball D, George K. Effects of Massage on Limb and Skin Blood Flow After Quadriceps Exercise. *Med Sci Sports Exercise* (2004) 36(8):1308–13. doi: 10.1249/01.MSS.0000135789.47716.DB
14. Sivanandam S, Anburajan M, Venkatraman B, Menaka M, Sharath D. Medical Thermography: A Diagnostic Approach for Type 2 Diabetes Based on Non-Contact Infrared Thermal Imaging. *Endocrine* (2012) 42(2):343–51. doi: 10.1007/s12020-012-9645-8
15. Jalil B, Hartwig V, Moroni D, Salvetti O, Benassi A, Jalil Z, et al. A Pilot Study of Infrared Thermography Based Assessment of Local Skin Temperature Response in Overweight and Lean Women During Oral Glucose Tolerance Test. *J Clin Med* (2019) 8(2):260. doi: 10.3390/jcm8020260
16. Thiruvengadam J, Mariamichael A. A Preliminary Study for the Assessment of Hypertension Using Static and Dynamic IR Thermograms. *Biomed Technik Biomed Eng* (2018) 63(2):197–206. doi: 10.1515/bmt-2016-0237
17. Law J, Morris DE, Izzi-Engbeaya C, Salem V, Coello C, Robinson L, et al. Thermal Imaging Is a Noninvasive Alternative to PET/CT for Measurement of Brown Adipose Tissue Activity in Humans. *J Nucl Med* (2018) 59(3):516–22. doi: 10.2967/jnumed.117.190546
18. Brasil S, Renck AC, de Meneck F, Brioschi ML, Costa EF, Teixeira MJ. A Systematic Review on the Role of Infrared Thermography in the Brown Adipose Tissue Assessment. *Rev Endocr Metab Disord* (2020) 21(1):37–44. doi: 10.1007/s11154-020-09539-8
19. Chinese Diabetes Society. Chinese Guideline for the Prevention and Treatment of Type 2 Diabetes Mellitus (2017 Edition). *Chin J Diabetes Mellitus* (2018) 10(1):4–67. doi: 10.3760/cma.j.issn.1674-5809.2018.01.003
20. Chudecka M, Lubkowska A. Thermal Maps of Young Women and Men. *Infrared Phys Technol* (2015) 69:81–7. doi: 10.1016/j.infrared.2015.01.012
21. Neves EB, Salamunes ACC, de Oliveira RM, Stadnik AMW. Effect of Body Fat and Gender on Body Temperature Distribution. *J Therm Biol* (2017) 70(Pt B):1–8. doi: 10.1016/j.jtherbio.2017.10.017
22. Song EM, Kim EJ, Kim KW, Cho JH, Song MY. Correlation Between Abdominal Fat Distribution and Abdominal Temperature in Korean Premenopausal Obese Women. *J Korean Med* (2013) 34(2):1–9. doi: 10.13048/jkm.13001
23. Ovechkin A, Kim KS, Lee JW, Lee SM. Thermo-Visual Evaluation of the Yin-Tang Acupuncture Point for Intracranial Hypertension Syndrome. *Am J Chin Med* (2003) 31(3):455–66. doi: 10.1142/S0192415X03001041
24. McNamara M, Batur P, DeSapri KT. In the Clinic. Perimenopause. *Ann Internal Med* (2015) 162(3):ITC1–15. doi: 10.7326/AITC201502030
25. Hallajzadeh J, Khoramdad M, Izadi N, Karamzad N, Almasi-Hashiani A, Ayubi E, et al. Metabolic Syndrome and its Components in Premenopausal and Postmenopausal Women: A Comprehensive Systematic Review and Meta-Analysis on Observational Studies. *Menopause* (2018) 25(10):1155–64. doi: 10.1097/GME.0000000000001136
26. Iorga A, Cunningham CM, Moazeni S, Ruffenach G, Umar S, Eghbali M. The Protective Role of Estrogen and Estrogen Receptors in Cardiovascular Disease

and the Controversial Use of Estrogen Therapy. *Biol Sex Differences* (2017) 8 (1):33. doi: 10.1186/s13293-017-0152-8
27. Barros RPA, Gustafsson JÅ. Estrogen Receptors and the Metabolic Network. *Cell Metab* (2011) 14(3):289–99. doi: 10.1016/j.cmet.2011.08.005
28. Danko M, Hudak R, Foffová P, Zivcak J. An Importance of Camera - Subject Distance and Angle in Musculoskeletal Application of Medical Thermography. *Acta Electrotech Inform* (2010) 10(2):57–60.

Conflict of Interest: Author RC is employed by Beijing Wholelife Medical Science Co., Ltd.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers | Frontiers in Endocrinology

ORIGINAL RESEARCH
published: 14 April 2022
doi: 10.3389/fendo.2022.850784

Maternal Urinary Cotinine Concentrations During Pregnancy Predict Infant BMI Trajectory After Birth: Analysis of 89617 Mother-Infant Pairs in the Japan Environment and Children's Study

Hiroyuki Hirai[1,2], Shiki Okamoto[3], Hiroaki Masuzaki[3], Tsuyoshi Murata[4,5], Yuka Ogata[4], Akiko Sato[4], Sayaka Horiuchi[6], Ryoji Shinohara[6], Kosei Shinoki[7], Hidekazu Nishigori[4,8], Keiya Fujimori[4,5], Mitsuaki Hosoya[4,9], Seiji Yasumura[4,10], Koichi Hashimoto[4,9], Zentaro Yamagata[11], Michio Shimabukuro[1] and the JECS Group[†]*

[1] Department of Diabetes, Endocrinology and Metabolism, Fukushima Medical University School of Medicine, Fukushima, Japan, [2] Department of Internal Medicine, Shirakawa Kosei General Hospital, Fukushima, Japan, [3] Division of Endocrinology, Diabetes and Metabolism, Hematology, Rheumatology (Second Department of Internal Medicine), Graduate School of Medicine, University of the Ryukyus, Okinawa, Japan, [4] Fukushima Regional Center for the Japan Environmental and Children's Study, Fukushima, Japan, [5] Department of Obstetrics and Gynecology, School of Medicine, Fukushima Medical University, Fukushima, Japan, [6] Center for Birth Cohort Studies, School of Medicine, University of Yamanashi, Yamanashi, Japan, [7] Koriyama Office, Fukushima Regional Center for the Japan Environmental and Children's Study, Fukushima, Japan, [8] Fukushima Medical Center for Children and Women, Fukushima Medical University, Fukushima, Japan, [9] Department of Pediatrics, School of Medicine, Fukushima Medical University, Fukushima, Japan, [10] Department of Public Health, School of Medicine, Fukushima Medical University, Fukushima, Japan, [11] Department of Health Sciences, School of Medicine, University of Yamanashi, Yamanashi, Japan

Background: Clinical or epidemiological conclusions remain undecided on the direct effects of active and second-hand smoking during pregnancy on childhood obesity. Urinary cotinine (UC) concentration, an accurate and quantitative marker for smoking, may elucidate the dose-dependent relationship between smoking during pregnancy and childhood obesity. To analyze the relationship between UC concentration and smoking questionnaire (SQ) classes for active and second-hand smoking in pregnant mothers and trajectory of infant Kaup index (body mass index: BMI).

Methods: This multicenter prospective cohort study was conducted using a list-wise complete set of 35829 among 89617 mother-infant singleton pairs, recruited between 2011 and 2014, in the Japan Environment and Children's Study (JECS). Pairs were categorized according to UC levels (1 to 4 classes) or SQ (0 to 4 classes).

Results: Maternal BMI at delivery was the highest in UC class 4 (highest). Maternal and paternal education of ≥16 years and annual household income were lowest in UC class 4. Infant BMI was lower at birth, but trends in BMI and ΔBMI were higher from six to 36 months step-wise in the UC classes. The above tendency was observed in the list-wise complete dataset but was emphasized after multiple imputations and corrections of

OPEN ACCESS

Edited by:
Mostafa Qorbani, Alborz University of Medical Sciences, Iran

Reviewed by:
Dimitrios T. Papadimitriou, National and Kapodistrian University of Athens, Greece
Wenjing Hu, Chongqing Medical University, China

***Correspondence:**
Michio Shimabukuro
mshimabukuro-ur@umin.ac.jp

[†]Members listed in Appendix

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *08 January 2022*
Accepted: *28 February 2022*
Published: *14 April 2022*

Citation:
Hirai H, Okamoto S, Masuzaki H, Murata T, Ogata Y, Sato A, Horiuchi S, Shinohara R, Shinoki K, Nishigori H, Fujimori K, Hosoya M, Yasumura S, Hashimoto K, Yamagata Z, Shimabukuro M and the JECS Group (2022) Maternal Urinary Cotinine Concentrations During Pregnancy Predict Infant BMI Trajectory After Birth: Analysis of 89617 Mother-Infant Pairs in the Japan Environment and Children's Study. Front. Endocrinol. 13:850784. doi: 10.3389/fendo.2022.850784

cofounders. UC concentration in five SQ classes largely fluctuated, and the relationship between SQ classes and trends in BMI and ΔBMI was not statistically significant.

Conclusion: Infants from high UC mothers had a low BMI at birth, increasing from six to 36 months of age. UC concentrations, but not smoking questionnaire classes, predict infant BMI trajectory, suggesting that active and second-hand smoking affect child obesity in a dose-dependent manner.

Keywords: body mass index - BMI, infant obesity, cotinine concentrations, maternal smoking during pregnancy, second-hand smoking (SHS)

INTRODUCTION

Childhood obesity is often carried over into adulthood (1), leading to an increased risk of atherosclerotic cardiovascular disease (ASCVD) in early life (2). Over 60% of prepubertal childhood obesity results in adulthood obesity and lifestyle-related diseases, which account for 86% of premature death (3). Childhood obesity can be grouped into infancy (0–1.9 years), preschool age (2·0–4·9 years), school-age (5·0–12·9 years), and adolescence (13·0–18·0 years), and either case causes adulthood obesity (4). Factors associated with early childhood (newborn to preschool age) obesity such as birth weight, artificial milk, maternal smoking, and low socioeconomic status have been reported (4). Active or second-hand smoking during pregnancy (5) is a well-known factor in increasing infant Kaup index (body mass index: BMI) trajectory after birth (6–8). Riedel et al. indicated that maternal and paternal smoking during pregnancy increased the odds ratio for childhood obesity (8).

Childhood obesity due to such active or second-hand smoking is associated with lower birth weight and a compensatory increase in BMI (catch-up-growth) (9–11); the direct effects of tobacco substances on postnatal feeding behavior have also been suggested (12, 13). However, clinical or epidemiological conclusions remain undecided on the direct impact of smoking during pregnancy on childhood obesity. The reasons for the undecided conclusions: first, low socioeconomic status followed by low birth weight and less breast milk in smoking mothers can be confounded with childhood obesity; second, there are no standardized methods for quantifying smoking doses, and the difficulty in estimating concentrations of smoking substances by questionnaires limits the interpretations of tobacco substances. Measuring the urinary concentration of cotinine, a nicotine metabolite, is useful for estimating accurate tobacco intake (6, 7, 14). Therefore, the relationship between tobacco substances and BMI trajectory in childhood can be interpreted more precisely by minimizing the above confounding factors.

This study aims to analyze the relationship between urinary cotinine (UC) concentration and smoking questionnaire (SQ) classes for active and second-hand smoking in pregnant mothers and infant BMI trajectory using a large Japanese mother-infant longitudinal dataset.

Abbreviations: ANCOVA, analysis of covariance; ANOVA, analysis of variance; ASCVD, atherosclerotic cardiovascular disease; BMI, body mass index; IQR, interquartile range; JECS, Japan Environment and Children's Study; ROC, receiver operating characteristic curve; SD, standard deviation; SQ, smoking questionnaire; UC, urinary cotinine.

MATERIAL AND METHODS

Study Design and Participants

The protocol for the Japan Environment and Children's Study (JECS), an ongoing Japanese nationwide birth cohort study, has been published elsewhere (15). Briefly, 15 nationwide, regional centers were responsible for recruiting pregnant women who lived in study areas (city, town, or village) between 2011 and 2014. The current report is based on the dataset of jecs-ta-20190930, which included 104,062 datasets of fetal records up to three years of age (**Dataset 1**, **Supplement 1**). Pairs of abortion, stillbirth, other causes of unbirth, multiple births, and missing data for infant BMI and maternal UC or SQ classes were excluded, and 89617 pairs were eligible for (**Dataset 2**, **Supplement 1**). Further, missing for infant BMI at zero, six, 12, 18, 24, 30, and 36 months, mother's age at delivery (years), body weight gain during pregnancy, pre-pregnancy BMI, regular alcohol drinking, maternal and paternal education, and household income were excluded, and 35829 pairs were eligible for (**Dataset 3**, **Supplement 1**).

The JECS protocol was reviewed and approved by the Ministry of the Environment's Institutional Review Board on Epidemiological Studies and the Ethics Committees of all participating institutions. The JECS was conducted according to the principles of Helsinki Declaration and other nationally valid regulations and guidelines. Written informed consent was obtained from all participants.

Measurement of Infant BMI

The primary outcomes of this study were infant BMI trajectories and differences in the BMI (ΔBMI) at six, 12, 18, 24, 30, and 36 months from baseline according to the class of UC levels of pregnant mothers. BMI was calculated as weight (kg)/length or height $(m)^2$. Weight and length or height were collected by records of the infant's caregivers.

Questionnaire and Measurement of Covariates

Mothers completed a baseline questionnaire during the first trimester of pregnancy. The questionnaire included maternal information regarding age, height, weight before pregnancy,

parity, drinking and smoking during pregnancy, gestational age, and medical history of mothers during pregnancy such as hypertension, gestational diabetes, infant sex, single or multiple births, live birth, stillbirth, or abortion. SQ, annual household income, and parental educational background were interviewed in the second/third trimester of pregnancy.

Measurement and Classification of UC Concentrations

A maternal urine sample was collected at the second or third trimester of pregnancy, transferred to a contract laboratory at 1–10°C, and stored at −80°C until analysis. UC concentrations were determined using a high-performance liquid chromatography-tandem mass spectrometer (16). In brief, 100 μL aliquots of urine were used to measure the concentrations. The 1.4% ammonia solution (400 μL) and internal standard solution including 3 ng/mL of 13C3-cotinine (10 μL) were added in and mixed with the urine subject. The mixtures were loaded into 96-well preconditioned plates. The cartridge was centrifuged (1000 times/minute, 4°C) and cleaned with ammonia. After repeated centrifugations, elution was performed in 50% (v/v) methanol. Finally, the elute was dissolved in water of 300 μL, and a 10 μL aliquot was injected into the HPLC system. JECS Native Mixture solution 500 ng/mL in water (ES-5536) https://shop.isotope.com/productdetails.aspx?itemno=ES-5536 and JECS Labeled Mixture solution in water (ES-5535) https://shop.isotope.com/productdetails.aspx?itemno=ES-5535 were purchased from CIL (Cambridge isotope laboratories, Inc. Cambridge, England). The minimum reporting level was 0.03 ng/mL. Reproducibility and intermediate precision for cotinine analysis were 4.0% and 4.7%, respectively (16). UC concentrations normalized relative to creatinine concentrations were $\log_{10}$-transformed (16). Then, participants were categorized into four classes according to the $\log_{10}$ UC levels [log (ng/mL)]: UC class 1 (UC1), <−1; UC class 2 (UC2), <−1 to 0; UC class 3 (UC3), <0–1; and UC class 4 (UC4).

Questionnaire on Smoking Status

Active smoking status and exposure to second-hand smoke were evaluated using self-administered questionnaires during the second or third trimester when samples of UC levels were collected. A mother was asked to choose an active smoking status from the following questionnaire: 1 = never, 2 = previously did, but quit before realizing current pregnancy, 3 = previously did, but quit after realizing current pregnancy, and 4 = currently smoking. For second-hand smoking, mothers answered how often they inhaled tobacco smoke at home, workplace, or any other indoor places before and during pregnancy and from whom, including husbands, cohabitants, and colleagues at workplaces. To quantitatively assess exposure to active and second-hand smoking (6) simultaneously, we made a classification by combining two above questionnaires: SQ class 0 (SQ0), no second-hand smoking; SQ class 1 (SQ1), second-hand smoking <7 hours/week; SQ class 2 (SQ2), second-hand smoking <7–14 hours/week; SQ class 3 (SQ3), second-hand smoking >14 hours/week, and SQ class 4 (SQ4), active smoking.

Statistical Analysis

Parametric variables were presented as mean ± standard deviation (SD), and non-parametric variables were presented as median (interquartile range [IQR]). For multi-group comparison, parametric variables were analyzed using the one-way or two-way analysis of variance (ANOVA), and non-parametric variables were analyzed using the Kruskal-Wallis test. The frequencies of categorical variables were reported as percentages, and the Pearson χ^2 test was used for multi-group comparison. Trends in the BMI and the absolute increase in BMI (ΔBMI) from the baseline were calculated for overall, four UC classes, and five SQ classes and were evaluated using repeated measures ANOVA. The null hypothesis for equal distribution among four or five classes was assessed using the Mauchly's sphericity test. If the sphericity was not satisfied, the P values in repeated measures ANOVA were adjusted using the Greenhouse–Geisser ϵ correction. BMI was evaluated as the main effect, and BMI × UC classification was assessed as the interaction. After comparing the four and five classes in the complete set (n = 35829), BMI and ΔBMI were analyzed using repeated measures analysis of covariance (ANCOVA) corrected for the covariates: mother's age at delivery (years), pre-pregnancy BMI (kg/m^2), regular alcohol drinking (yes or no), hypertension (yes or no), diabetes mellitus (yes or no), infant sex, pregnancy period, maternal education of ≥16 years (yes or no), paternal education of ≥16 years (yes or no), household income in Japanese Yen (class 1, <4 million; class 2, ≥4 million and <8 million; class 3, ≥8 million and <12 million; class 4, ≥12 million). *Post-hoc* group comparisons were made after the Bonferroni correction. For the sensitivity analysis, the missing values and multiple imputations were evaluated. The dropout rates in BMI at 36 months after birth were calculated. The visualization of missing and imputed values was performed using the R statistical software (R-4.0.2, The R Foundation Vienna, Austria) with VIM package 6.0.0 and ggplot2 package 3.3.2. Multiple imputations for the missing data were performed using the Bayesian method with the minimum and maximum values set for each variable. To create and analyze 89617 records, the missing confounders were imputed for BMI at six, nine,12,18, 24, and 36 months, maternal age, BMI before pregnancy, body weight gain during pregnancy, alcohol drinking during pregnancy, maternal and paternal education of ≥16 years, and an annual class of household income. The receiver operating characteristic curve (ROC) analysis of UC levels was done from the area under the curve (AUC) for active and second-hand smoking.

Unless otherwise indicated, statistical analyses were performed using SPSS version 25.0 (SPSS, Chicago, Illinois, USA). A value of $P < 0.05$ was considered statistically significant using a two-sided test.

RESULTS

Distribution of Maternal UC Levels

The median UC concentration in the list-wise dataset (**Dataset 3**, **Supplement 1**, n = 35829) was 0.12 ng/mL (IQR, 0.05–0.39 ng/mL), minimum was 0.03 ng/mL, maximum was 6,030 ng/mL, and approximately 86% was distributed below 1 ng/mL. Since there was

no normal distribution in UC concentration, data were converted to log_{10}. In **Dataset 3**, **Supplement 1**, UC levels in log_{10} had a median value of −0.92 (IQR, −1.30 to −0.41] and a bimodal distribution (**Figure 1**).

Characteristics of Participants in the UC Classes

The general characteristics of 35829 records in the classes of UC levels are shown in **Table 1**. Participants were categorized into four classes according to log_{10} UC levels [log (ng/mL)]: UC1, <−1; UC2, <−1 to 0; UC3, <0–1; and UC4 [log (ng/mL)] and their corresponding frequencies were 45.4%, 40.7%, 8.5%, and 5.4%, respectively. Gestational body weight gain, and BMI at delivery were highest in UC4. The age at delivery was lower in UC3 and UC4. UC1, UC2, UC3, and UC4 had 83.5%, 62.2%, 27.2%, and 13.0% of no active or secondhand smoking (SQ0), respectively. Regular alcohol consumption was highest in UC4. The rates of hypertension and diabetes mellitus were slightly higher in UC3 and UC4. Maternal and paternal education of ≥16 years and household income were the lowest in UC4.

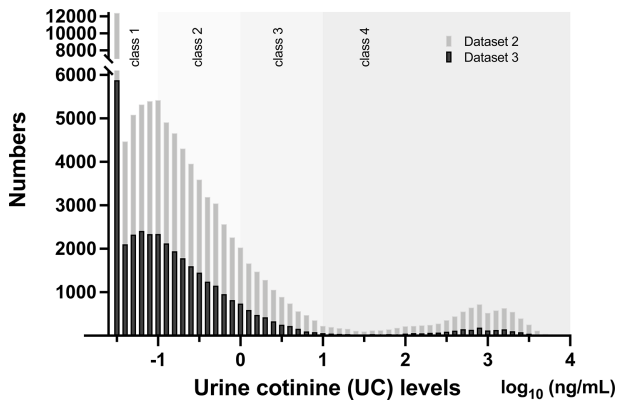


FIGURE 1 | Distribution of urinary cotinine (UC) levels in Dataset 2 (n = 89617) and Dataset 3 (n = 35829) Bars represent numbers in individual UC concentration in log_{10} (ng/mL) and are categorized by UC classes. Please see detail in the text and **Supplement 1**.

TABLE 1 | General characteristics of 35,829 list-wise participants in classes of urine cotinine (UC) concentration.

Factors		Definition	Overall	Classes by UC concentration				P
				UC1	UC2	UC3	UC4	
Numbers of pairs (%)			35,829	16,264 (45.4)	14,579 (40.7)	3,045 (8.5)	1,941 (5.4)	
Mothers								
Pre-pregnancy body weight, kg			52.9 (8.49)	52.7 (8.16)	52.8 (8.42)	53.6 (9.45)	53.8 (9.98)	0.001
Pre-pregnancy BMI, kg/m^2			21.11 (3.15)	21.02 (3.03)	21.08 (3.10)	21.44 (3.52)	21.51 (3.70)	<0.001
Gestational body weight gain, kg			10.09 (5.90)	9.65 (6.45)	10.29 (5.67)	10.85 (4.18)	11.02 (4.42)	<0.001
BMI at delivery (kg/m^2)			25.1 (3.63)	24.9 (3.71)	25.2(3.53)	25.8 (3.46)	26.0 (3.64)	<0.001
Pregnancy period (days)			275.19 (9.92)	274.85 (10.00)	275.54 (9.72)	276.04 (9.82)	274.08 (10.67)	<0.001
Age of mother at derlivery (years)			31.90 (4.68)	32.72 (4.35)	31.47 (4.69)	30.25 (5.16)	30.96 (5.21)	<0.001
Numbers of pairs by SQ class (%)	SQ0	No active nor SHS smoking	23,720 (66.2)	13,573 (83.5)	9,068 (62.2)	827 (27.2)	252 (13.0)	<0.001
	SQ1	SHS < 7h/week	10,047 (28.0)	2,645 (16.3)	5,140 (35.3)	1,711 (56.2)	551 (28.4)	
	SQ2	SHS 7h≤ and <14/ week	692 (1.9)	24 (0.1)	254 (1.7)	304 (10.0)	110 (5.7)	
	SQ3	SHS14h≤/week	433 (1.2)	21 (0.1)	114 (0.8)	201 (6.6)	97 (5.0)	
	SQ4	Active smoking	937 (2.6)	1 (0.0)	3 (0.0)	2 (0.1)	931 (48.0)	
UC (ng/mL)			0.12 [0.05, 0.39]	0.05 [0.03, 0.07]	0.23 [0.15, 0.42]	1.86 [1.32, 3.04]	574.00 [183, 1240]	<0.001
UC log_{10} (ng/mL)			-0.92 [-1.30, -0.41]	-1.34 [-1.52, -1.17]	-0.63 [-0.83, -0.38]	0.27 [0.12, 0.48]	2.76 [2.26, 3.09]	<0.001
Regular alcohol drinking (%)			850 (2.4)	304 (1.9)	350 (2.4)	80 (2.6)	116 (6.0)	<0.001
Hypertention (%)			429 (1.2)	195 (1.2)	153 (1.0)	45 (1.5)	36 (1.9)	0.008
Diabetes mellitus (%)			368 (1.0)	149 (0.9)	143 (1.0)	47 (1.5)	29 (1.5)	0.002
Maternal education ≥ 16 years (%)			9,314 (26.0)	5302 (32.6)	3,494 (24.0)	385 (12.6)	133 (6.9)	<0.001
Paternal education ≥ 16 years (%)			13,561 (37.8)	7,702 (47.4)	4,971 (34.1)	623 (20.5)	265 (13.7)	<0.001
Numbers of pairs by household income class (%)	1	< 4 million Japanese Yen	12,814 (35.8)	4,789 (29.4)	5,443 (37.3)	1,561 (51.3)	1,021 (52.6)	<0.001
	2	4 ≤ and <8	18,682 (52.1)	9,239 (56.8)	7,429 (51.0)	1,236 (40.6)	778 (40.1)	
	3	8 ≤ and < 12	3,683 (10.3)	1,903 (11.7)	1,467 (10.1)	201 (6.6)	112 (5.8)	
	4	12 ≤	650 (1.8)	333 (2.0)	240 (1.6)	47 (1.5)	30 (1.5)	
Infants								
Male gender (%)			18,280 (51.0)	8,271 (50.9)	7,435 (51.0)	1,576 (51.8)	998 (51.4)	0.810
Length or height at birth (cm)			49.0 (2.1)	48.97 (2.12)	49.01 (2.13)	49.01 (2.21)	48.49 (2.25)	<0.001
Body weight at birth (g)			3,026 (399)	3,025 (398)	3,035 (394)	3048 (412)	2,929 (407)	<0.001

Data are number (%), mean (standard deviation), or median [25%, 75%]. UC, urine cotinine; BMI, body mass index; UC1, UC class 1; UC2, UC class 2; UC3, UC class 3; UC4, UC class 4; SQ, smoking questionnaire; SQ0, SQ class 0; SQ1, SQ class 1; SQ2, SQ class 2; SQ3, SQ class 3; SQ4, SQ class 4; SHS, second-hand smoke; P, probability by ANOVA.

Association of UC Class and Infant BMI Trajectory

Trends in BMI of participants in the UC classes before (n = 35829) and after (n = 89617) multiple imputations are shown in **Table 2** and **Figure 2**. BMI data showed normal distributions at birth and six, 12, 18, 24, 30, and 36 months. Overall, the 35829 records including four UC classes showed an increase in the mean BMI from 12.6 (SD 1.17) kg/m^2 at birth to peak 17.2 (1.50) kg/m^2 at six months, which gradually decreased to 16.0 (1.23) kg/m^2 at 36 months, and ΔBMI from baseline peaked at 0.37 (0.15) kg/m^2 at six months and decreased to 0.28 (0.14) kg/m^2 at 36 months.

Since the null hypothesis for equal distribution among the four classes was rejected by the Mauchly's sphericity test, P values in repeated measures ANOVA were adjusted using the Greenhouse–Geisser ϵ correction (**Table 2**). The main effect was BMI (P = 0.000), and BMI × UC class (P = 0.000) was the interaction. BMI and ΔBMI were comparable before and after correction (**Table 2B**). In the 35829 records, UC4 showed the lowest BMI at birth and the highest BMI at six months, gradually decreasing to comparable levels to that of the other three classes at 18, 24, 30, and 36 months (**Table 2A**, **Figure 2A** upper panel). ΔBMI from baseline was the highest at 0.41 (0.16) kg/m^2 at six months and decreased gradually but remained highest at 36 months (**Figure 2A** lower panel). Trends in BMI and ΔBMI corrected by bellow covariates were compared using repeated measures ANCOVA (**Table 2B**, **Figure 2B**). The covariates included were maternal age at delivery (years), pre-pregnancy BMI (kg/m^2), regular alcohol drinking (yes or no), hypertension (yes or no), diabetes mellitus (yes or no), infant sex, maternal education of ≥16 years (yes or no), paternal education of ≥16 years (yes or no), and household income (class 1 to 4).

Multiple Imputation Analyses

The missing rate in BMI data of 89617 participants was significantly different among the four UC classes (**Table 3**): the missing rates at 36 months were the lowest in UC1 (17.6%) and the highest in UC4 (42.6%). Because the missing patterns of variables were considered to be missing not at random (MNAR) (**Supplement 2**), the database was used after multiple imputations (**Table 3**). There were no significant differences in the general characteristics before and after multiple imputations. In Dataset 2, UC levels in $\log_{10}$ showed a bimodal distribution as in Dataset 3 (**Figure 1**). The trends in BMI of participants in UC classes before (n = 35829) and after (n = 89617) multiple imputations are presented in **Table 2C** and **Figure 2C**. After multiple imputations, the statistical differences were largely enhanced, indicating that the trend in BMI was step-wise increased according to the class of UC levels (UC1 < UC2 < UC3 < UC4). The trends in BMI and ΔBMI in the multiple imputation datasets were comparable before and after correction by covariates (**Table 2D** and **Figure 2D**).

Association Between Self-Reported Smoking Status and UC Levels

As shown in **Table 4**, UC levels (median) increased in a stepwise manner for SQ classes [$\log_{10}$ (ng/mL)]: SQ0, −1.09 [IQR, −1.41 to −0.72]; SQ1, −0.57 [−1.03 to −0.06]; SQ2, 0.20 [−0.30 to 0.69]; SQ3, 0.38 [−0.12 to 0.94]; and SQ4, 2.89 [2.57 to 3.18]. However, as shown in **Figure 3A**, fluctuations in UC levels were large. In SQ0, 42.8% showed UC2–4 and in UC1, UC2, and UC3, 73.7%, 96.5%, and 95.1% showed UC2–4, respectively. In SQ4, 99.4% showed UC4. There was a strong discrepancy between the UC classes and SQ classes.

Trends in BMI and ΔBMI of 35829 list-wise participants in the SQ class are shown in **Table 5** and **Figures 3B, C**. After the corrections for covariates, the trends in BMI and ΔBMI were not significantly different between the four classes (**Table 5**).

DISCUSSION

In the current study, we observed that BMI was lower at birth, but the trends in BMI and ΔBMI were higher from six to 36 months step-wise according to UC levels during pregnancy. The effects were observed in the list-wise complete dataset (35829 records), but it was emphasized after multiple imputations and corrections of confounders. In addition, the relationship between the SQ class and trends in BMI and ΔBMI was not statistically significant after the corrections of confounders. Since UC concentration in the five SQ classes largely fluctuated, the self-reported smoking status such as active and second-hand smoking could not be shown to be linked to infant BMI. Collectively, the results suggested that UC concentrations, but not self-reported smoking status, directly affect the BMI trajectory in a dose-dependent manner.

In a study with 630 multi-ethnic offspring, Moore et al. reported that infants exposed prenatally to active or second-hand smoking experience an increase in BMI until three years of age (6), which agrees with our findings. In recent decades, childhood obesity has been considered a major cause of obesity globally (17, 18). Obesity and obesity-related ASCVD have become a major health problem not only in Europe (3) and the United States (4) but also in Asian countries (19).

Previous studies have reported that urinary or serum concentrations of cotinine are related to parents' low educational background and low income (6, 7, 20), which agrees with our findings. However, our study first confirmed that UC levels are strongly linked to postnatal BMI trajectory after correcting socioeconomic statuses, such as parental education and household income. Since the missing rate in BMI was vastly different (UC1, 17.6% vs. UC4, 42.6%), and the missing pattern was MNAR, the analysis in the list-wise complete dataset for 36 months may have caused bias. A multiple imputation model for correcting missing patterns was adopted to minimize this bias, confirming that UC levels were strongly linked to postnatal BMI trajectory. Altogether, our results support that tobacco substances, not *via* altered socioeconomic statuses and other confounding factors, largely change BMI trajectory.

Previous studies have reported underestimating smoking status using a questionnaire and difficulty quantifying active and second-hand smoking (7). There was also a large

TABLE 2 | Trends in body mass index (BMI) of participants in urinary cotinine (UC) classes before (n=35,829) and after (n= 89,617) multiple imputation.

A. Complete set (n = 35,829).

BMI (kg/m^2)	Classes by UC concentration				vs UC1			vs UC2		vs UC3
	UC1	UC2	UC3	UC4	UC2	UC3	UC4	UC3	UC 4	UC4
at birth	12.58 (12.56–12.60)	12.60 (12.58–12.62)	12.65 (12.61–12.69)	12.41 (12.36–12.46)						
at 6 month	17.09 (17.07–17.12)	17.19 (17.16–17.21)	17.28 (17.23–17.33)	17.43 (17.36–17.49)						
at 12 month	16.98 (16.96–17.00)	17.05 (17.03–17.07)	17.12 (17.07–17.16)	17.23 (17.17–17.29)						
at 18 month	16.56 (16.54–16.58)	16.64 (16.61–16.66)	16.69 (16.64–16.75)	16.70 (16.64–16.77)	P = 0.000	P = 0.000	P = 0.000	P = 0.040	P = 0.172	P = 1.000
at 24 month	16.35 (16.33–16.37)	16.42 (16.40–16.44)	16.44 (16.40–16.49)	16.42 (16.36–16.48)						
at 30 month	16.23 (16.21–16.25)	16.26 (16.24–16.28)	16.28 (16.24–16.33)	16.28 (16.22–16.34)						
at 36 month	15.98 (15.96–16.00)	16.00 (15.98–16.02)	16.05 (16.01–16.10)	16.04 (15.99–16.10)						

B. ANCOVA (n = 35,829)

BMI (kg/m^2)	Classes by UC concentration				vs UC1			vs UC2		vs UC3
	UC1	UC2	UC3	UC4	UC2	UC3	UC4	UC3	UC 4	UC4
at birth	12.60 (12.58–12.61)	12.59 (12.57–12.61)	12.60 (12.56–12.64)	12.43 (12.38–12.48)						
at 6 month	17.12 (17.10–17.15)	17.18 (17.15–17.20)	17.21 (17.16–17.27)	17.36 (17.29–17.42)						
at 12 month	17.00 (16.98–17.02)	17.04 (17.02–17.07)	17.08 (17.03–17.13)	17.19 (17.13–17.25)						
at 18 month	16.57 (16.55–16.60)	16.63 (16.61–16.66)	16.66 (16.61–16.71)	16.67 (16.61–16.74)	P = 0.007	P = 0.012	P = 0.016	P = 1.000	P = 0.853	P = 1.000
at 24 month	16.36 (16.34–16.38)	16.41 (16.39–16.44)	16.43 (16.38–16.47)	16.40 (16.34–16.46)						
at 30 month	16.23 (16.21–16.25)	16.26 (16.24–16.28)	16.27 (16.22–16.32)	16.27 (16.21–16.33)						
at 36 month	15.98 (15.97–16.00)	16.00 (15.99–16.02)	16.04 (15.99–16.08)	16.03 (15.98–16.09)						

C. Multiple imputation (n = 89,617)

BMI (kg/m^2)	Classes by UC concentration				vs UC1			vs UC2		vs UC3
	UC1	UC2	UC3	UC4	UC2	UC3	UC4	UC3	UC 4	UC4
at birth	12.59 (12.58–12.61)	12.63 (12.61–12.64)	12.66 (12.63–12.68)	12.47 (12.44–12.49)						
at 6 month	17.14 (17.12–17.16)	17.23 (17.21–17.24)	17.34 (17.31–17.37)	17.49 (17.46–17.53)						
at 12 month	17.00 (16.99–17.02)	17.07 (17.05–17.08)	17.16 (17.13–17.18)	17.31 (17.28–17.34)						
at 18 month	16.56 (16.55–16.58)	16.63 (16.61–16.64)	16.68 (16.65–16.71)	16.79 (16.76–16.83)	P = 0.000	P = 0.000	P = 0.000	P = 0.000	P = 0.000	P = 0.014
at 24 month	16.35 (16.34–16.36)	16.40 (16.39–16.41)	16.43 (16.40–16.45)	16.48 (16.45–16.51)						
at 30 month	16.22 (16.21–16.24)	16.25 (16.23–16.26)	16.29 (16.26–16.32)	16.32 (16.29–16.35)						
at 36 month	15.98 (15.97–15.99)	16.00 (15.99–16.01)	16.04 (16.01–16.06)	16.07 (16.04–16.09)						

D. Multiple imputation + ANCOVA (n = 89,617)

BMI (kg/m^2)	Classes by UC concentration				vs UC1			vs UC2		vs UC3
	UC1	UC2	UC3	UC4	UC2	UC3	UC4	UC3	UC 4	UC4
at birth	12.62 (12.60–12.63)	12.61 (12.60–12.62)	12.62 (12.59–12.64)	12.48 (12.45–12.50)						
at 6 month	17.18 (17.17–17.20)	17.22 (17.21–17.24)	17.27 (17.24–17.30)	17.42 (17.39–17.46)						
at 12 month	17.02 (17.00–17.04)	17.07 (17.05–17.08)	17.12 (17.09–17.14)	17.26 (17.23–17.29)						
at 18 month	16.58 (16.57–16.60)	16.62 (16.61–16.64)	16.65 (16.62–16.68)	16.76 (16.73–16.79)	P = 0.001	P = 0.000	P = 0.000	P = 0.059	P = 0.000	P = 0.004
at 24 month	16.36 (16.34–16.37)	16.40 (16.39–16.41)	16.42 (16.39–16.45)	16.46 (16.44–16.49)						
at 30 month	16.23 (16.21–16.24)	16.25 (16.23–16.26)	16.28 (16.25–16.31)	16.31 (16.28–16.34)						
at 36 month	15.99 (15.97–16.00)	16.00 (15.99–16.01)	16.03 (16.00–16.05)	16.06 (16.03–16.08)						

Data are mean (95% confidenctial interval). UC, urine cotinine; BMI, body mass index; UC1, UC class 1; UC2, UC class 2; UC3, UC class 3; UC4, UC class 4. P, provability for group differences made by the Bonferroni correction post-hoc after repeated measures ANOVA adjusted using the Greenhouse–Geisser ϵ correction.

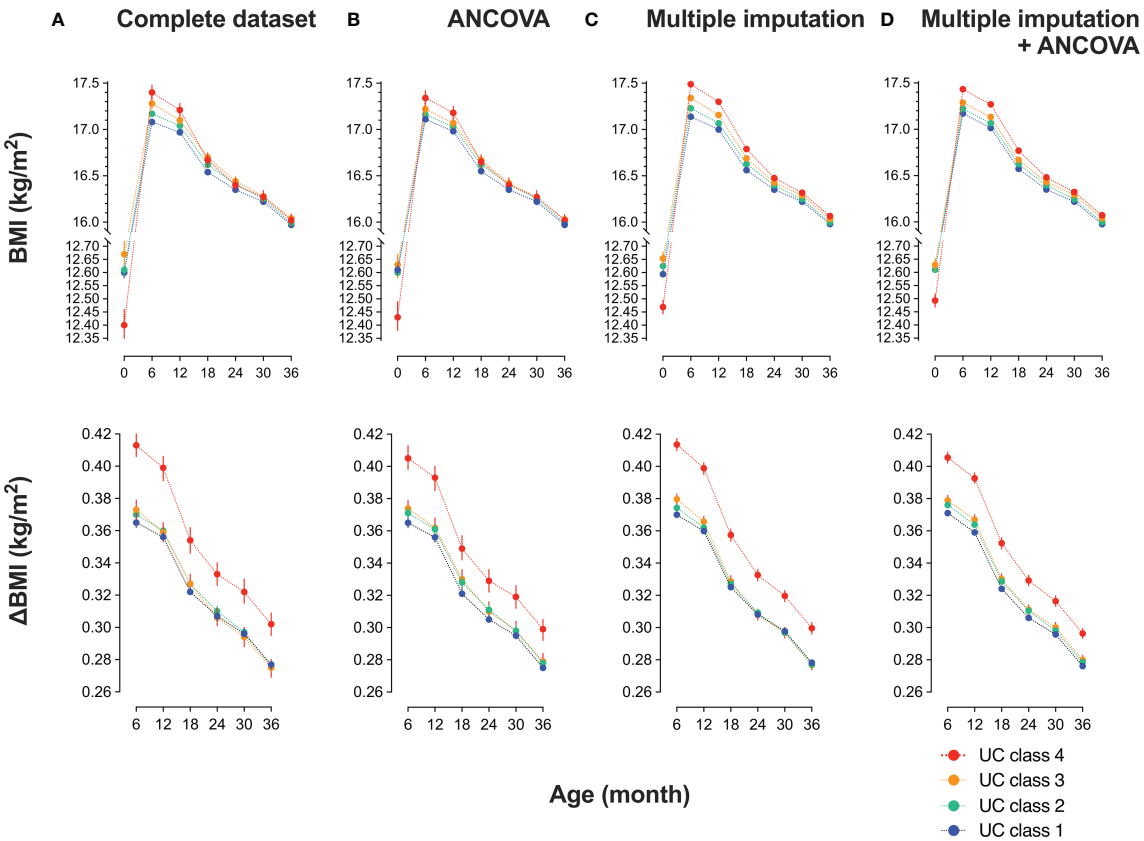


FIGURE 2 | Trends in body mass index (BMI) of participants in urinary cotinine (UC) classes before and after multiple imputations Participants were categorized into four classes according to $\log_{10}$ UC levels [$\log_{10}$ (ng/mL)]: class 1: <−1, class 2: <−1 to 0, class 3: <0–1, class 4: ≥1. The trend in BMI and the differences from baseline (ΔBMI) are shown according to UC class before and after multiple imputations. Values are shown in **(A)** a complete set (n = 35829); **(B)** complete set plus repeated measured analysis of covariance (ANCOVA) corrected by covariates: mother's age at delivery (years), pre-pregnancy BMI (kg/m^2), regular alcohol drinking (yes or no), hypertension (yes or no), diabetes mellitus (yes or no), infant sex, pregnancy period, maternal education of ≥16 years (yes or no), paternal education of ≥16 years (yes or no), household income (1, <4 million; 2, ≥4 million and <8 million; 3, ≥8 million and <12 million; 4, ≥12 million in Japanese Yen); **(C)** dataset (n = 89617) imputed the missing confounders on BMI at six, nine, 12, 18, 24, and 36 months, maternal age, BMI before pregnancy, body weight gain during pregnancy, alcohol drinking during pregnancy, maternal and paternal education of ≤16 years, and class of annual household income; and **(D)** dataset imputed as in **(C)** plus ANCOVA done as in **(B)**.

discrepancy between the classes categorized by UC levels and the smoking status questionnaire in our research. In the absence of active or second-hand smoking (SQ0), 42.8% showed UC2–4. In contrast, UC4 included 48.0% active smoking class (SQ 4) and 52.0% no active smoking class (SQ0, SQ1, SQ2, and SQ3). These results indicate that smoking status by SQ includes potential uncertainty. Combined, although urinary cotinine can estimate active smoking, it would be challenging to accurately detect passive smoking based on smoking questionnaires. Therefore, it may be reasonable to consider urinary cotinine as desirable for evaluating true smoking status (especially passive smoking and second-hand smoke) because interview alone is insufficient, as claimed in this paper. Clearly, the validity and reliability of this self-reported SQ class need to be evaluated compared to the UC classes in future studies of different ethnicities and populations. In our study, the ROC analysis of UC levels showed that the AUC for active smoking was 0.980 [95% CI 0.979–0.982], and the AUC for second-hand smoking was 0.762 [95% CI 0.759–0.766], which are similar to the findings of the previous profile paper (16). Therefore, it is considered that the accurate assessment of smoking during pregnancy needs a smoking status questionnaire and an evaluation of cotinine levels.

In our study, BMI was lower at birth, but trends in BMI and ΔBMI were higher from six to 36 months of age with the levels of maternal UC. This indicates that the catch-up-growth in infants from mothers exposed to active or second-hand smoking occurs six months after birth (11). A hypothesis concerning the mechanism that causes a rapid rise in BMI after delivery of low birth weight infants includes the effects of the neuroendocrine system, including growth hormone (9, 21) and acceleration of compensatory cell proliferation (22). An earlier BMI peak has been reported contributing to an increased risk of future obesity and ASCVD (23, 24). Lindström et al. said in an observational study in Sweden that the catch-up-growth rate of children born small for gestational age from smoking mothers was greater than that of children from nonsmoking mothers (11).

TABLE 3 | General characteristics of 89,617 participants in classes by urine cotinine (UC) concentration before and after multiple imputation.

Factors		Definition	Classes by UC concentration						Missing					Multiple imputation					
			Overall	UC1	UC2	UC3	UC4	*P*	Overall	UC1	UC2	UC3	UC4	Overall	UC1	UC2	UC3	UC4	*P*
Numbers of pairs (%)			89,617	35,357 (39.5)	36,207 (40.4)	9,657 (10.8)	8,396 (9.4)												
Mothers																			
Pre-pregnancy body weight (kg)			53.1 (8.87)	52.9 (8.32)	53.0 (8.65)	53.6 (9.89)	54.0 (10.57)	<0.001	39 (0.0)	7 (0.0)	17 (0.0)	7 (0.1)	8 (0.1)	53.1 (8.87)	52.9 (8.31)	53.0 (8.65)	53.6 (9.89)	54.0 (10.57)	<0.001
Pre-pregnancy BMI (kg/m^2)			21.2 (3.30)	21.1 (3.08)	21.2 (3.21)	21.5 (3.69)	21.6 (3.95)	<0.001	51 (0.1)	9 (0.0)	20 (0.1)	12 (0.1)	10 (0.1)	21.2 (3.30)	21.1 (3.07)	21.2 (3.21)	21.5 (3.69)	21.6 (3.95)	<0.001
Gestational body weight gain (kg)			10.3 (4.94)	9.74 (5.23)	10.42 (4.76)	11.14 (4.43)	11.20 (4.67)	<0.001	1,538 (1.7)	724 (2.0)	609 (1.7)	121 (1.3)	84 (1.0)	10.32 (4.93)	9.77 (5.21)	10.44 (4.76)	11.13 (4.44)	11.20 (4.68)	<0.001
BMI at delivery (kg/m^2)			25.4 (3.49)	25.0 (3.42)	25.4 (3.40)	26.0 (3.61)	26.1 (3.77)	<0.001	1,516 (1.7)	719 (2.0)	596 (1.6)	121 (1.3)	80 (1.0)	25.4 (3.47)	25.0 (3.39)	25.4 (3.38)	26.0 (3.59)	26.1 (3.76)	<0.001
Pregnancy period (days)			274.88 (11.23)	274.52 (11.53)	275.32 (10.63)	275.50 (11.54)	273.81 (11.93)	<0.001	0	0	0	0	0	274.88 (11.22)	274.52 (11.53)	275.32 (10.63)	275.50 (11.54)	273.81 (11.93)	<0.001
Age of mother at derlivery (years)			31.19 (5.03)	32.48 (4.46)	30.88 (4.91)	28.92 (5.48)	29.68 (5.60)	<0.001	5 (0.0)	3 (0.0)	2 (0.0)	0	0	31.19 (5.02)	32.48 (4.46)	30.88 (4.91)	28.92 (5.48)	29.68 (5.60)	<0.001
Numbers of pairs by SQ class (%)	SQ0	No	54,578 (60.9)	29,412 (83.2)	21,866 (60.4)	2,359 (24.4)	941 (11.2)	<0.001	0	0	0	0	0	54,578 (60.9)	29,412 (83.2)	21,866 (60.4)	2,359 (24.4)	941 (11.2)	<0.001
	SQ1	SHS < 7h/week	26,532 (29.6)	5,828 (16.5)	13,255 (36.6)	5,311 (55.0)	2,138 (25.5)		0	0	0	0	0	26,532 (29.6)	5,828 (16.5)	13,255 (36.6)	5,311 (55.0)	2,138 (25.5)	
	SQ2	SHS 7h≤ and <14/week	2,509 (2.8)	74 (0.2)	731 (2.0)	1,114 (11.5)	590 (7.0)		0	0	0	0	0	2,509 (2.8)	74 (0.2)	731 (2.0)	1,114 (11.5)	590 (7.0)	
	SQ3	SHS14h≤/week	1,756 (2.0)	39 (0.1)	328 (0.9)	828 (8.6)	561 (6.7)		0	0	0	0	0	1,756 (2.0)	39 (0.1)	328 (0.9)	828 (8.6)	561 (6.7)	
	SQ4	Maternal smoking	4,242 (4.7)	4 (0.0)	27 (0.1)	45 (0.5)	4,166 (49.6)		0	0	0	0	0	4,242 (4.7)	4 (0.0)	27 (0.1)	45 (0.5)	4,166 (49.6)	
UC (ng/mL)			0.15 [0.06, 0.64]	0.05 [0.03, 0.07]	0.24 [0.15, 0.44]	1.99 [1.37, 3.42]	626 [188, 1,370]	<0.001	0	0	0	0	0	0.15 [0.06, 0.64]	0.05 [0.03, 0.07]	0.24 [0.15, 0.44]	1.99 [1.37, 3.42]	626 [188, 1,370]	<0.001
UC log_{10} (ng/mL)			-0.81 [-1.24, -0.19]	-1.33 [-1.52, -1.16]	-0.61 [-0.82, -0.36]	0.30 [0.14, 0.53]	2.80 [2.27, 3.14]	<0.001	0	0	0	0	0	-0.81 [-1.24, -0.19]	-1.33 [-1.52, -1.16]	-0.61 [-0.82, -0.36]	0.30 [0.14, 0.53]	2.80 [2.27, 3.14]	<0.001
Regular alcohol drinking (%)			2,510 (2.8)	739 (2.1)	923 (2.6)	303 (3.2)	545 (6.6)	<0.001	703 (0.8)	268 (0.8)	256 (0.7)	90 (0.9)	112 (1.3)	2526.8 (2.82)	745.2 (2.11)	925.9 (2.56)	305.2 (3.16)	550.6 (6.56)	<0.001
Hypertention (%)			1,092 (1.2)	405 (1.1)	402 (1.1)	144 (1.5)	141 (1.7)	<0.001	0	0	0	0	0	1,092 (1.2)	405 (1.1)	402 (1.1)	144 (1.5)	141 (1.7)	<0.001
Diabetes mellitus (%)			964 (1.1)	349 (1.0)	366 (1.0)	135 (1.4)	114 (1.4)	<0.001	0	0	0	0	0	964 (1.1)	349 (1.0)	366 (1.0)	135 (1.4)	114 (1.4)	<0.001
Maternal education ≥ 16 years (%)			19,291 (21.6)	10,620 (30.2)	7,409 (20.6)	909 (9.5)	353 (4.2)	<0.001	464 (0.5)	141 (0.4)	172 (0.5)	63 (0.7)	88 (1.0)	19,372.5 (21.62)	10,659.8 (30.14)	7,441.2 (20.55)	914.7 (9.47)	356.9 (4.25)	<0.001
Paternal education ≥ 16 years (%)			29,451 (33.2)	15,961 (45.5)	11,098 (30.9)	1,497 (15.8)	895 (10.9)	<0.001	1,014 (1.1)	268 (0.8)	331 (0.9)	196 (2.0)	219 (2.6)	29,685.1 (33.12)	16,067.8 (45.44)	11,184.0 (30.89)	1,519.4 (15.73)	914.0 (10.89)	<0.001
Numbers of pairs by household income class (%)	1	< 4 million Japanese Yen	33,581 (40.3)	10,356 (31.1)	13,671 (40.5)	5,006 (57.3)	4,548 (60.2)	<0.001	6,233 (7.0)	2,055 (5.8)	2,425 (6.7)	918 (9.5)	835 (9.9)	36,553.6 (40.79)	11,063.3 (31.29)	14,796.5 (40.87)	5,593.5 (57.92)	5,100.3 (60.75)	<0.001
	2	4 ≤ and <8	40,784 (48.9)	18,547 (55.7)	16,482 (48.8)	3,155 (36.1)	2,600 (34.4)							43,531.6 (48.58)	19,657.6 (55.60)	17,579.1 (48.55)	3,443.4 (35.66)	2,851.6 (33.96)	
	3	8 ≤ and < 12	7,456 (8.9)	3,661 (11.0)	3,011 (8.9)	468 (5.4)	316 (4.2)							7,880.4 (8.79)	3,860.1 (10.92)	3,179.7 (8.78)	501.9 (5.20)	338.8 (4.04)	
	4	12 ≤	1,563 (1.9)	738 (2.2)	618 (1.8)	110 (1.3)	97 (1.3)							1,651.5 (1.84)	776.2 (2.20)	651.7 (1.80)	118.3 (1.22)	105.4 (1.26)	
Infants																			
Male gender (%)			45,955 (51.3)	18,120 (51.2)	18,542 (51.2)	4,957 (51.3)	4336 (51.6)	0.897	0	0	0	0	0	45,955 (51.3)	18,120 (51.2)	18,542 (51.2)	4,957 (51.3)	4336 (51.6)	0.911
Length or height at birth (cm)			48.96 (2.2)	48.98 (2.20)	49.03 (2.16)	49.00 (2.21)	48.47 (2.30)	<0.001	0	0	0	0	0	48.96 (2.2)	48.98 (2.20)	49.03 (2.16)	49.00 (2.21)	48.47 (2.30)	<0.001
Body weight at birth (kg)			3,030.1 (406.17)	3,032.5 (408.26)	3,043.9 (400.23)	3,047.8 (407.35)	2,940.5 (410.12)	<0.001	0	0	0	0	0	3,030.1 (406.17)	3,032.5 (408.26)	3,043.9 (400.23)	3,047.8 (407.35)	2,940.5 (410.12)	<0.001
BMI (kg/m2)																			
at birth			12.6 (1.19)	12.6 (1.18)	12.6 (1.20)	12.7 (1.19)	12.5 (1.22)	<0.001	0	0	0	0	0	12.6 (1.19)	12.6 (1.18)	12.6 (1.20)	12.7 (1.19)	12.5 (1.22)	<0.001
at 6 month			17.2 (1.51)	17.1 (1.50)	17.2 (1.50)	17.3 (1.56)	17.5 (1.54)	<0.001	7,807 (8.7)	2,064 (5.8)	2,892 (8.0)	1,185 (12.3)	1,666 (19.8)	17.2 (1.51)	17.1 (1.50)	17.2 (1.50)	17.3 (1.55)	17.5 (1.54)	<0.001
at 12 month			17.0 (1.35)	17.0 (1.34)	17.0 (1.34)	17.1 (1.39)	17.3 (1.39)	<0.001	28,226 (31.5)	9,614 (27.2)	11,155 (30.8)	3,480 (36.0)	3,977 (47.4)	17.1 (1.36)	17.0 (1.34)	17.1 (1.35)	17.2 (1.38)	17.3 (1.40)	<0.001
at 18 month			16.6 (1.50)	16.6 (1.46)	16.6 (1.49)	16.7 (1.55)	16.8 (1.68)	<0.001	19,563 (21.8)	5,993 (16.9)	7,555 (20.9)	2,767 (28.7)	3,248 (38.7)	16.6 (1.50)	16.6 (1.47)	16.6 (1.49)	16.7 (1.54)	16.8 (1.62)	<0.001
at 24 month			16.4 (1.35)	16.4 (1.33)	16.4 (1.35)	16.4 (1.35)	16.5 (1.41)	<0.001	18,336 (20.5)	5,493 (15.5)	7,066 (19.5)	2,567 (26.6)	3,210 (38.2)	16.4 (1.35)	16.4 (1.33)	16.4 (1.35)	16.4 (1.35)	16.5 (1.40)	<0.001
at 30 month			16.2 (1.34)	16.2 (1.31)	16.3 (1.33)	16.3 (1.40)	16.3 (1.46)	<0.001	21,316 (23.8)	6,428 (18.2)	8,303 (22.9)	3,026 (31.3)	3,559 (42.4)	16.2 (1.34)	16.2 (1.31)	16.2 (1.33)	16.3 (1.38)	16.3 (1.42)	<0.001
at 36 month			16.0 (1.25)	16.0 (1.22)	16.0 (1.25)	16.0 (1.29)	16.1 (1.37)	<0.001	20,815 (23.2)	6,212 (17.6)	8,043 (22.2)	2,985 (30.9)	3,575 (42.6)	16.0 (1.26)	16.0 (1.23)	16.0 (1.26)	16.0 (1.28)	16.1 (1.33)	<0.001

Data are number (%), mean (standard deviation), or median [25%, 75%]. UC, urine cotinine; BMI, body mass index; UC1, UC class 1; UC2, UC class 2; UC3, UC class 3; UC4, UC class 4; SQ, smoking questionnaire; SQ0, SQ class 0; SQ1, SQ class 1; SQ2, SQ class 2; SQ3, SQ class 3; SQ4, SQ class 4; SHS, second-hand smoke; P, probability by ANOVA.

TABLE 4 | General characteristics of 35,829 list-wise participants in classes by smoking questionnaire (SQ).

Factors		Definition	Overall	Classes by SQ					P
				SQ0 No active nor SHS smoking	SQ1 SHS < 7h/week	SQ2 SHS 7h≤ and <14/ week	SQ3 SHS14h≤/ week	SQ4 Active smoking	
Numbers of pairs (%)			35,829	23,720 (66.2)	10,047 (28.0)	692 (1.9)	433 (1.2)	937 (2.6)	
Mothers									
Pre-pregnancy body weight (kg)			52.9 (8.49)	52.61 (8.13)	53.41 (8.98)	53.65 (9.75)	54.11 (10.20)	53.75 (9.77)	<0.001
Pre-pregnancy BMI (kg/m^2)			21.1 (3.15)	21.0 (3.00)	21.3 (3.35)	21.4 (3.55)	21.7 (3.70)	21.5 (3.66)	<0.001
Gestational body weight gain (kg)			10.10 (5.90)	9.90 (4.99)	10.35 (7.81)	11.03 (4.18)	11.18 (5.05)	11.16 (4.42)	<0.001
BMI at delivery (kg/m^2)			25.1 (3.63)	24.9 (3.33)	25.5 (4.22)	25.8 (3.55)	26.2 (3.61)	26.0 (3.58)	<0.001
Pregnancy period (days)			275.19 (9.92)	275.14 (9.87)	275.34 (9.93)	275.89 (10.11)	275.54 (9.95)	274.00 (10.83)	<0.001
Age of mother at derlivery (years)			31.90 (4.68)	32.25 (4.52)	31.32 (4.82)	30.52 (5.29)	30.30 (5.45)	31.13 (5.22)	<0.001
UC (ng/mL)			0.12 [0.05, 0.39]	0.08 [0.04, 0.19]	0.27 [0.09, 0.86]	1.60 [0.50, 4.89]	2.39 [0.77, 8.72]	769 [370, 1520]	<0.001
UC log10 (ng/mL)			-0.92 [-1.31, -0.41]	-1.09 [-1.41, -0.72]	-0.57 [-1.03, -0.06]	0.20 [-0.30, 0.69]	0.38 [-0.12, 0.94]	2.89 [2.57, 3.18]	<0.001
Numbers of pairs by UC class (%)	UC1	<-1 log (ng/mL)	16,264 (45.4)	13,573 (57.2)	2,645 (26.3)	24 (3.5)	21 (4.8)	1 (0.1)	<0.001
	UC2	-1≤ and <0 log (ng/ mL)	14,579 (40.7)	9,068 (38.2)	5,140 (51.2)	254 (36.7)	114 (26.3)	3 (0.3)	
	UC3	0≤ and <1 log (ng/mL)	3,045 (8.5)	827 (3.5)	1,711 (17.0)	304 (43.9)	201 (46.4)	2 (0.2)	
	UC4	1≤ log (ng/mL)	1,941 (5.4)	252 (1.1)	551 (5.5)	110 (15.9)	97 (22.4)	931 (99.4)	
Regular alcohol drinking (%)			850 (2.4)	470 (2.0)	260 (2.6)	21 (3.0)	18 (4.2)	81 (8.6)	<0.001
Hypertention (%)			429 (1.2)	261 (1.1)	132 (1.3)	12 (1.7)	6 (1.4)	18 (1.9)	0.610
Diabetes mellitus (%)			368 (1.0)	225 (0.9)	114 (1.1)	11 (1.6)	6 (1.4)	12 (1.3)	0.209
Maternal education ≥ 16 years (%)			9,314 (26.0)	7,217 (30.4)	1,944 (19.3)	67 (9.7)	37 (8.5)	49 (5.2)	<0.001
Paternal education ≥ 16 years (%)			13,561 (37.8)	10,312 (43.5)	2,935 (29.2)	135 (19.5)	74 (17.1)	105 (11.2)	<0.001
Numbers of pairs by household income class (%)	1	< 4 million Japanese Yen	12,814 (35.8)	7,594 (32.0)	4,099 (40.8)	367 (53.0)	244 (56.4)	510 (54.4)	<0.001
	2	4 ≤ and <8	18,682 (52.1)	12,990 (54.8)	4,903 (48.8)	269 (38.9)	153 (35.3)	367 (39.2)	
	3	8 ≤ and < 12	3,683 (10.3)	2,672 (11.3)	886 (8.8)	49 (7.1)	28 (6.5)	48 (5.1)	
	4	12 ≤	650 (1.8)	464 (2.0)	159 (1.6)	7 (1.0)	8 (1.8)	12 (1.3)	
Infants									
Male gender (%)			18,280 (51.0)	12,127 (51.1)	5,093 (50.7)	345 (49.9)	237 (54.7)	478 (51.0)	0.508
Height at birth (cm)			49.0 (2.1)	48.98 (2.13)	48.97 (2.15)	48.96 (2.23)	49.09 (2.23)	48.24 (2.25)	<0.001
Body weight at birth (g)			3,026 (399)	3,030 (395)	3,029 (402)	3,043 (408)	3,040 (419)	2,893 (402)	<0.001

Data are number (%), mean (standard deviation), or median [25%, 75%]. UC, urine cotinine; BMI, body mass index; SQ, smoking questionnaire; SQ0, SQ class 0; SQ1, SQ class 1; SQ2, SQ class 2; SQ3, SQ class 3; SQ4, SQ class 4; SHS, second-hand smoke; P, probability by ANOVA.

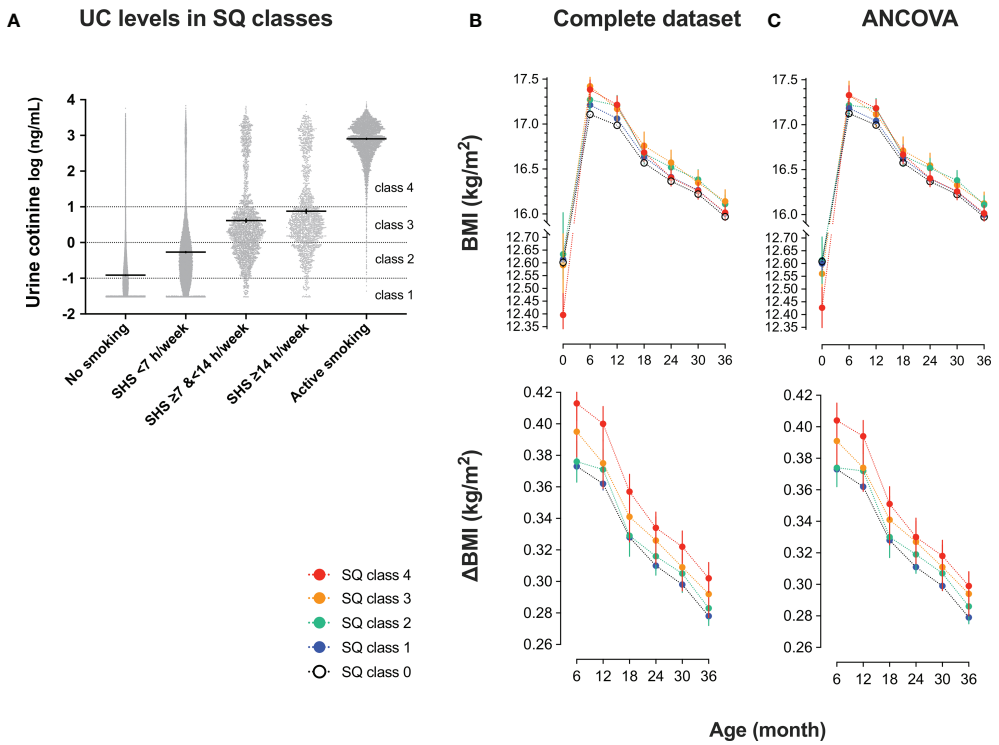


FIGURE 3 | Urinary cotinine (UC) levels during pregnancy and trends in body mass index (BMI) of 35829 list-wise participants in classes by smoking questionnaire (SQ) **(A)** UC levels in SQ classes. Participants were categorized into five SQ classes for active and second-hand smoking: SQ class 0 (no active nor second-hand smoking); SQ class 1 (second-hand smoking, <7 hours/week); SQ class 2 (second-hand smoking, ≥7 hours, and <14 hours/week); SQ class 3 (second-hand smoking, ≥14 hours/week); SQ class 4 (active smoking). According to SQ classes, trends in BMI and the differences from baseline (ΔBMI) are shown. Values are shown in **(B)** complete set (n = 35829) and **(C)** complete set plus repeated measured analysis of covariance corrected by covariates: mother's age at delivery (years), pre-pregnancy BMI (kg/m^2), regular alcohol drinking (yes or no), hypertension (yes or no), diabetes mellitus (yes or no), infant sex, pregnancy period, maternal education of ≥16 years (yes or no), paternal education of ≥16 years (yes or no), household income (1, <4 million; 2, ≥4 million and <8 million; 3, ≥8 million and <12 million; 4, ≥12 million in Japanese Yen).

It has been hypothesized that overeating in obese individuals shares, at least partly, common mechanisms with addiction to nicotine as well as to alcohol and narcotics in the brain reward system (25, 26). Richardson and Tizabi reported that neonates from nicotine-treated rats showed altered dopaminergic pathways in the striatum and ventral tegmental area of the reward system (27). Romoli et al. reported that neonatal nicotine exposure primes the reward system to display increased susceptibility to nicotine and alcohol consumption in adulthood (28). Furthermore, Thomas et al. described that nicotine exposure during adolescence affects the reward system of mice and increases alcohol abuse (29). Although the direct effects of maternal smoking on neonatal overeating and obesity remain undetermined, exposure of the fetus and neonate to nicotine may be linked to excessive eating behavior *via* alteration in the reward system (25, 26), and it needs to be evaluated whether high UC levels during pregnancy alter BMI trajectory *via* modified eating behavior.

Strengths and Limitations

A strength of our study is its size, covering almost the whole area of Japan and nearly 100,000 pairs of mother and infant. However, our study has several limitations. First, there is an issue regarding the reproducibility of UC levels. This study measured only the UC levels in the second or third trimester. The half-life of UC was reported to be approximately 72–96 hours (30), and it might be considered valuable even once. Future studies need to evaluate the reproducibility, effectiveness, and usefulness of UC measurements. Second, there were many missing cases because of the large observational cohort. Particularly, there were many cases of dropouts in the high UC group. This was addressed by supplementing the missing values and conducting a sensitivity analysis. Third, quantification of milk intake immediately after delivery and evaluation of nutritional aspects were insufficient. Energy intake exceeding energy expenditure is a primary cause of BMI rise, and it is a problem that needs to be evaluated as much as possible in the future. BMI, height, weight-for-height, and upper arm circumference are nutritional indices in infants (31), but only BMI was evaluated as a nutritional index in the current study, limiting the interpretation. Although there have been discussions about developing infant nutrition indicator tools in recent years, no representative tools have been established (32). While validating nutrition assessment indicators, it is asked in future studies to clarify the effects of maternal smoking on outcomes and infant growth through the analysis of these nutrition indicators. Fourth, the observation period was as short as three years after birth. In this

TABLE 5 | Trend in body mass index of 35,829 list-wise participants in classes by smoking questionnaire (SQ).

A. Complete set

BMI (kg/m^2)	Classes by SQ					vs SQ0				vs SQ1			vs SQ2		vs SQ3
	SQ0	SQ1	SQ2	SQ3	SQ4	SQ1	SQ2	SQ3	SQ4	SQ2	SQ3	SQ4	SQ3	SQ4	SQ4
at birth	12.59 (12.57–12.60)	12.59 (12.57–12.61)	12.66 (12.58–12.75)	12.57 (12.46–12.68)	12.38 (12.31–12.46)										
at 6 month	17.12 (17.10–17.14)	17.22 (17.20–17.25)	17.31 (17.20–17.43)	17.42 (17.28–17.56)	17.40 (17.30–17.49)										
at 12 month	17.00 (16.98–17.02)	17.08 (17.05–17.10)	17.20 (17.10–17.30)	17.17 (17.04–17.30)	17.23 (17.14–17.32)										
at 18 month	16.59 (16.57–16.60)	16.65 (16.62–16.68)	16.72 (16.61–16.82)	16.79 (16.65–16.93)	16.71 (16.62–16.80)	P =	P =	P =	P =	P =	P =	P =	P =	P =	P =
at 24 month	16.37 (16.35–16.38)	16.42 (16.40–16.50)	16.50 (16.40–16.59)	16.58 (16.45–16.70)	16.40 (16.32–16.49)	0.000	0.001	0.003	0.170	0.201	0.156	1.000	1.000	1.000	1.000
at 30 month	16.23 (16.21–16.25)	16.27 (16.25–16.30)	16.38 (16.28–16.47)	16.37 (16.25–16.49)	16.27 (16.19–16.36)										
at 36 month	15.98 (15.97–16.00)	16.02 (16.00–16.05)	16.13 (16.04–16.22)	16.17 (16.05–16.28)	16.02 (15.94–16.10)										

B. ANCOVA

	SQ classes by SQ					vs SQ0				vs SQ1			vs SQ2		vs SQ3
BMI (kg/m^2)	SQ0	SQ1	SQ2	SQ3	SQ4	SQ1	SQ2	SQ3	SQ4	SQ2	SQ3	SQ4	SQ3	SQ4	SQ4
at birth	12.60 (12.58–12.61)	12.58 (12.55–12.60)	12.62 (12.54–12.70)	12.53 (12.43–12.63)	12.41 (12.34–12.48)										
at 6 month	17.14 (17.12–17.16)	17.19 (17.16–17.22)	17.26 (17.15–17.36)	17.32 (17.18–17.46)	17.33 (17.23–17.42)										
at 12 month	17.01 (17.00–17.03)	17.06 (17.03–17.08)	17.17 (17.07–17.26)	17.10 (16.98–17.22)	17.18 (17.10–17.27)										
at 18 month	16.60 (16.58–16.61)	16.63 (16.60–16.66)	16.70 (16.58–16.80)	16.73 (16.60–16.87)	16.68 (16.58–16.77)	P =	P =	P =	P =	P =	P =	P =	P =	P =	P =
at 24 month	16.37 (16.36–16.39)	16.41 (16.39–16.44)	16.49 (16.39–16.58)	16.54 (16.41–16.66)	16.38 (16.30–16.47)	0.216	0.033	0.222	1.000	0.285	0.891	1.000	1.000	1.000	1.000
at 30 month	16.24 (16.22–16.25)	16.26 (16.24–16.29)	16.37 (16.27–16.46)	16.34 (16.22–16.46)	16.26 (16.18–16.34)										
at 36 month	15.99 (15.97–16.00)	16.01 (15.99–16.03)	16.12 (16.03–16.21)	16.14 (16.02–16.25)	16.01 (15.93–16.09)										

Data are mean (95% confidenctial interval). UC, urine cotinine; BMI, body mass index; SQ, smoking questionnaire; SQ0, SQ class 0; SQ1, SQ class 1; SQ2, SQ class 2; SQ3, SQ class 3; SQ4, SQ class 4; P, provability for group differences made by the Bonferroni correction post-hoc after repeated measures ANOVA adjusted using the Greenhouse–Geisser ε correction.

regard, the JECS is still continuously conducting an observational study for the mother-infant relationship. Sixth, length at birth is rather overestimated as no proper neonatal length equipment is used for measurements performed inside the delivery rooms settings (33). We did not use identical measuring equipment at multiple recruitment sites because of the large numbers. The child's length is used instead of height 1.5-2 cm higher than height (33). We did not assess the supine position (length) or the standing position (height) in the current study. Thus, BMI calculated from length in our measurements might be lower. Assessing a BMI trajectory from 0-3 years of age must be carefully interpreted. Seventh, the question that remains open is whether more children from mothers with active or second-hand smoking present or not an early adiposity rebound, even before the age of 3 years earlier than the one observed in small for gestational age (SGA) children. Although we could not answer this question in the current study, future studies need to clarify the underlying mechanisms for the link between mothers' UC levels and infant BMI trajectory. Eighth, we did not collect "participants' medication history," which may affect body weight.

In conclusion, BMI was lower at birth, but the trends in BMI and ΔBMI were higher from six to 36 months step-wise according to UC levels during pregnancy. The effects were observed in the list-wise complete dataset, but it was emphasized after multiple imputations and corrections of confounders. The relationship between the categorical class of smoking status questionnaire and trends in BMI was not statistically significant. In mothers, UC concentrations, but not self-reported smoking status for active and second-hand smoking, can predict infant BMI trajectory in a dose-dependent manner. The study findings have important implications for mothers with active or second-hand smoking, healthcare professionals, and policymakers.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Ministry of the Environment's Institutional Review Board on Epidemiological Studies and the Ethics Committees of all participating institutions. Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

JAPAN ENVIRONMENT AND CHILDREN'S STUDY (JECS) GROUP MEMBERS

The work was performned on behalf of the Japan Environment and Children's Study (JECS) group (as of 2021): Michihiro

Kamijima (principal investigator, Nagoya City University, Nagoya, Japan), Shin Yamazaki (National Institute for Environmental Studies, Tsukuba, Japan), Yukihiro Ohya (National Center for Child Health and Development, Tokyo, Japan), Reiko Kishi (Hokkaido University, Sapporo, Japan), Nobuo Yaegashi (Tohoku University, Sendai, Japan), KH (Fukushima Medical University, Fukushima, Japan), ChisatoMori (Chiba University, Chiba, Japan), Shuichi Ito (Yokohama City University, Yokohama, Japan), ZY (University of Yamanashi, Chuo, Japan), Hidekuni Inadera (University of Toyama, Toyama, Japan), Takeshi Ebara(Nagoya City University Graduate School of Medical Sciences, Nagoya, Japan),Takeo Nakayama (Kyoto University, Kyoto, Japan), Hiroyasu Iso (Osaka University, Suita, Japan), Masayuki Shima (Hyogo College of Medicine, Nishinomiya, Japan), Youichi Kurozawa (Tottori University, Yonago, Japan), Narufumi Suganuma (Kochi University, Nankoku, Japan), Koichi Kusuhara (University of Occupational and Environmental Health, Kitakyushu, Japan), and Takahiko Katoh (Kumamoto University, Kumamoto, Japan).

AUTHOR CONTRIBUTIONS

HH and MS conceptualized the study, analyzed the data, and wrote the manuscript. SO, HM, TM, YO, AS, SH, RS, KS, HN, KF, MH, SY KH, and ZY reviewed and approved the final draft. The Japan Environment and Children's Study Group collected data and reviewed and approved the final draft.

ACKNOWLEDGMENTS

This work was supported by the Ministry of the Environment, Japan. The authors sincerely thank all the study participants, research coordinators, and researchers for their role in the JECS.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2022.850784/full#supplementary-material

REFERENCES

1. Juonala M, Magnussen CG, Berenson GS, Venn A, Burns TL, Sabin MA, et al. Childhood Adiposity, Adult Adiposity, and Cardiovascular Risk Factors. *N Engl J Med* (2011) 365:1876–85. doi: 10.1056/NEJMoa1010112
2. Bjerregaard LG, Adelborg K, Baker JL. Change in Body Mass Index From Childhood Onwards and Risk of Adult Cardiovascular Disease. *Trends Cardiovasc Med* (2020) 30:39–45. doi: 10.1016/j.tcm.2019.01.011
3. Nittari G, Scuri S, Petrelli F, Pirillo I, di Luca NM, Grappasonni I. Fighting Obesity in Children From European World Health Organization Member States. Epidemiological Data, Medical-Social Aspects, and Prevention Programs. *Clin Ter* (2019) 170:e223–e30. doi: 10.7417/ct.2019.2137
4. Cheung PC, Cunningham SA, Narayan KMV, Kramer MR. Childhood Obesity Incidence in the United States: A Systematic Review. *Child Obes* (2016) 12:1–11. doi: 10.1089/chi.2015.0055
5. Monasta L, Batty GD, Cattaneo A, Lutje V, Ronfani L, Van Lenthe FJ, et al. Early-Life Determinants of Overweight and Obesity: A Review of Systematic Reviews. *Obes Rev* (2010) 11:695–708. doi: 10.1111/j.1467-789X.2010.00735.x
6. Moore BF, Starling AP, Magzamen S, Harrod CS, Allshouse WB, Adgate JL, et al. Fetal Exposure to Maternal Active and Second-Hand Smoking With Offspring Early-Life Growth in the Healthy Start Study. *Int J Obes (2005)* (2019) 43:652–62. doi: 10.1038/s41366-018-0238-3
7. Aurrekoetxea JJ, Murcia M, Rebagliato M, Lopez MJ, Castilla AM, Santa-Marina L, et al. Determinants of Self-Reported Smoking and Misclassification During Pregnancy, and Analysis of Optimal Cut-Off Points for Urinary Cotinine: A Cross-Sectional Study. *BMJ Open* (2013) 3:e002034. doi: 10.1136/bmjopen-2012-002034
8. Riedel C, Schönberger K, Yang S, Koshy G, Chen YC, Gopinath B, et al. Parental Smoking and Childhood Obesity: Higher Effect Estimates for Maternal Smoking in Pregnancy Compared With Paternal Smoking–a Meta-Analysis. *Int J Epidemiol* (2014) 43:1593–606. doi: 10.1093/ije/dyu150
9. de Wit CC, Sas TC, Wit JM, Cutfield WS. Patterns of Catch-Up Growth. *J Pediatr* (2013) 162:415–20. doi: 10.1016/j.jpeds.2012.10.014
10. Raghuveer G, White David A, Hayman Laura L, Woo Jessica G, Villafane J, Celermajer D, et al. Cardiovascular Consequences of Childhood Second-Hand Tobacco Smoke Exposure: Prevailing Evidence, Burden, and Racial and Socioeconomic Disparities: A Scientific Statement From the American Heart Association. *Circulation* (2016) 134:e336–e59. doi: 10.1161/CIR.0000000000000443
11. Lindström L, Wikström AK, Bergman E, Mulic-Lutvica A, Högberg U, Ahlsson F, et al. Postnatal Growth in Children Born Small for Gestational Age With and Without Smoking Mother. *Pediatr Res* (2019) 85:961–6. doi: 10.1038/s41390-019-0352-5
12. Potvin S, Tikàsz A, Dinh-Williams LL, Bourque J, Mendrek A. Cigarette Cravings, Impulsivity, and the Brain. *Front Psychiatry* (2015) 6:125. doi: 10.3389/fpsyt.2015.00125
13. Stojakovic A, Espinosa EP, Farhad OT, Lutfy K. Effects of Nicotine on Homeostatic and Hedonic Components of Food Intake. *J Endocrinol* (2017) 235:R13–31. doi: 10.1530/joe-17-0166
14. Argalasova L, Zitnanova I, Vondrova D, Dvorakova M, Laubertova L, Jurkovicova J, et al. Self-Reported Exposure to ETS (Environmental Tobacco Smoke), Urinary Cotinine, and Oxidative Stress Parameters in Pregnant Women-the Pilot Study. *Int J Environ Res Public Health* (2019) 16:1656. doi: 10.3390/ijerph16091656
15. Kawamoto T, Nitta H, Murata K, Toda E, Tsukamoto N, Hasegawa M, et al. Rationale and Study Design of the Japan Environment and Children's Study (JECS). *BMC Public Health* (2014) 14:25. doi: 10.1186/1471-2458-14-25
16. Nishihama Y, Nakayama SF, Tabuchi T, Isobe T, Jung C-R, Iwai-Shimada M, et al. Determination of Urinary Cotinine Cut-Off Concentrations for Pregnant Women in the Japan Environment and Children's Study (JECS). *Int J Environ Res Public Health* (2020) 17:5537. doi: 10.3390/ijerph17155537
17. Callo Quinte G, Barros F, Gigante DP, de Oliveira IO, Dos Santos Motta JV, Horta BL. Overweight Trajectory and Cardio Metabolic Risk Factors in Young Adults. *BMC Pediatr* (2019) 19:75. doi: 10.1186/s12887-019-1445-3
18. Stein AD, Obrutu OE, Behere RV, Yajnik CS. Developmental Undernutrition, Offspring Obesity and Type 2 Diabetes. *Diabetologia* (2019) 62:1773–8. doi: 10.1007/s00125-019-4930-1
19. Huang MY, Wang MY, Lin YS, Lin CJ, Lo K, Chang IJ, et al. The Association Between Metabolically Healthy Obesity, Cardiovascular Disease, and All-Cause Mortality Risk in Asia: A Systematic Review and Meta-Analysis. *Int J Environ Res Public Health* (2020) 17:1320. doi: 10.3390/ijerph17041320
20. Kobayashi S, Sata F, Hanaoka T, Braimoh TS, Ito K, Tamura N, et al. Association Between Maternal Passive Smoking and Increased Risk of Delivering Small-for-Gestational-Age Infants at Full-Term Using Plasma Cotinine Levels From The Hokkaido Study: A Prospective Birth Cohort. *BMJ Open* (2019) 9:e023200. doi: 10.1136/bmjopen-2018-023200
21. Griffin IJ. Catch-Up Growth: Basic Mechanisms. *Nestle Nutr Inst Workshop Ser* (2015) 81:87–97. doi: 10.1159/000365806
22. Finkielstain GP, Lui JC, Baron J. Catch-Up Growth: Cellular and Molecular Mechanisms. *World Rev Nutr Diet* (2013) 106:100–4. doi: 10.1159/000342535

23. Aris IM, Bernard JY, Chen LW, Tint MT, Pang WW, Lim WY, et al. Infant Body Mass Index Peak and Early Childhood Cardio-Metabolic Risk Markers in a Multi-Ethnic Asian Birth Cohort. *Int J Epidemiol* (2017) 46:513–25. doi: 10.1093/ije/dyw232
24. Chen LW, Aris IM, Bernard JY, Tint MT, Colega M, Gluckman PD, et al. Associations of Maternal Macronutrient Intake During Pregnancy With Infant BMI Peak Characteristics and Childhood BMI. *Am J Clin Nutr* (2017) 105:705–13. doi: 10.3945/ajcn.116.148270
25. DiLeone RJ, Taylor JR, Picciotto MR. The Drive to Eat: Comparisons and Distinctions Between Mechanisms of Food Reward and Drug Addiction. *Nat Neurosci* (2012) 15:1330–5. doi: 10.1038/nn.3202
26. Masuzaki H, Kozuka C, Okamoto S, Yonamine M, Tanaka H, Shimabukuro M. Brown Rice-Specific γ-Oryzanol as a Promising Prophylactic Avenue to Protect Against Diabetes Mellitus and Obesity in Humans. *J Diabetes Investig* (2019) 10:18–25. doi: 10.1111/jdi.12892
27. Richardson SA, Tizabi Y. Hyperactivity in the Offspring of Nicotine-Treated Rats: Role of the Mesolimbic and Nigrostriatal Dopaminergic Pathways. *Pharmacol Biochem Behav* (1994) 47:331–7. doi: 10.1016/0091-3057(94)90018-3
28. Romoli B, Lozada AF, Sandoval IM, Manfredsson FP, Hnasko TS, Berg DK, et al. Neonatal Nicotine Exposure Primes Midbrain Neurons to a Dopaminergic Phenotype and Increases Adult Drug Consumption. *Biol Psychiatry* (2019) 86:344–55. doi: 10.1016/j.biopsych.2019.04.019
29. Thomas AM, Ostroumov A, Kimmey BA, Taormina MB, Holden WM, Kim K, et al. Adolescent Nicotine Exposure Alters GABA(A) Receptor Signaling in the Ventral Tegmental Area and Increases Adult Ethanol Self-Administration. *Cell Rep* (2018) 23:68–77. doi: 10.1016/j.celrep.2018.03.030
30. Torres S, Merino C, Paton B, Correig X, Ramírez N. Biomarkers of Exposure to Second-Hand and Thirdhand Tobacco Smoke: Recent Advances and Future Perspectives. *Int J Environ Res Public Health* (2018) 15:2693. doi: 10.3390/ijerph15122693
31. Becker PJ, Nieman Carney L, Corkins MR, Monczka J, Smith E, Smith SE, et al. Consensus Statement of the Academy of Nutrition and Dietetics/ American Society for Parenteral and Enteral Nutrition: Indicators Recommended for the Identification and Documentation of Pediatric Malnutrition (Undernutrition). *J Acad Nutr Diet* (2014) 114:1988–2000. doi: 10.1016/j.jand.2014.08.026
32. Becker PJ, Gunnell Bellini S, Wong Vega M, Corkins MR, Spear BA, Spoede E, et al. Validity and Reliability of Pediatric Nutrition Screening Tools for Hospital, Outpatient, and Community Settings: A 2018 Evidence Analysis Center Systematic Review. *J Acad Nutr Diet* (2020) 120:288–318.e2. doi: 10.1016/j.jand.2019.06.257
33. World Health Organization. *WHO Child Growth Standards : Length/Height-for-Age, Weight-for-Age, Weight-for-Length, Weight -for-Height and Body Mass Index-for-Age: Methods and Development*. Geneva: World Health Organization (2006). Available at: https://apps.who.int/iris/handle/10665/43413.

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers | Frontiers in Endocrinology

ORIGINAL RESEARCH
published: 16 May 2022
doi: 10.3389/fendo.2022.881093

Association Between Fat Mass or Fat Fibrotic Gene Expression and Polyneuropathy in Subjects With Obesity: A Korean Metabolic Bariatric Surgery Cohort

Kyuho Kim[1], Tae Jung Oh[1,2], Young Suk Park[3], Won Chang[4], Hyen Chung Cho[1], Jihye Lee[1], Yun Kyung Lee[1], Sung Hee Choi[1,2] and Hak Chul Jang[1,2]*

[1] Department of Internal Medicine, Seoul National University Bundang Hospital, Seongnam, South Korea, [2] Department of Internal Medicine, Seoul National University College of Medicine, Seoul, South Korea, [3] Department of Surgery, Seoul National University Bundang Hospital, Seongnam, South Korea, [4] Department of Radiology, Seoul National University Bundang Hospital, Seongnam, South Korea

OPEN ACCESS

Edited by:
Mostafa Qorbani, Alborz University of Medical Sciences, Iran

Reviewed by:
Solaleh Emamgholipour, Tehran University of Medical Sciences, Iran
Fatemeh Bandarian, Tehran University of Medical Sciences, Iran

***Correspondence:**
Tae Jung Oh
ohtjmd@gmail.com

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *22 February 2022*
Accepted: *28 March 2022*
Published: *16 May 2022*

Citation:
Kim K, Oh TJ, Park YS, Chang W, Cho HC, Lee J, Lee YK, Choi SH and Jang HC (2022) Association Between Fat Mass or Fat Fibrotic Gene Expression and Polyneuropathy in Subjects With Obesity: A Korean Metabolic Bariatric Surgery Cohort. Front. Endocrinol. 13:881093. doi: 10.3389/fendo.2022.881093

Aim: We aimed to investigate the association between obesity-related parameters and polyneuropathy (PN) and to evaluate inflammatory and fibrotic gene expression of fat as a potential mediator in subjects scheduled to undergo metabolic bariatric surgery (MBS).

Methods: This was a cross-sectional study of MBS cohort. Body composition and visceral fat area (VFA) were quantified by bioimpedance analysis and computed tomography scan. PN was defined by Michigan Neuropathy Screening Instrument–Physical Examination score was > 2. We measured mRNA expression level of *FN1*, *TIMP1*, *CCL2*, and *CXCL8* in omental fat tissue.

Results: Of 189 subjects (mean age, 39.4 years; 69 [36.5%] male; mean body mass index, 38.5 kg/m^2), prevalence of PN was 9.1% in subjects without diabetes (n = 110) and 20.3% in those with diabetes (n = 79). Nondiabetic subjects with PN had higher homeostatic model assessment-insulin resistance (6.8 ± 3.5 *vs* 4.5 ± 2.8, p = 0.041), and increased fat mass (58.5 ± 12.5 kg *vs* 50.5 ± 10.7 kg, p = 0.034), and VFA (309.4 ± 117.6 cm^2 *vs* 243.5 ± 94.2 cm^2, p = 0.046) compared to those without PN. These obesity-related parameters were significantly associated with the presence of PN after adjusting for conventional risk factors of PN only in subjects without diabetes. In contrast, a fibrotic gene such as *TIMP1* was independently associated with PN (adjusted odds ratio of 1.56; 95% confidence interval 1.06, 2.30) only in subjects with diabetes.

Conclusion: Increased adiposity was independently associated with PN in obese subjects without diabetes. In contrast, this association was not significant after adjusting conventional risk factors of PN in obese subjects with diabetes but increased fibrotic gene expression in fat was associated with PN in this group.

Keywords: fibrosis, insulin resistance, obesity, polyneuropathy, bariatric surgery

INTRODUCTION

Polyneuropathy (PN), one of the most common types of peripheral neuropathy, is prevalent in subjects with diabetes and even in those with prediabetes. While increased duration of diabetes and poor glycemic control are established risk factors for diabetic PN (1, 2), intensive glycemic control shows limited efficacy in prevention of PN in type 2 diabetes (3). This suggests that risk factors other than glucose, such as the components of metabolic syndrome, might contribute to nerve damage to a considerable extent.

The Cooperative Health Research in the Region of Augsburg (KORA) study (4) and the Anglo–Danish–Dutch study of Intensive Treatment in People with Screen-Detected Diabetes in Primary Care (ADDITION) study (5) revealed an association between general and abdominal obesity (weight and waist circumference) and development of PN. Besides the amount of fat, the quality of the fat tissue could induce detrimental effects on the metabolic milieu. Considering that chronic adipose tissue inflammation was associated with obesity-related metabolic complications (6) and adipose tissue is the main site where systemic inflammation begins (7), it is logically reasonable to investigate an association of the expression level of inflammatory and fibrosis genes in fat tissue with PN.

Meanwhile, a previous observational study showed that 11.1% of obese subjects already had PN despite normoglycemia, and that the presence of PN is positively associated with waist circumference (8). This suggests that the association between obesity and PN needs to be analyzed in a subgroup stratified by diabetes status.

Therefore, in this study, we aimed to investigate the association between obesity-related parameters and PN in obese subjects stratified by diabetes status. We used computed tomography scan to assess the quantity of VAT, and analyzed mRNA levels of inflammatory and fibrosis genes such as *FN1* (9), *TIMP1* (10), *CCL2* (11), and *CXCL8* (12) from omental fat tissue. These genes have been studies as a driver of systemic inflammation and insulin resistance.

METHODS

Population

We recruited subjects scheduled to undergo metabolic bariatric surgery (MBS) and intensively evaluated metabolic parameters and the presence of diabetic vascular complications. A total of 205 obese subjects were enrolled from Seoul National University Bundang Hospital (SNUBH), a tertiary academic hospital from April 2019 to December 2020, the aim of original prospective observational study is to discover predictive markers for weight loss and metabolic improvement after MBS. The original cohort study has been registered at Clinical research Information Service (CRIS Registration No. KCT0005777). Inclusion criteria were age ≥ 20 years old and a body mass index (BMI) ≥ 35 kg/m^2 with no comorbidity; BMI ≥ 30 kg/m^2 with at least one comorbidity; or BMI ≥ 27.5 kg/m^2 with medically uncontrolled type 2 diabetes. Type 2 diabetes was defined according to the criteria of American Diabetes Association: fasting plasma glucose (FPG) ≥ 7.0 mmol/l or HbA_{1c} ≥ 6.5% (48 mmol/mol) (13). Subjects on diabetes medications were also considered to have diabetes. We excluded the subjects who did not meet the indication of MBS according to the local guideline (14). We also excluded the subjects who previously underwent MBS. This study was a cross-sectional study analyzing data of 189 participants (**Figure 1**) after excluding subjects with missing information of PN (n = 16). We applied 2:1 propensity score matching using age, sex, and BMI to compare mRNA expression levels in omental fat tissue between subjects without PN and those with PN. However, values were excluded in analysis if quality of cDNA sample was unacceptable. The study was approved by the Institutional Review Board of SNUBH (no. B-2111-718-301), and each participant provided written informed consent. The study was conducted in accordance with the Declaration of Helsinki.

Anthropometric and Biochemical Analyses

Anthropometric indices were measured by a well-trained research nurse. BMI was calculated as weight (kg) divided by the square of the height in meters. Waist circumference was measured at the midpoint between the margin of the lowest rib and the iliac crest. Systolic blood pressure (BP) and diastolic BP were measured by an electronic BP monitor after 10 minutes of rest in a sitting position. Smoking status was classified as never smoker (< 100 cigarettes in a lifetime and currently a nonsmoker), ex-smoker (≥ 100 cigarettes in a lifetime and currently a nonsmoker), and current smoker (≥ 100 cigarettes in a lifetime and currently a smoker). We defined drinkers as those who drank any alcoholic beverage more than once a month. Positive exercise was defined as exercising for > 150 min/week. Blood samples were collected after an overnight fast. FPG levels were measured by the hexokinase method, and glycated hemoglobin (HbA_{1c}) levels were measured by high-performance liquid chromatography (Bio-Rad, Hercules, CA, USA). Serum insulin levels were measured by immunoradiometric assay (DIAsource, Nivelles, Belgium). Total cholesterol, triglyceride, high-density lipoprotein (HDL) cholesterol, and low-density lipoprotein (LDL) cholesterol were measured by enzymatic colorimetric assay. Creatinine was measured by the protocol of the central laboratory of SNUBH, and estimated glomerular filtration rate (eGFR) was calculated by the Modification of Diet in Renal Disease equation (15). Homeostasis model assessment-insulin resistance (HOMA-IR) was calculated using the following

Abbreviations: ADDITION, Anglo–Danish–Dutch study of Intensive Treatment in People with Screen-Detected Diabetes in Primary Care; BMI, Body mass index; BP, Blood pressure; CI, confidence interval; CT, Computed tomography; DNER, Delta/Notch-like epidermal growth factor-related receptor; eGFR, Estimated glomerular filtration rate; FPG, Fasting plasma glucose; $HbA1_c$, Glycated hemoglobin; HDL, High-density lipoprotein; HOMA-IR, Homeostasis model assessment-insulin resistance; KORA, Cooperative Health Research in the Region of Augsburg; LBM, Lean body mass; LDL, Low-density lipoprotein; MBS, Metabolic bariatric surgery; MNSI-PE, Michigan Neuropathy Screening Instrument-Physical Examination; MNSI-Q, Michigan Neuropathy Screening Instrument-Questionnaire; OR, odds ratio; PN, Polyneuropathy; SAT, Subcutaneous adipose tissue; SNUBH, Seoul National University Bundang Hospital; VAT, Visceral adipose tissue; VFA, Visceral fat area.

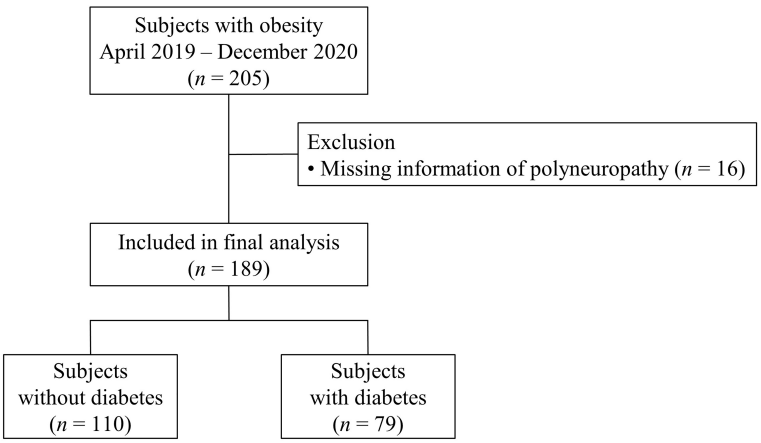


FIGURE 1 | Flow chart of the selection of subjects for the analysis. Among 205 obese subjects, after excluding subjects with missing information of polyneuropathy, 189 subjects were included in the final analysis. They were divided into two groups according to diabetes status.

formula: HOMA-IR = (fasting insulin [μU/ml] × FPG [mmol/l]/ 22.5) (16).

Body Composition Measurements

Body fat mass, fat percent, and lean body mass (LBM) were estimated by a bioimpedance analysis (InBody770, InBody, Seoul, Korea). Regarding the visceral fat area (VFA), cross-sectional abdominal computed tomography (CT) images at the level of the third lumbar vertebral body (L3) were acquired, and VFA was calculated from areas within a range of –150 to –50 Hounsfield units (17).

Assessment of PN

We used the Michigan Neuropathy Screening Instrument (MNSI), which includes two separate assessments, a 15-item self-administered questionnaire (MNSI-Q) and a lower-extremity physical examination (MNSI-PE) (18). PN was diagnosed when the MNSI-PE score was > 2. A trained nurse performed all neurologic examinations.

Measurement of Inflammatory and Fibrosis Markers

Omental fat tissue was obtained during MBS and stored at –80°C. Total RNA was extracted from frozen human fat tissue samples using TRIzol (Thermo Fisher Scientific, Waltham, MA, USA). For quantitative real-time PCR analysis, 1 μg of total RNA was reverse-transcribed using the High-Capacity cDNA Reverse Transcription kit (Thermo Fisher Scientific, Waltham, MA, USA). SYBR Green reactions using the SYBR Green PCR Master mix (Enzynomics, Daejeon, Korea) were assembled along with primers according to the manufacturer's instructions and were performed using the QuantStudio 7 Flex Real-Time PCR System (Thermo Fisher Scientific, Waltham, MA, USA). Relative mRNA levels were calculated using the comparative threshold cycle method and normalized to *cyclophilin* mRNA. All primers used are listed with their sequences in **Supplementary Table 1**.

Statistical Analysis

Data were expressed as the mean ± standard deviation or number (%). For checking normality of distribution of variables, Kolmogorov-Smirnov and Shapiro-Wilk tests were used. Categorical variables were compared using χ^2 tests, and continuous variables were compared using Student's unpaired *t* tests for parametric data or Mann–Whitney U tests for nonparametric data. Since variables were not normally distributed, Spearman's correlation coefficient was used to evaluate the correlation between variables. Univariable and multivariable logistic regression models were used to analyze the associations between obesity-related parameters and PN. In all cases, $p < 0.05$ was considered statistically significant. Statistical analyses were performed using IBM SPSS version 25.0 (IBM Inc., Armonk, NY, USA). Figures were drawn using GraphPad Prism software (version 9.1.2; GraphPad Software Inc., CA, USA).

RESULTS

Among 189 subjects who were candidates for MBS, 79 (41.8%) had diabetes. Prevalence of PN was 9.1% in subjects without diabetes and 20.3% in those with diabetes. Among subjects without diabetes, insulin and HOMA-IR were higher in subjects with PN compared with those without PN. Among subjects with diabetes, body weight, BMI, waist circumference, eGFR, and the proportion of subjects taking insulin therapy were higher in subjects with PN compared with those without PN. However, diabetes duration, FPG, and HbA_{1c} were comparable between subjects with PN and those without PN (**Table 1**). Body composition analysis showed that fat mass and VFA were higher in subjects with PN compared with those without PN, irrespective of diabetes status. Among subjects with diabetes,

TABLE 1 | Baseline characteristics of obese subjects stratified by diabetes and polyneuropathy.

Variable	Diabetes (–) (*n* = 110)		*p* value	Diabetes (+) (*n* = 79)		*p* value
	PN (–) (*n* = 100)	PN (+) (*n* = 10)		PN (–) (*n* = 63)	PN (+) (*n* = 16)	
Male, *n* (%)	34 (34.0)	3 (30.0)	1.000	24 (38.1)	8 (50.0)	0.386
Age (years)	36.5 ± 9.3	38.3 ± 15.9	0.983	42.6 ± 10.6	45.1 ± 12.7	0.490
Body weight (kg)	107.8 ± 20.2	121.2 ± 28.4	0.154	101.6 ± 23.5	118.7 ± 22.7	0.003
BMI (kg/m^2)	38.7 ± 5.2	42.1 ± 7.7	0.206	36.8 ± 5.9	42.3 ± 7.5	0.005
Obesity classes, *n* (%)			0.496			0.144
Class I (BMI 25–29.9)	1 (1.0)	0 (0.0)		5 (7.9)	0 (0.0)	
Class II (BMI 30–34.9)	26 (26.0)	1 (10.0)		23 (36.5)	3 (18.8)	
Class III (BMI ≥ 35)	73 (73.0)	9 (90.0)		35 (55.6)	13 (81.3)	
Waist circumference (cm)	115.2 ± 12.3	117.8 ± 11.0	0.571	113.2 ± 14.4	124.7 ± 15.9	0.007
Systolic BP (mmHg)	134.0 ± 15.8	134.2 ± 13.6	0.743	136.3 ± 19.0	145.1 ± 15.7	0.089
Diastolic BP (mmHg)	80.4 ± 12.0	78.3 ± 10.8	0.685	83.0 ± 13.4	80.3 ± 15.2	0.526
Diabetes duration (years)	NA	NA	NA	4.5 ± 5.3	10.0 ± 10.8	0.074
FPG (mmol/l)	5.5 ± 0.6	5.5 ± 0.5	0.720	8.5 ± 3.1	8.9 ± 3.0	0.522
HbA_{1c} (%)	5.5 ± 0.4	5.6 ± 0.7	0.707	7.8 ± 1.8	7.3 ± 1.3	0.490
HbA_{1c} (mmol/mol)	36.5 ± 4.0	37.1 ± 7.7	0.707	61.7 ± 19.5	56.4 ± 14.4	0.490
Triglyceride (mmol/l)	1.8 ± 1.5	1.9 ± 0.9	0.491	2.1 ± 1.4	2.1 ± 0.9	0.357
HDL cholesterol (mmol/l)	1.4 ± 0.3	1.2 ± 0.2	0.091	1.3 ± 0.2	1.3 ± 0.2	0.877
LDL cholesterol (mmol/l)	3.4 ± 0.6	3.2 ± 0.4	0.574	2.9 ± 0.9	3.0 ± 0.7	0.353
eGFR (mL min^{-1} [1.73 m]^2)	115.6 ± 24.0	124.5 ± 34.6	0.743	110.8 ± 24.8	94.7 ± 28.6	0.025
Insulin (pmol/l)	126.5 ± 72.8	190.2 ± 94.7	0.042	126.0 ± 91.8	136.0 ± 100.6	0.829
HOMA-IR	4.5 ± 2.8	6.8 ± 3.5	0.041	6.6 ± 4.4	7.0 ± 4.7	0.830
10-g monofilament, Right	9.8 ± 0.5	9.4 ± 0.7	0.013	9.6 ± 0.6	9.3 ± 0.9	0.148
10-g monofilament, Left	9.8 ± 0.5	9.3 ± 0.8	0.012	9.7 ± 0.6	9.4 ± 0.7	0.198
MNSI-Q	2.2 ± 1.8	3.3 ± 2.1	0.076	3.2 ± 2.1	4.6 ± 2.0	0.019
MNSI-PE	0.4 ± 0.7	3.3 ± 0.5	<0.001	0.6 ± 0.7	3.2 ± 0.5	<0.001
Smoking status			0.768			0.201
Never smoker, *n* (%)	67 (67.0)	6 (60.0)		27 (42.9)	10 (62.5)	
Ex-smoker, *n* (%)	12 (12.0)	2 (20.0)		14 (22.2)	4 (25.0)	
Current smoker, *n* (%)	21 (21.0)	2 (20.0)		22 (34.9)	2 (12.5)	
Alcohol, *n* (%)	74 (74.0)	7 (70.0)	0.784	38 (60.3)	10 (62.5)	0.873
Exercise, *n* (%)	43 (43.0)	4 (40.0)	0.834	30 (47.6)	3 (18.8)	0.048
Hypertension, *n* (%)	36 (36.0)	5 (50.0)	0.383	39 (61.9)	12 (75.0)	0.328
Dyslipidaemia, *n* (%)	37 (37.0)	4 (40.0)	0.852	45 (71.4)	10 (62.5)	0.488
Insulin therapy, *n* (%)	NA	NA	NA	10 (15.9)	7 (43.8)	0.015
Fat mass (kg)	50.5 ± 10.7	58.4 ± 12.5	0.034	44.2 ± 13.8	54.0 ± 16.4	0.028
Fat percent (%)	46.1 ± 6.5	48.5 ± 4.0	0.165	43.2 ± 6.3	44.9 ± 7.6	0.390
LBM (kg)	58.0 ± 12.8	62.9 ± 17.1	0.431	57.1 ± 13.1	64.6 ± 12.2	0.012
VFA (cm^2)	243.5 ± 94.2	309.4 ± 117.6	0.046	271.7 ± 78.7	334.2 ± 113.4	0.020

Data are expressed as the mean ± standard deviation or number (%).

BMI, body mass index; BP, blood pressure; eGFR, estimated glomerular filtration rate; FPG, fasting plasma glucose; HbA_{1c}, glycated haemoglobin; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment-insulin resistance; LBM, lean body mass; LDL, low-density lipoprotein; MNSI-PE, Michigan Neuropathy Screening Instrument-Physical Examination; MNSI-Q, Michigan Neuropathy Screening Instrument-Questionnaire; NA, not applicable; PN, polyneuropathy; VFA, visceral fat area.

LBM was higher in subjects with PN compared with those without PN (**Table 1**).

Among subjects without diabetes, unadjusted odds ratio (OR) (95% confidence interval [CI]) for PN was 1.23 (1.02, 1.49) for HOMA-IR and 1.06 (1.00, 1.12) for fat mass. Among subjects with diabetes, unadjusted OR (95% CI) for PN was 1.05 (1.01, 1.09) for waist circumference, 1.13 (1.03, 1.23) for BMI, 1.04 (1.01, 1.08) for fat mass, and 1.01 (1.00, 1.02) for VFA. After adjustment of known risk factors for PN (19), adjusted OR (95% CI) for PN was 1.47 (1.10, 1.95) for HOMA-IR, 1.08 (1.00, 1.16) for fat mass, and 1.01 (1.00, 1.02) for VFA in subjects without diabetes. In contrast, waist circumference, BMI, fat mass, and VFA showed no significant association with PN in subjects with diabetes after adjustment of covariates (**Table 2**).

Among subjects without diabetes, any mRNA expression of omental fat was not different according to the presence of PN. In contrast, among subjects with diabetes, mRNA expression of *TIMP1* and *CXCL8* was significantly higher in subjects with PN (**Figure 2**), and positively correlated with MNSI-PE scores (*rho* = 0.469, *p* = 0.001; *rho* = 0.454, *p* = 0.002) (**Figure 3**). In addition, adjusted OR (95% CI) for PN was 1.56 (1.06, 2.30) for *TIMP1* in subjects with diabetes (**Table 2**).

DISCUSSION

In this study, we found increased fat mass and VFA, and higher HOMA-IR in subjects with PN compared with those without PN among subjects without diabetes. These variables were still significantly associated with PN even after adjusting for known risk factors for PN in this group. On the other hand, among subjects

TABLE 2 | ORs and 95% CIs for polyneuropathy stratified by diabetes.

Variable	Diabetes (-) (*n* = 110)				Diabetes (+) (*n* = 79)			
	Unadjusted OR (95% CI)	*p* value	Adjusted OR (95% CI)	*p* value	Unadjusted OR (95% CI)	*p* value	Adjusted OR (95% CI)	*p* value
Age	1.02 (0.96–1.09)	0.585	–	–	1.02 (0.97–1.07)	0.415	–	–
Sex								
Female	1.20 (0.29–4.95)	0.799	–	–	0.62 (0.20–1.86)	0.389	–	–
Male (Reference)	1	1	–	–	1	1	–	–
BMI	1.11 (0.99–1.23)	0.067	1.12 (0.98–1.28)	0.095	1.13 (1.03–1.23)	0.006	1.12 (0.99–1.27)	0.066
Waist circumference	1.02 (0.96–1.08)	0.547	1.04 (0.96–1.12)	0.372	1.05 (1.01–1.09)	0.014	1.04 (0.98–1.09)	0.180
Systolic BP	1.00 (0.96–1.04)	0.966	–	–	1.03 (1.00–1.06)	0.096	–	–
Diabetes duration	–	–	–	–	1.01 (1.02–1.18)	0.012	–	–
HbA_{1c}	1.49 (0.29–7.59)	0.634	–	–	0.82 (0.56–1.20)	0.303	–	–
Triglyceride	1.00 (1.00–1.01)	0.878	–	–	1.00 (1.00–1.01)	0.937	–	–
HDL cholesterol	0.95 (0.88–1.01)	0.114	–	–	1.00 (0.94–1.06)	0.917	–	–
HOMA-IR	1.23 (1.02–1.49)	0.031	1.47 (1.10–1.95)	0.009	1.02 (0.90–1.16)	0.751	1.06 (0.90–1.25)	0.521
Smoking status								
Current smoker	0.94 (0.19–4.76)	0.941	–	–	0.27 (0.06–1.28)	0.098	–	–
Ex-smoker	1.86 (0.33–10.33)	0.478	–	–	0.77 (0.21–2.91)	0.702	–	–
Never smoker (Reference)	1	1	–	–	1	1	–	–
Alcohol status								
Drinker	0.82 (0.20–3.41)	0.785	–	–	1.10 (0.35–3.40)	0.873	–	–
Non-drinker (Reference)	1	1	–	–	1	1	–	–
Exercise	0.87 (0.23–3.27)	0.835	–	–	0.25 (0.07–0.98)	0.046	–	–
Fat mass	1.06 (1.00–1.12)	0.040	1.08 (1.01–1.16)	0.034	1.04 (1.01–1.08)	0.025	1.04 (0.99–1.10)	0.137
VFA	1.01 (1.00–1.01)	0.052	1.01 (1.00–1.02)	0.031	1.01 (1.00–1.02)	0.021	1.00 (1.00–1.01)	0.440
TIMP1[a]	1.13 (0.82–1.57)	0.459	1.41 (0.77–2.56)	0.262	1.34 (1.09–1.66)	0.007	1.51 (1.02–2.23)	0.040
CXCL8[b]	0.97 (0.85–1.11)	0.705	1.14 (0.88–1.47)	0.317	1.68 (1.04–2.70)	0.033	1.90 (0.92–3.92)	0.081

Data are presented as ORs and 95% CIs.

Adjusted analysis: adjustment for age, sex, systolic blood pressure, glycated haemoglobin (HbA_{1c}), triglyceride, HDL cholesterol, smoking status, alcohol status, and exercise (+ diabetes duration in diabetes (+) group). In diabetes (+) group, for TIMP1 and CXCL8, VFA was adjusted additionally.

[a]*Diabetes (–) (n = 26), Diabetes (+) (n = 47).*

[b]*Diabetes (–) (n = 27), Diabetes (+) (n = 46).*

BMI, body mass index; BP, blood pressure; CI, confidence interval; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment-insulin resistance; OR, odds ratio; VFA, visceral fat area.

with diabetes we found no difference in HOMA-IR between subjects with or without PN, and the association of fat mass and VFA with PN was not significant after adjusting covariates. In contrast to these insignificant associations, *TIMP1* gene expression as a marker of fat fibrosis was significantly higher in the presence of PN in the diabetes subgroup.

Hyperglycemia is a major risk factor for PN (19), and glucose control is the mainstay of prevention and management of PN (20). However, enhanced glucose control modestly reduced (21), or did not reduce the incidence of PN in type 2 diabetes (22, 23). Therefore, it is necessary to identify other risk factors for PN in subjects with type 2 diabetes or at high risk for type 2 diabetes. From the data of obese subjects without diabetes, we confirmed that obesity is the main metabolic driver of PN. Furthermore, insulin resistance assessed by HOMA-IR was also an independent risk factor for PN in this population. Considering that insulin is a neurotrophic factor responsible for neuronal growth, survival, and differentiation (24, 25), it is possible that disruption of insulin signaling due to insulin resistance contributed to the pathogenesis of PN. However, the association between HOMA-IR and PN was not significant in subjects with overt diabetes, a finding that might be due to the strong influence of severe insulin resistance in these subjects who were candidates for MBS. Therefore, the impact of obesity on PN might depend on whether the severity of insulin resistance of subjects caused diabetes.

A prospective study of the general population with and without diabetes from the KORA cohort showed that serum levels of CCL7, CXCL10, and DNER partly mediated the association between obesity and PN (4). Another prospective study of subjects with diabetes showed an association of plasma levels of TNF and ICAM1 with development of PN (26). A cross-sectional study showed associations of plasma levels of MMP9 and TIMP1 with PN in type 1 diabetes (27). Considering that adipose tissue is the main site where systemic inflammation begins (7), it is reasonable to investigate an association of the expression level of inflammatory and fibrosis genes in fat tissue with PN. In our analysis, including subjects with diabetes, mRNA levels of *TIMP1* and *CXCL8* were significantly higher in subjects with PN compared with their counterparts. These results suggest the possibility that inflammatory cytokines released from VAT during tissue inflammation and fibrosis contribute to the pathogenesis of PN in diabetes. In contrast, no significant association between inflammatory gene expression in the omental fat and PN was found in the nondiabetic subgroup. Therefore, inflammation or fibrosis of fat tissue might contribute

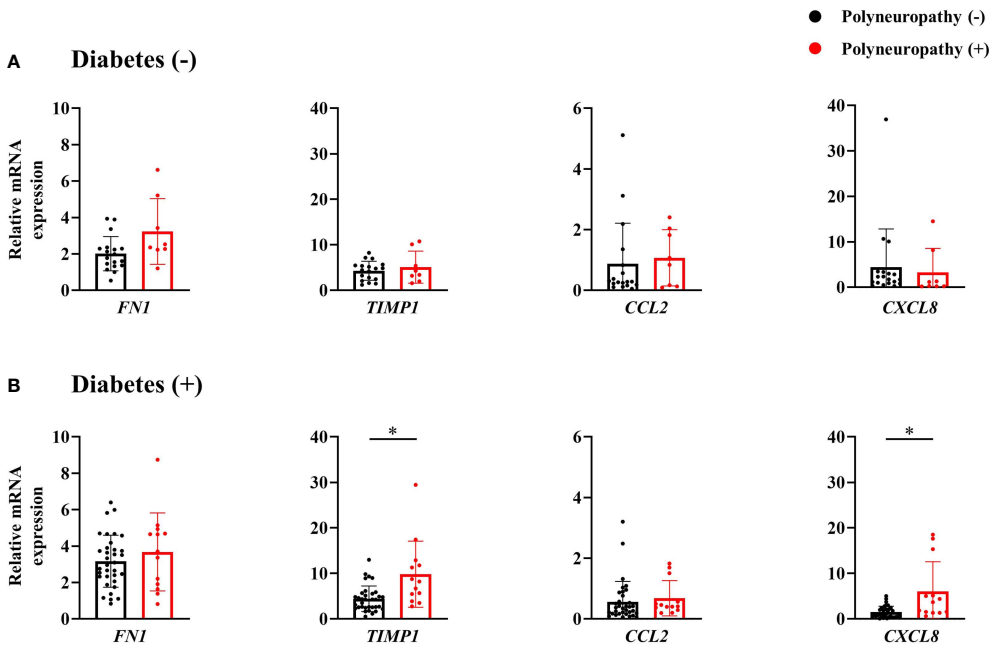


FIGURE 2 | mRNA expression of inflammatory and fibrotic genes of omental fat tissue in subjects without diabetes **(A)** and those with diabetes **(B)**. *$p < 0.05$.

to PN differently according to diabetes status, however this hypothesis needs to be investigated in further study.

The current study has a number of strengths. First, we performed analysis of PN stratified by diabetes status, thereby suggesting the step-wise contribution of hyperglycemia and obesity involved in the pathogenesis of PN. Second, VFA was measured by CT scan, the gold standard method. Finally, we directly measured gene expression from omental fat tissue. To the best of our knowledge, this study is the first analysis combining gene expression of fat tissue and PN. Nevertheless, this study has some limitations. First, there were no lean controls, and we analyzed the mRNA data in the subgroup due to the availability of fat tissue. Second, a neurophysiological study was not used to confirm PN. Therefore, the diagnosis of PN is not confirmative, but possible PN was adopted. Third, we cannot establish a causal relationship based on the cross-sectional study design. Fourth, because the original cohort is based on a single center in Korea, results cannot be generalized to other races or ethnicities. Finally, we did not separate subjects with prediabetes from subjects without diabetes.

In conclusion, fat mass, VFA ("quantity of fat"), and HOMA-IR were independent risk factors for PN in obese subjects without diabetes. In contrast, advanced pathology of fat tissue such as fibrosis ("quality of fat") might be a considerable

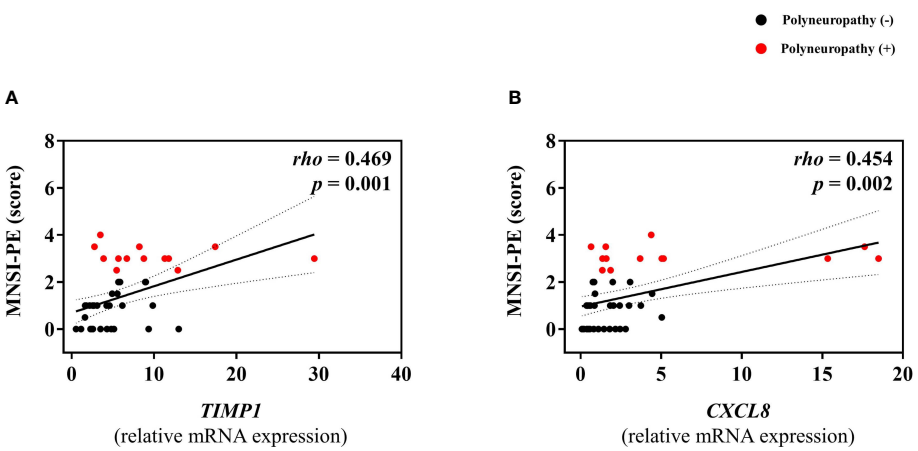


FIGURE 3 | Correlations between mRNA expression level of *TIMP1* **(A)** or *CXCL8* **(B)**, and MNSI-PE scores in subjects with diabetes.

abnormality for PN in obese subjects who already have diabetes. Therefore, the early evaluation of PN and the early management of obesity might be necessary in obese subjects at high risk of diabetes. Future studies are needed to test whether weight loss intervention can prevent and delay PN in these high-risk subjects.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/**Supplementary Material**, further inquiries can be directed to the corresponding author.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Institutional Review Board of SNUBH. The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

KK drafted the article. TJO, YSP, WC, SHC, and HCJ contributed to the conception and design of the study. KK performed the statistical analyses. KK, TJO, YSP, WC, HCC, JL, and YKL contributed to the acquisition of data. TJO is the guarantor of this work. All authors critically reviewed the manuscript and approved this version to be published.

FUNDING

This work was supported by the Korean Society for the Study of Obesity (Grant No. KSSO201904); the National Research Foundation of Korea (NRF) (Grant No. NRF-2020R1C1 C1013766); the Medical Research Center through the NRF (Grant No. NRF-2018R1A5A2024425) funded by the Korea Government; and the SNUBH Research Fund (Grant No. 13-2019-0013).

ACKNOWLEDGMENTS

The authors sincerely thank all the participants for their cooperation. In addition, we thank Ms. Chae Won Kim for assistance.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2022.881093/full#supplementary-material

REFERENCES

1. Adler AI, Boyko EJ, Ahroni JH, Stensel V, Forsberg RC, Smith DG. Risk Factors for Diabetic Peripheral Sensory Neuropathy. Results of the Seattle Prospective Diabetic Foot Study. *Diabetes Care* (1997) 20(7):1162–7. doi: 10.2337/diacare.20.7.1162
2. Yang H, Sloan G, Ye Y, Wang S, Duan B, Tesfaye S, et al. New Perspective in Diabetic Neuropathy: From the Periphery to the Brain, A Call for Early Detection, and Precision Medicine. *Front Endocrinol (Lausanne)* (2019) 10:929. doi: 10.3389/fendo.2019.00929
3. Callaghan BC, Little AA, Feldman EL, Hughes RA. Enhanced Glucose Control for Preventing and Treating Diabetic Neuropathy. *Cochrane Database Syst Rev* (2012), CD007543. doi: 10.1002/14651858.CD007543.pub2
4. Schlesinger S, Herder C, Kannenberg JM, Huth C, Carstensen-Kirberg M, Rathmann W, et al. General and Abdominal Obesity and Incident Distal Sensorimotor Polyneuropathy: Insights Into Inflammatory Biomarkers as Potential Mediators in the Kora F4/Ff4 Cohort. *Diabetes Care* (2019) 42 (2):240–7. doi: 10.2337/dc18-1842
5. Andersen ST, Witte DR, Dalsgaard EM, Andersen H, Nawroth P, Fleming T, et al. Risk Factors for Incident Diabetic Polyneuropathy in a Cohort With Screen-Detected Type 2 Diabetes Followed for 13 Years: Addition-Denmark. *Diabetes Care* (2018) 41(5):1068–75. doi: 10.2337/dc17-2062
6. Zatterale F, Longo M, Naderi J, Raciti GA, Desiderio A, Miele C, et al. Chronic Adipose Tissue Inflammation Linking Obesity to Insulin Resistance and Type 2 Diabetes. *Front Physiol* (2019) 10:1607. doi: 10.3389/fphys.2019.01607
7. Xu H, Barnes GT, Yang Q, Tan G, Yang D, Chou CJ, et al. Chronic Inflammation in Fat Plays a Crucial Role in the Development of Obesity-Related Insulin Resistance. *J Clin Invest* (2003) 112(12):1821–30. doi: 10.1172/JCI19451
8. Callaghan BC, Xia R, Reynolds E, Banerjee M, Rothberg AE, Burant CF, et al. Association Between Metabolic Syndrome Components and Polyneuropathy in an Obese Population. *JAMA Neurol* (2016) 73(12):1468–76. doi: 10.1001/jamaneurol.2016.3745
9. Chun TH. Peri-Adipocyte Ecm Remodeling in Obesity and Adipose Tissue Fibrosis. *Adipocyte* (2012) 1(2):89–95. doi: 10.4161/adip.19752
10. Meissburger B, Stachorski L, Roder E, Rudofsky G, Wolfrum C. Tissue Inhibitor of Matrix Metalloproteinase 1 (Timp1) Controls Adipogenesis in Obesity in Mice and in Humans. *Diabetologia* (2011) 54(6):1468–79. doi: 10.1007/s00125-011-2093-9
11. Dahlman I, Kaaman M, Olsson T, Tan GD, Bickerton AS, Wahlen K, et al. A Unique Role of Monocyte Chemoattractant Protein 1 Among Chemokines in Adipose Tissue of Obese Subjects. *J Clin Endocrinol Metab* (2005) 90 (10):5834–40. doi: 10.1210/jc.2005-0369
12. Kobashi C, Asamizu S, Ishiki M, Iwata M, Usui I, Yamazaki K, et al. Inhibitory Effect of Il-8 on Insulin Action in Human Adipocytes *Via* Map Kinase Pathway. *J Inflammation (Lond)* (2009) 6:25. doi: 10.1186/1476-9255-6-25
13. American Diabetes A. 2. Classification and Diagnosis of Diabetes: Standards of Medical Care in Diabetes-2021. *Diabetes Care* (2021) 44(Suppl 1):S15–33. doi: 10.2337/dc21-S002
14. Kim BY, Kang SM, Kang JH, Kang SY, Kim KK, Kim KB, et al. 2020 Korean Society for the Study of Obesity Guidelines for the Management of Obesity in Korea. *J Obes Metab Syndr* (2021) 30(2):81–92. doi: 10.7570/jomes21022
15. Levey AS, Coresh J, Greene T, Stevens LA, Zhang YL, Hendriksen S, et al. Using Standardized Serum Creatinine Values in the Modification of Diet in Renal Disease Study Equation for Estimating Glomerular Filtration Rate. *Ann Intern Med* (2006) 145(4):247–54. doi: 10.7326/0003-4819-145-4-200608150-00004
16. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis Model Assessment: Insulin Resistance and Beta-Cell Function From Fasting Plasma Glucose and Insulin Concentrations in Man. *Diabetologia* (1985) 28(7):412–9. doi: 10.1007/BF00280883

17. Lee JK, Park YS, Kim K, Oh TJ, Chang W. Comparison of Bioelectrical Impedance Analysis and Computed Tomography on Body Composition Changes Including Visceral Fat After Bariatric Surgery in Asian Patients With Obesity. *Obes Surg* (2021) 31(10):4243–50. doi: 10.1007/s11695-021-05569-6
18. Feldman EL, Stevens MJ, Thomas PK, Brown MB, Canal N, Greene DA. A Practical Two-Step Quantitative Clinical and Electrophysiological Assessment for the Diagnosis and Staging of Diabetic Neuropathy. *Diabetes Care* (1994) 17(11):1281–9. doi: 10.2337/diacare.17.11.1281
19. Papanas N, Ziegler D. Risk Factors and Comorbidities in Diabetic Neuropathy: An Update 2015. *Rev Diabetes Stud* (2015) 12(1-2):48–62. doi: 10.1900/RDS.2015.12.48
20. Pop-Busui R, Boulton AJ, Feldman EL, Bril V, Freeman R, Malik RA, et al. Diabetic Neuropathy: A Position Statement by the American Diabetes Association. *Diabetes Care* (2017) 40(1):136–54. doi: 10.2337/dc16-2042
21. Ismail-Beigi F, Craven T, Banerji MA, Basile J, Calles J, Cohen RM, et al. Effect of Intensive Treatment of Hyperglycaemia on Microvascular Outcomes in Type 2 Diabetes: An Analysis of the Accord Randomised Trial. *Lancet* (2010) 376(9739):419–30. doi: 10.1016/S0140-6736(10)60576-4
22. Charles M, Ejskjaer N, Witte DR, Borch-Johnsen K, Lauritzen T, Sandbaek A. Prevalence of Neuropathy and Peripheral Arterial Disease and the Impact of Treatment in People With Screen-Detected Type 2 Diabetes: The Addition-Denmark Study. *Diabetes Care* (2011) 34(10):2244–9. doi: 10.2337/dc11-0903
23. Duckworth W, Abraira C, Moritz T, Reda D, Emanuele N, Reaven PD, et al. Glucose Control and Vascular Complications in Veterans With Type 2 Diabetes. *N Engl J Med* (2009) 360(2):129–39. doi: 10.1056/NEJMoa0808431
24. Recio-Pinto E, Rechler MM, Ishii DN. Effects of Insulin, Insulin-Like Growth Factor-Ii, and Nerve Growth Factor on Neurite Formation and Survival in Cultured Sympathetic and Sensory Neurons. *J Neurosci* (1986) 6(5):1211–9. doi: 10.1523/JNEUROSCI.06-05-01211.1986
25. Brussee V, Cunningham FA, Zochodne DW. Direct Insulin Signaling of Neurons Reverses Diabetic Neuropathy. *Diabetes* (2004) 53(7):1824–30. doi: 10.2337/diabetes.53.7.1824
26. Zheng H, Sun W, Zhang Q, Zhang Y, Ji L, Liu X, et al. Proinflammatory Cytokines Predict the Incidence of Diabetic Peripheral Neuropathy Over 5 Years in Chinese Type 2 Diabetes Patients: A Prospective Cohort Study. *EClinicalMedicine* (2021) 31:100649. doi: 10.1016/j.eclinm.2020.100649
27. Papadopoulou-Marketou N, Whiss PA, Eriksson AC, Hyllienmark L, Papassotiriou I, Wahlberg J. Plasma Levels of Tissue Inhibitor of Metalloproteinase-1 in Patients With Type 1 Diabetes Mellitus Associate With Early Diabetic Neuropathy and Nephropathy. *Diabetes Vasc Dis Res* (2021) 18(2):14791641211002470. doi: 10.1177/14791641211002470

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

frontiers | Frontiers in Endocrinology

ORIGINAL RESEARCH
published: 24 May 2022
doi: 10.3389/fendo.2022.889785

Association Between Four Anthropometric Indexes and Metabolic Syndrome in US Adults

Yaling Li[1], *Rui Zheng*[2], *Shuting Li*[1], *Ruyi Cai*[1], *Feihua Ni*[1], *Huiyan Zheng*[1], *Ruying Hu*[1] *and Ting Sun*[1]*

[1] Department Health Management Center, the Second Affiliated Hospital, School of Medicine, Zhejiang University, Hangzhou, China, [2] Department of Critical Care Medicine, Sir Run Run Shaw Hospital, School of Medicine, Zhejiang University, Hangzhou, China

Objective: To study the association between anthropometric indexes [lipid accumulation products (LAP), visceral obesity index (VAI), triglyceride and glucose index (TyG) and waist triglyceride index (WTI)] and metabolic syndrome (MetS) in a representative sample of American adult population surveyed by National Health and Nutrition Examination Survey (NHANES).

Methods: Cross-sectional data from the NHANES were used. Participants were adults aged 18–80 y from 1996–2006. MetS were defined by the updated National Cholesterol Education Program/Adult Treatment Panel III criteria (NCEP-ATP III) for Americans. Receiver operating characteristic (ROC) curve was drawn and the areas under the curve (AUC) were used to assess the ability of these indexes in screening MetS. Statistical differences among the AUC values of these indexes were compared. The association between the anthropometric indexes and MetS was investigated using weighted multivariable-adjusted logistic regression.

Results: 560 (35.2%) males and 529 (26.4%) females were diagnosed with MetS. LAP was the strongest predictor of MetS for men (AUC=0.87, 95% CI 0.85-0.89), and also was the strongest for women [AUC=0.85, 95% confidence interval (CI) 0.83-0.86], according to the ROC curve analysis. In men, differences in AUC values between LAP and other anthropometric indicators were also significant (all $P<0.001$). In women, there was a significant difference in AUC values between LAP and WTI ($P<0.001$), but differences in AUC values between LAP and TyG, VAI were not significant.

Conclusion: The present study indicated that LAP is a better predictor in the clinical setting for identifying individuals with MetS in the American adult population.

Keywords: metabolic syndrome, triglyceride and glucose index (TyG), adiposity index (VAI), lipid accumulation product (LAP), waist-triglyceride index (WTI), anthropometric indexes

Abbreviations: BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; TMFA, total monounsaturated fatty acids; TPFA, total polyunsaturated fatty acids; TSFA, total saturated fatty acids; eGFR, estimated glomerular filtration rate; HDL-C, high density lipoprotein cholesterol; CRP, C-reactive protein; LAP, lipid accumulation product; VAI, visceral adiposity index; TyG, triglyceride and glucose index; WTI, waist-triglyceride index; AUC, area under the curve.

OPEN ACCESS

Edited by:
Patricia Khashayar, Ghent University, Belgium

Reviewed by:
Enoch Odame Anto, Kwame Nkrumah University of Science and Technology, Ghana
Gang Yuan, Huazhong University of Science and Technology, China

***Correspondence:**
Ting Sun
1195037@zju.edu.cn

Specialty section:
This article was submitted to Obesity, a section of the journal Frontiers in Endocrinology

Received: *04 March 2022*
Accepted: *25 March 2022*
Published: *24 May 2022*

Citation:
Li Y, Zheng R, Li S, Cai R, Ni F, Zheng H, Hu R and Sun T (2022) Association Between Four Anthropometric Indexes and Metabolic Syndrome in US Adults. Front. Endocrinol. 13:889785. doi: 10.3389/fendo.2022.889785

INTRODUCTION

Metabolic syndrome (MetS) is a complicated disorder characterized by impaired glucose tolerance, dyslipidemia, elevated blood pressure, abdominal obesity (1–3). MetS is associated with higher risks of cardiovascular diseases, type 2 diabetes, some cancers, and all-cause mortality and has become one of the major challenges facing global and national public health institutions (4, 5). According to the National Health and Nutrition Examination Survey, more than one-third of adults suffer from MetS (6).

Obesity as the core manifestation of MetS has attracted more and more attention (7). There is some evidence to support the assumption that abdominal visceral fat has a stronger correlation with MetS (8, 9). Hence, it is reasonable to define visceral fat as a predictor of MetS. Magnetic resonance imaging (MRI) and computed tomography (CT) are considered as the gold standard for evaluating visceral fat (10). However, they cannot be used in epidemiological studies and clinical routine due to expensive, time-consuming, and exposure patients to radiation and contrast agents. Thus, it is very important to identify a simple and clinically suitable visceral obesity substitute indicator. Body mass index (BMI) is the most commonly used indicator of obesity, but it has limitations in assessing fat distribution (11, 12). Therefore, anthropometric indexes have been suggested to evaluate the amount and location of body fat to track metabolic disorders (13). Recently, visceral obesity index (VAI) and lipid accumulation products (LAP) have been recommended as reliable indicators of visceral obesity. VAI is calculated based on waist circumference (WC), high-density lipoprotein cholesterol (HDL-C), BMI, and triglyceride (TG) and has a separate formula for men and women (14). It has been reported to have a good ability to predict metabolic syndrome in Chinese and Iranian populations (15, 16). LAP is an index of abdominal fat over-accumulation based on TG and WC (13), which is considered as the best predictor of MetS in middle-aged and elderly people in Korea (17).

Insulin resistance (IR) is another core issue of MetS (7). Hyperinsulinaemic-euglycaemic clamp (HEC) is the gold standard for testing IR (18), but this approach is time-consuming and is not suitable for clinical application. The triglyceride and glucose (TyG) index combined with fasting plasma glucose (FPG) and TG has been proposed as an effective substitute for IR and has been reported to have a good predictive ability for MetS in Korean and Chinese populations (17, 19). Recently, inspired by the formula of the TyG index, Liu et al. combined WC with TG to develop a new index called waist-triglyceride index (WTI), which showed a strong ability to distinguish MetS (20).

Even though several papers on the association between anthropometric indicators and the MetS have been published (13), it is still hard to determine explicitly which indicator is the most predictable indicator of MetS. And these studies have limitations in adjusting confounding factors, most of them do not adjust the factors that may affect MetS, such as smoking, drinking, exercise, and socioeconomic factors (21). Postmenopausal women tend to deposit more visceral fat have shown by a large number of studies. Thus, gender may affect the relationship between anthropometric indicators and MetS. As far as we know, up to now, there are limited research on comparing anthropometric indicators of the American population with the predicted strength of MetS by gender. Accordingly, the purpose of this study was to investigate the relationship between anthropometric indicators (VAI, LAP, TyG, WTI) and MetS in American adults and to compare their predictive ability according to gender.

MATERIALS AND METHODS

Data Source

The NHANES is a repeated national representative cross-sectional health examination survey conducted in the United States (US), on behalf of the non-institutionalized population of the US civilian population, which provides estimate of the lifestyle, nutritional status, and health of the US civilian population (22). Since 1999, NHANES has become a continuous survey, with data released every two years. During the survey, participants will complete a questionnaire survey, a series of tests, and offer blood and other biological samples at the mobile screening center (23). Five main parts make up the NHANES database, including demographic, questionnaire, laboratory, diet, and examination data.

More details are available on the official website (https://www.cdc.g-ov/nchs/nhanes/index.htm). The NHANES datasets (1999–2006) were downloaded from DataDryad (https://doi.org/10.5061/dryad.d5h62). Participants provided written informed consents. The National Center for Health Statistics (NCHS) Ethics Review Board approved the collection of the NHANES data.

Participants Selection

We conducted a secondary data analysis based on data extracted from NHANES cycles: 1999-2000, 2001-2002, 2003-2004, and 2004-2006. After a series of screenings, 3894 subjects were included in the final data analysis. Subjects were filtered based on the following exclusion criteria, and were shown in **Figure 1**:

(1) subjects without components of metabolic syndrome data (n=33786);

(2) people aged < 18 years or aged >80 years (n=2342);

(3) drug therapy (diuretics or corticosteroids) that could influence weight (n=207);

(4) with a suspected renal or liver insufficiency: an estimated glomerular filtration rate (eGFR) ≤60 mL/minute/1.73 m^2 (n=182) or serum total bilirubin concentration ≥1.5 mg/L (n=131), or alanine aminotransferase (ALT) level ≥120 U/L (n=45).

(5) any cancer or malignancy (n= 905).

Anthropometric Indexes Measurement

Each participant had a home interview and finished a physical examination at a mobile examination center (MEC). Participants

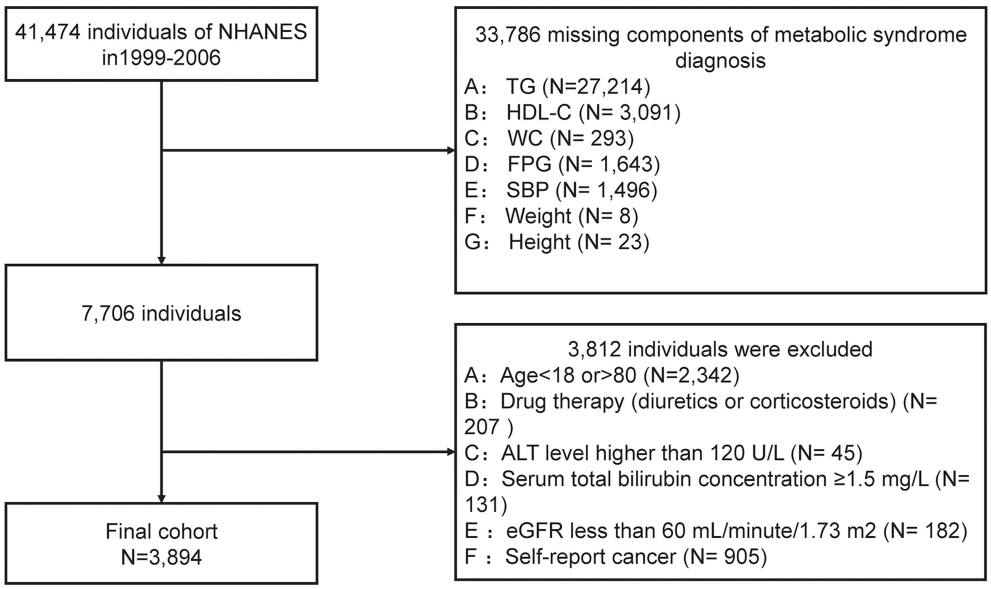


FIGURE 1 | Flow chart.

were required to fast at least 9 hours before the health examination (22). Height and weight were measured at the MEC by a standardized protocol. BMI was calculated by dividing the weight in kilograms by the square of the height in meters and then rounding to the nearest 1/10 cm. WC was measured by an inelastic ruler with a minimum scale of one millimeter at the end of a normal exhalation, and when standing naturally with legs opened about 25-30 cm apart. Placed the ruler at the midpoint of the connecting line between the upper edge of the top of the iliac crest and the lower edge of the 12th rib (usually the natural narrowest part of the waist) and horizontally circled the abdomen, and then rounded to 0.1cm (24). After at least 5 minutes of rest at the MEC, using a standardized mercury sphygmomanometer to measure blood pressure in a sitting position (25).

LAP, VAI, TyG, and WTI were calculated by using the following formulas (14, 20, 26, 27):

$$\begin{aligned}&\mathrm{LAP} = [\mathrm{WC\ (cm)} - 65] \times \mathrm{TG(mmol/L)}\\ &\text{for male and } [\mathrm{WC\ (cm)} - 58] \times \mathrm{TG}\\ &\mathrm{(mmol/L)\ \ for\ \ female}\end{aligned}$$

$$\mathrm{VAI} = \begin{bmatrix}\mathrm{WC\ \ (cm)}/39.68 + \\ (1.88 \times \mathrm{BMI(kg/m2)})\end{bmatrix} \times (\mathrm{TG(mmol/L)}/1.03) \times (1.31/\mathrm{HDL-C(mmol/L)}) \text{ for male and } [\mathrm{WC\ (cm)}/36.58 + (1.89 \times \mathrm{BMI\ (kg/m2)})]\ \ (\mathrm{TG(mmol/L)}/0.81) \times (1.52/\mathrm{HDL-C(mmol/L)})\ \text{ for female}$$

$$\mathrm{TyG} = \mathrm{Ln}\ \ [\mathrm{TG(mg/dL)} \times \mathrm{FPG\ \ (mg/dL)}/2]$$

$$\mathrm{WTI} = \mathrm{Ln}\ \ [\mathrm{TG(mg/dL)} \times \mathrm{WC(cm)}/2]$$

Biochemical Measurements

Total cholesterol (TC), TG, low-density lipoprotein cholesterol (LDL-C), and HDL-C were estimated in subjects who fasted for at least 8.5 hours but less than 24 hours. Venous blood samples of participants were collected and processed in MECs following the NHANES protocols.

According to the established protocols, the samples were packed in cold bags or dry ice, and directly transported to the Collaborative Studies Clinical Laboratory by Federal Express and stored at-70°C for analysis (28). Johns Hopkins University School of Medicine Lipoprotein Analytical Laboratory tested the blood samples of lipid. Interlaboratory quality control carried out by the laboratories met the Centers for Disease Control and Prevention (CDC) program's acceptable performance of allowable bias and imprecision.

HDL-C was determined using a nephelometric immunoassay on the Hitachi 717 Analyzer (Hitachi Global Storage Technologies, California). The FPG was measured using the enzyme hexokinase (HK) method. TG was measured using an automatic direct chemiluminescence analyzer (Beckman Synchron LX20, USA). All laboratory measurements met the requirements of the standardization and certification program. More detailed information about the analyzers and methods used can be obtained from the laboratory method file available on the NHANES website.

MetS Definition

MetS was defined according to the updated National Cholesterol Education Program/Adult Treatment Panel III criteria (NCEP-ATP III) for Americans, that was, meeting the following three or more components: WC ≥102cm for male or ≥88cm for female; blood pressure ≥130/85mmHg or treated with anti-hypertensive

drugs; or FPG ≥5.6 mmol/L or drugs used for treating diabetes; TG ≥150mg/dL or treated with drugs for this lipid abnormality; HDL-C <40mg/dL for male or <50mg/dL for female or treated with drugs for this lipid abnormality (29).

Variables

In this study, the independent variables were VAI, LAP, TyG, and WTI respectively. The dependent variable was MetS. Covariates were prioritized according to the previous research on risk factors for MetS (29–32). Socio-demographic characteristics such as sex, education, race/ethnicity were collected. A self-reported questionnaire was applied to evaluate medication use (glucose-lowering drugs, lipid-lowering drugs, and anti-hypertensive drugs). According to the self-completed questionnaire, physical activities were divided into four categories (moderate, low, moderate, and high), and smoking was separated into current smokers, former smokers, and non-smokers. We also collected a series of laboratory data such as homocysteine, glucose, insulin, hs-CRP, TG, TC, HDL-C, LDL-C, albumin, total bilirubin, total protein, uric acid, and BUN, and a set of the dietary condition like alcohol intake, energy, total saturated fatty acids (TSFA), total polyunsaturated fatty acids (TPFA), total monounsaturated fatty acids (TMFA), total fat, protein. The physical activity categories were based on the distribution of MET-minute levels for the present NHANES sample. Diabetes was defined as a self-reported physician diagnosis of diabetes or a fasting glucose concentration >126 mg/dL. Hypertension was defined by ≥1 of the following criteria: systolic blood pressure ≥140 mmHg or diastolic blood pressure ≥90 mmHg or self-reported physician diagnosis of hypertension. Drink consumption is defined as 5 gm or more drinks per day. Insulin resistance (HOMA-IR) was calculated as following formula (33):

$$\text{HOMA} - \text{IR} = [\text{fasting insulin concentration } (\mu\text{IU/mL}) \times \text{FPG } (\text{mml/L})/22.5]$$

Statistical Methods

The statistical analysis was conducted by the guidelines of the CDC (https://www.cdc.gov/nchs/nhanes/tutorials/default.aspx). All analyses used EmpowerStats (http://www.empower.stats.com, X&Y Solutions, Inc., Boston, MA) and the statistical software packages R (http://www.R-project.org, The R Foundation R.3.4.3).

In this study, sample weights were adjusted according to the recommendations of the NCHS. To present nationally representative estimates, survey analysis procedures were used to account for the sample weights (MEC exam weight), stratification, and clustering of the complex sampling design (34). We calculated the sample weight for the 8 years of data from 1999 to 2006 as $WT_{99-06} = (1/4) \times WT_{05-06} + (1/4) \times WT_{03-04} + (1/2) \times WT_{99-02}$, WT_{99-02} is the variable WTMEC4YR from the NHANES 1999–2000 and NHANES 2001–2002; $WT_{03-04}$and WT_{05-06} were the variable WTMEC2YR from the NHANES 2003–2004 and NHANES 2005–2006 demographic file, respectively (35, 36). Data were expressed as weighted proportions (± Standard Error (SE)) for categorical variables and as weighted means ± SE for continuous variables depending on their type. In estimating standard errors, the complex sample design was incorporated by using Taylor series linearization with provided survey design variables (37). We tested differences in characteristics between the MetS group and the non-MetS group with a one-way analysis of variance for continuous variables and with chi-square tests for categorical variables. Weighted logistic regression was applied to analyze the relationship between anthropometric indicators (VAI, LAP, TyG, WTI) and MetS. We selected these confounders on the basis of their associations with the MetS or a change in effect estimate of more than 10% (38). Further, the receiver operating characteristic (ROC) curve was drawn and the area under curve (AUC) value was calculated to evaluate the predictive ability of LAP, TyG, VAI, and WTI for MetS. DeLong et al's non-parametric method was performed to compare the AUC between LAP and other indexes (32). According to the maximum value of the sum of sensitivity and specificity, the best cutoff values of LAP, VAI, TyG, and WTI for predicting MetS were determined. All statistical significance was set to $P<0.05$.

RESULTS

Baseline Characteristics of the Subjects

As shown in **Figure 1**, according to the exclusion criteria, 3794 subjects (1893 males and 2001 females) were finally included in this study. **Table 1** describes the baseline characteristics of the population. At baseline, 560 (35.2%) males and 529 (26.4%) females were diagnosed with MetS. The average age of included subjects was 38.17± 0.47 years for the non-MetS group and 47.25 ± 0.42 years for the MetS group. Significantly higher levels of mean systolic, mean diastolic, glucose-plasma, TC, HOMA-IR, TG, LDL-C, uric acid, alanine aminotransferase, aspartate aminotransferase, and gamma-glutamyl transferase were observed in subjects with MetS. But they had significantly lower levels of HDL-C, albumin than those without MetS.

In addition, as shown in **Table 1**, all of the anthropometric indexes in the MetS group, including VAI, LAP, TyG, WTI, BMI, WC, height, BMI, and weight were significantly increased.

The Anthropometric Indexes for Predicting MetS

What can be seen in **Figure 2** is LAP, VAI, TyG, and WTI increased in proportion to the number of MetS components. **Table 2** and **Figure 3** show the AUC values [95% confidence interval (CI)] of the anthropometric indexes used to screen American adults with MetS. Of the four indexes examined, the highest AUC was LAP, 0.8458 for women (95% CI: 0.8272-0.8645) and 0.8685 for men (95% CI: 0.8504-0.8865). The optimum cutoff values of LAP predicted were 52.4291 (sensitivity 0.8117, specificity 0.7677) in women and 53.3125 (sensitivity 0.8013, specificity 0.7852) in men. The optimal cut-off points for TyG were 8.8221 in men and 8.6897 in women.

TABLE 1 | Baseline characteristics of the participants.

	Non-MetS Group	MetS Group	*P* value
Age (yr)	38.17 ± 0.47	47.25 ± 0.42	<0.0001
Sex, %			0.0037
Female	50.86 ± 0.80	45.58 ± 1.85	
Male	49.14 ± 0.80	54.42 ± 1.85	
Race, %			0.006
Non-Hispanic Black	11.24 ± 1.10	7.28 ± 1.12	
Mexican American	8.82 ± 1.04	7.99 ± 1.15	
Other Hispanic	6.03 ± 1.22	6.47 ± 1.72	
Non-Hispanic White	69.1 ± 0.83	73.06 ± 2.62	
Other race	4.8 ± 0.62	5.2 ± 1.13	
Education, %			<0.0001
< high school	18.22 ± 1.13	21.56 ± 1.30	
High school	24.13 ± 1.20	29.91 ± 1.77	
> high school	57.65 ± 1.68	48.53 ± 1.74	
Poverty to income ratio	3.0 3 ± 0.07	3.11 ± 0.07	0.3338
Smoking, %			<0.0001
Never	51.84 ± 1.49	46.54 ± (2.17	
Former	20.42 ± 1.22	27.39 ± 1.68	
Current	27.74 ± 1.20	26.07 ± 1.65	
Drink consumption, %			<0.0001
No	70.35 ± 1.52	78.59 ± 1.51	
Yes	29.65 ± 1.52	21.4 1 ± 1.51	
Physical activity, %			0.013
Sedentary	15.9 ± 1.00	18.21 ± 1.71	
Low	28.22 ± 1.61	31.49 ± 2.12	
Moderate	20.63 ± 0.86	16.84 ± 1.71	
High	35.24 ± 1.38	33.46 ± 2.20	
Medication use			
Glucose-lowering drugs, %			<0.0001
No	99.43 ± 0.15	92.95 ± 0.92	
Yes	0.57 ± 0.15	7.05 ± 0.92	
Lipid-lowering drugs, %			<0.0001
No	98.19 ± 0.29	85.53 ± 1.17	
Yes	1.81 ± 0.29	14.47 ± 1.17	
Antihypertensive drugs, %			<0.0001
No	96.94 ± 0.43	79.58 ± 1.32	
Yes	3.06 ± 0.43	20.42 ± 1.32	
Laboratory data			
Triglyceride (mg/dL)	109.92 ± 1.56	235.24 ± 9.63	<0.0001
LDL-cholesterol (mg/dL)	118.33 ± 0.92	127.25 ± 1.73	<0.0001
Albumin (g/dL)	4.36 ± 0.01	4.26 ± 0.01	<0.0001
Alanine aminotransferase ALT (U/L)	23.44 ± 0.28	29.73 ± 0.7	<0.0001
Aspartate aminotransferase AST (U/L)	23.19 ± 0.20	24.82 ± 0.49	0.003
Gamma glutamyl transferase (U/L)	24.37 ± 0.53	37.2 ± 2.07	<0.0001
Glucose, serum (mg/dL)	89.43 ± 0.40	106.81 ± 1.07	<0.0001
Total bilirubin (mg/dL)	0.69 ± 0.01	0.68 ± 0.01	0.3674
Total protein (g/dL)	7.330 ± 0.02	7.29 ± 0.02	0.2184
Triglycerides (mg/dL)	101.23 ± 1.64	222.2 ± 10.33	<0.0001
Uric acid (mg/dL)	5.05 ± 0.02	5.83 ± 0.06	<0.0001
Plasma glucose (mmol/L)	5.19 ± 0.02	6.23 ± 0.07	<0.0001
HDL-cholesterol (mg/dL)	54.37 ± 0.34	42.03 ± 0.60	<0.0001
HOMA-IR	2.09 ± 0.04	5.12 ± 0.20	<0.0001
Dietary			
Energy (kcal)	2330.21± 22.24	2255.53 ± 31.96	0.0984
Total monounsaturated fatty acids (gm)	32.54 ± 0.46	32.71 ± 0.69	0.8449
Total polyunsaturated fatty acids (gm)	18.01 ± 0.25	17.57 ± 0.41	0.3831
Protein (gm)	85.99 ± 0.96	85.05 ± 1.64	0.645
Total saturated fatty acids (gm)	28.43 ± 0.40	28.54 ± 0.62	0.8887
Total fat (gm)	86.78 ± 1.12	86.78 ± 1.60	0.9993
Anthropometry			
Weight (kg)	75.37 ± 0.38	92.64 ± 0.88	<0.0001
Standing height (cm)	169.79 ± 0.24	170.84 ± 0.38	0.0113
Body mass index (kg/m^2)	26.07 ± 0.13	31.61 ± 0.25	<0.0001

(Continued)

TABLE 1 | Continued

	Non-MetS Group	MetS Group	*P* value
Waist circumference (cm)	90.18 ± 0.32	106.83 ± 0.56	<0.0001
mean systolic	114.44 ± 0.34	123.8 ± 0.55	<0.0001
mean diastolic	70.26 ± 0.27	75.2 ± 0.49	<0.0001
VAI	1.44 ± 0.03	4.33 ± 0.25	<0.0001
LAP	34.15 ± 0.77	110.23 ± 4.55	<0.0001
TyG.	8.32 ± 0.01	9.18 ± 0.03	<0.0001
WTI	8.29 ± 0.01	9.16 ± 0.03	<0.0001
MetS Components			
Elevated BP, %			<0.0001
No	86.86 ± 0.75	46.68 ± 1.81	
Yes	13.14 ± 0.75	53.32 ± 1.81	
Elevated TG level, %			<0.0001
No	88.26 ± 0.72	34.05 ± 1.84	
Yes	11.74 ± 0.72	65.95 ± 1.84	
Reduced HDL-C level, %			<0.0001
No	76.76 ± 1.05	21.47 ± 1.82	
Yes	23.24 ± 1.05	78.53 ± 1.82	
Drugs used for low level of high-density lipoprotein cholesterol, %			<0.0001
No	98.19 ± 0.29	85.76 ± 1.19	
Yes	1.81 ± 0.289	14.24 ± 1.19	
Drugs used for high level of triglyceride, %			0.0047
No	100 ± 0	99.72 ± 0.14	
Yes	0 ± 0	0.28 ± 0.14	
Elevated WC, %			
No	70.23 ± 1.18	15.16 ± 1.54	
Yes	29.77 ± 1.18	84.84 ± 1.54	
HDM, %			<0.0001
No	85.41 ± 0.89	32.8 ± 2.12	
Yes	14.59 ± 0.89	67.2 ± 2.12	

Data are expressed as weighted proportions [± Standard Error (SE)] for categorical variables and as weighted means ± Standard Error for continuous variables depending on its type. Variables between groups with and without MetS were compared using one-way analysis of variance for continuous variables and with chi-square tests for categorical variables.

This study also reported other details of all the anthropometric indexes such as negative predictive value (NPV) and positive predictive value (PPV) in **Table 2**.

Comparison of AUC Values Between LAP and Other Indexes in Men and Women

Table 3 shows the differences in AUC values between LAP and other indexes for screening MetS. In men, differences in AUC values between LAP and TyG, WTI, VAI were significant (all $P<0.001$). In women, the AUC value between LAP and WTI was significantly different ($P<0.001$), but the statistical difference between LAP and TyG, VAI was not significant. The above results showed that LAP had a stronger ability to identify MetS than other anthropometric indexes.

Associations Between Four Anthropometric Indexes and MetS

Table 4 shows the adjusted odds ratios (ORs) (95% CIs) of anthropometric indexes for MetS in women and men. After adjusting for age, education, alcohol, current or a past cigarette smoker, poverty to income ratio, physical activity, uric acid, energy intake, protein intake, TMFA intake, TPFA intake, TSFA intake, total fat intake, glucose-lowering drugs, lipid-lowering drugs and anti-hypertensive drugs, the prevalence of MetS is higher in the third and fourth quartiles (Q3 and Q4) of LAP, TyG, VAI, and WTI in women. For VAI, Q3 was at 6.084 (2.320, 15.955) and Q4 was at 71.681 (26.334, 195.112), showing a higher risk for MetS compared to Q1. Q3 of LAP was at 24.174 (5.690, 102.698) and Q4 was at 199.843 (46.394, 860.825), which indicated that MetS was risker than the first quartile (Q1) of LAP. For TyG, Q3 was at 6.058 (2.871, 12.783) and Q4 was at 37.708 (17.214, 82.598), revealing that the risk of MetS was higher than Q1. For WTI, Q3 was at 4.747 (2.269, 9.929) and Q4 was at 38.472 (17.723, 83.513), indicating a higher risk for MetS compared to Q1.

In the fully adjusted model in men, each 1 unit increase in VAI increased the MetS risk by 237.5%. LAP increased by 1 unit, the incidence of MetS increased by 5.2%. The fully adjusted OR (95%CI) for TyG and WTI in men, respectively, were 14.796 (8.771, 24.961) and 20.115 (11.454, 35.325).

For sensitivity analysis, we converted VAI, LAP, TyG, WTI from continuous variables to categorical variables. The *P* for the trend of VAI, LAP, TyG, WTI with categorical variables was consistent with the result when VAI, LAP, TyG, WTI was a continuous variable.

DISCUSSION

This study assessed the capability of four low-cost, non-invasive and easily-calculated anthropometric indicators, including VAI,

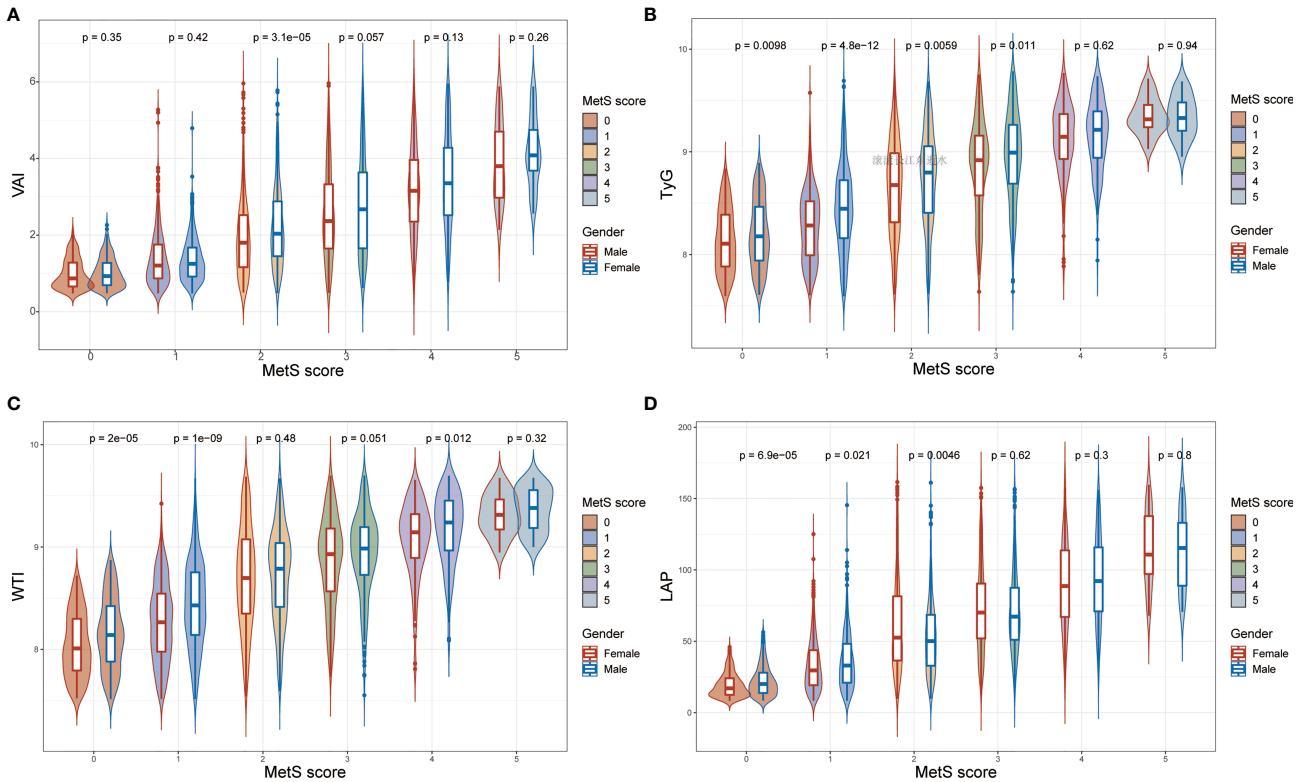


FIGURE 2 | The values of **(A)** visceral adiposity index (VAI), **(B)** triglycerides and glucose (TyG), **(C)** waist-triglyceride index and **(D)** lipid accumulation product (LAP) according to MetS components in both genders.

LAP, TyG, and WTI, to predict MetS. In this cross-sectional analysis of American adults, LAP, VAI, TyG, and WTI were significantly associated with MetS in both genders. Furthermore, ROC curve analysis showed that all parameters could distinguish subjects with MetS, and the AUC values were higher than 0.7 in both genders, of which LAP showed the greatest diagnostic accuracy.

To the best of our knowledge, this was the first study to explore the relationship between anthropometric parameters (LAP, VAI, TyG, and WTI) and MetS in the American population according to different genders, and their ability to diagnose MetS has been further evaluated.

LAP is reported to be associated with MetS, cardiovascular disease, and type 2 diabetes (13, 39, 40). Among these four indicators, LAP has the advantages of simplicity, low cost, and wide applicability to different populations. Shin et al. (17). reported that among the middle-aged and elderly people (aged 40 years or older) in South Korea, LAP was the best index for

TABLE 2 | The anthropometric indexes for predicting Mets.

	Test	AUC	95%CI low	95%CI upp	Cutoff Value	Specificity	Sensitivity	PPV	NPV
Women	VAI	0.8261	0.8038	0.8483	2.0786	0.7901	0.7332	0.5274	0.9026
	LAP	0.8458	0.8272	0.8645	52.4291	0.7677	0.8117	0.5403	0.9238
	TyG	0.8315	0.8093	0.8537	8.6897	0.7770	0.7983	0.5527	0.9178
	WTI	0.8179	0.7961	0.8396	8.8231	0.8206	0.7043	0.5787	0.8880
Men	VAI	0.8309	0.8088	0.8530	1.8196	0.7590	0.7862	0.5656	0.8989
	LAP	0.8685	0.8504	0.8865	53.3125	0.7852	0.8013	0.5940	0.9097
	TyG	0.8237	0.8016	0.8458	8.8221	0.7937	0.7330	0.5668	0.8898
	WTI	0.8335	0.8121	0.8550	8.8820	0.8296	0.7063	0.6078	0.8831
Overall	VAI	0.8263	0.8106	0.8420	2.0798	0.8063	0.7183	0.5691	0.8893
	LAP	0.8565	0.8435	0.8695	53.3255	0.7805	0.7989	0.5689	0.9146
	TyG	0.8279	0.8123	0.8435	8.8478	0.8337	0.7096	0.5718	0.8901
	WTI	0.8251	0.8098	0.8404	8.8233	0.8036	0.7256	0.5718	0.8901

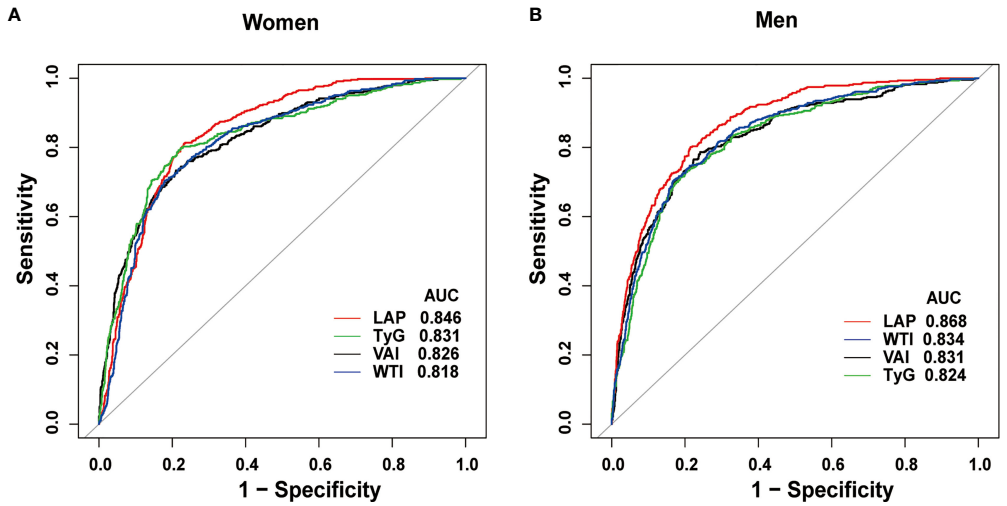


FIGURE 3 | Receiver operating characteristic curves of LAP and other indexes in women **(A)** and men **(B)** for identifying MetS.

predicting MetS comparing with VAI, WHtR, and TyG. Similar results were observed in subsequent studies, which compared more different anthropometric indicators (41). In the present study, the best threshold of LAP for predicting MetS was 53.3125 in males and 52.4291 in females. In Argentinian healthy individuals, a similar value was achieved (53.63 in all subjects). However, a slightly lower best threshold was suggested in the Iranian population (49.71 for females and 39.89 females) (42) and in Spanish adults (48.09 for males and 31.77 for females) (43).The optimal threshold of MetS predicted by LAP is different from other studies, which may be due to the ethnic modification of insulin resistance and abdominal fat distribution, age of the enrolled population, as well as the diagnostic criteria of MetS used.

VAI is an important indicator for insulin resistance and visceral obesity and is associated with CVD risk (14, 19). In this study, the AUC values of VAI predicting MetS were 0.8309 and 0.8261 in males and females, respectively. The best cutoff point for VAI to predict MetS in female was 2.0786 and in male was 1.8196, close to the optimal critical point 2 of VAI for predicting MetS in the middle-aged and elderly in China (44).

TABLE 3 | Comparison of AUC values between LAP and other indexes in both genders.

	Difference between Area (95%CI)	*P*-value
Women		
LAP *vs* TyG	0.0016 (-0.0015-0.0046)	0.8409
LAP *vs* WTI	0.0215 (0.0187-0.0243)	<0.001
LAP *vs* VAI	0.0087 (0.0055-0.0120)	0.2628
Men		
LAP *vs* TyG	0.0553 (0.0506-0.0600)	<0.001
LAP *vs* WTI	0.0429 (0.0391-0.0468)	<0.001
LAP *vs* VAI	0.0521 (0.0469-0.0573)	<0.001

Delong. Clarke-Pearson's nonparametric approach was used to compare the AUCs of indexes.

However, the cutoff in overweight and obese Turkish patients was higher with a value of 2.205 (45).

The results of our analysis also revealed the relatively high usefulness of TyG in identifying individuals with MetS. TyG is an index that combines FPG and TG and is considered to be a substitute for insulin resistance. Its ability to identify MetS has been confirmed by several studies. In middle-aged and elderly Chinese (44), TyG was suggested to be a credible surrogate marker for identifying MetS with the AUC of 0.802, and the best cut-off values were 8.9 and 8.7 for males and females, respectively. In the present population, the optimal cut-off values for males and females were 8.8221 and 8.6897 respectively, which was similar to their results. Furthermore, in the present study, the difference between TyG and LAP in predicting MetS of women was not significant.

In both genders, the predictive ability of WTI was significantly different from that of LAP, which suggested that WTI was weaker than LAP in predicting MetS. Inspired by the TyG, WTI was first proposed by Liu et al. (20). Their study showed that WTI and MetS risk in the Chinese population was associated, the AUC of WTI predicting MetS is 0.881 in women and 0.830 in men. In the present study, with the increase of the number of components of MetS, the value of WTI increases gradually. WTI has a good ability to predict MetS, and AUC is 0.8335 and 0.8179 in men and women, respectively, although it is weaker than LAP. And considering that the formulas of WTI and LAP are both combinations of TG and WC, and the calculation of WTI is more complex, WTI may not be the best index in identifying MetS.

Strength and Limitations

One advantage of the study is that, first and foremost, the analysis included several confounders associated with MetS, such as smoking, alcohol consumption, physical activity,

TABLE 4 | Associations between MetS and LAP, VAI, TyG and WTI.

Exposure	Adjusted Odds Ratio (95%CI)	*P*-value	Adjusted Odds Ratio (95%CI)	*P*-value
	Women		Men	
VAI (continuous variable)	4.174 (3.224, 5.404)	<0.001	3.375 (2.716, 4.194)	<0.001
VAI Quartile				
Q1 0.479-0.968	Reference		Reference	
Q2 0.968-1.543	2.838 (1.044, 7.716)	0.041	2.288 (0.969, 5.407)	0.059
Q3 1.544-2.510	6.084 (2.320, 15.955)	<0.001	5.622 (2.532, 12.482)	<0.001
Q4 2.510-5.963	71.681 (26.334, 195.112)	<0.001	36.702 (16.414, 82.067)	<0.001
P for trend	<0.001		<0.001	
LAP (continuous variable)	1.047 (1.038, 1.056)	<0.001	1.052 (1.043, 1.061)	<0.001
LAP Quartile				
Q1 8.337-22.744	Reference		Reference	
Q2 22.761-40.577	11.817 (2.734, 51.083)	<0.001	4.865 (1.245, 19.005)	0.023
Q3 40.626-68.526	24.174 (5.690, 102.698)	<0.001	23.137 (6.320, 84.704)	<0.001
Q4 68.542-161.564	199.843 (46.394, 860.825)	<0.001	125.125 (33.737, 464.075)	<0.001
P for trend	<0.001		<0.001	
TyG (continuous variable)	27.128 (14.724, 49.983)	<0.001	14.796 (8.771, 24.961)	<0.001
TyG Quartile				
Q1 7.593-8.163	Reference		Reference	
Q2 8.164-8.545	1.582 (0.712, 3.513)	0.260	3.452 (1.266, 9.407)	0.015
Q3 8.546-8.952	6.058 (2.871, 12.783)	<0.001	6.354 (2.476, 16.306)	<0.001
Q4 8.954-9.785	37.708 (17.214, 82.598)	<0.001	38.935 (15.069, 100.601)	<0.001
P for trend	<0.001		<0.001	
WTI (continuous variable)	20.556 (11.610, 36.395)	<0.001	20.115 (11.454, 35.325)	<0.001
WTI Quartile				
Q1 7.514-8.132	Reference		Reference	
Q2 8.133-8.543	1.535 (0.697, 3.378)	0.287	2.109 (0.759, 5.861)	0.153
Q3 8.543-8.968	4.747 (2.269, 9.929)	<0.001	5.976 (2.346, 15.228)	<0.001
Q4 8.969-9.698	38.472 (17.723, 83.513)	<0.001	38.645 (15.085, 99.003)	<0.001
P for trend	<0.001		<0.001	

Adjusted for age (years); race; education; alcohol; smoker; poverty to income ratio; physical activity; uric acid; energy; total monounsaturated fatty acids; total polyunsaturated fatty acids; protein; total saturated fatty acids; total fat; glucose-lowering drugs; lipid-lowering drugs; antihypertensive drugs.

dietary intake, and socioeconomic factors. Moreover, the data analyzed in this study were from the NHANES database, which was national and representative in scope, the anthropometric data and laboratory data are of high quality.

The limitations of this study need to be pointed out (1). This was a cross-sectional study and cannot draw any conclusions about the anthropometric index changes over time (2). The study was limited to American adults, the applicability of these results to other populations may be limited. (3) For females, due to the lack of data on menopause, their menopausal status cannot be taken into account in data analysis. (4) This study defined MetS using NCEP-ATP III criteria. Thus, whether a consistent conclusion can be obtained under other criteria requires further studies. (5) A common problem in observational studies is unmeasured confunders. Although we have adjusted many potential confounding factors, we can't rule out the possibility of residual confounding caused by unmeasured or unidentified factors.

The results of this study showed that LAP, VAI, TyG, WTI were reliable predictors of MetS for American adults, and LAP has the largest AUC in predicting MetS. Among females, the difference in AUC between LAP and TyG, VAI was not significant. We suggest that LAP is a useful screening indicator to identify MetS at a minimum cost in the clinical setting, considering the superiority and simplicity of LAP in identifying MetS.

CONCLUSIONS

The present study indicated that LAP is a better predictor in the clinical setting for identifying individuals with MetS in the US adult population.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding author.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by The National Center for Health Statistics (NCHS) Ethics Review Board. The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

Conceptualization: YL and TS; Methodology: RZ; Software: RZ; Validation: SL, RC, and HZ; Formal analysis: RZ, FN, and TS;

Writing—original draft preparation: YL and RH; Writing—review and editing: YL and TS; All authors contributed to the article and approved the submitted version.

ACKNOWLEDGMENTS

We thank Patel et al. for sharing the data.

REFERENCES

1. Ford ES, Li C, Sattar N. Metabolic Syndrome and Incident Diabetes: Current State of the Evidence. *Diabetes Care* (2008) 31(9):1898–904. doi: 10.2337/dc08-0423
2. Kassi E, Pervanidou P, Kaltsas G, Chrousos G. Metabolic Syndrome: Definitions and Controversies. *BMC Med* (2011) 9:48. doi: 10.1186/1741-7015-9-48
3. Oda E. Metabolic Syndrome: Its History, Mechanisms, and Limitations. *Acta Diabetol* (2012) 49(2):89–95. doi: 10.1007/s00592-011-0309-6
4. Expert Panel on Detection E and Treatment of High Blood Cholesterol in A. Executive Summary of the Third Report of the National Cholesterol Education Program (Ncep) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). *JAMA* (2001) 285(19):2486–97. doi: 10.1001/jama.285.19.2486
5. Ford ES, Giles WH, Dietz WH. Prevalence of the Metabolic Syndrome Among Us Adults: Findings From the Third National Health and Nutrition Examination Survey. *JAMA* (2002) 287(3):356–9. doi: 10.1001/jama.287.3.356
6. Aguilar M, Bhuket T, Torres S, Liu B, Wong RJ. Prevalence of the Metabolic Syndrome in the United States, 2003-2012. *JAMA* (2015) 313(19):1973–4. doi: 10.1001/jama.2015.4260
7. Reaven GM. Banting Lecture 1988. Role of Insulin Resistance in Human Disease. *Diabetes* (1988) 37(12):1595–607. doi: 10.2337/diab.37.12.1595
8. Lebovitz HE, Banerji MA. Point: Visceral Adiposity Is Causally Related to Insulin Resistance. *Diabetes Care* (2005) 28(9):2322–5. doi: 10.2337/diacare.28.9.2322
9. Borel AL, Nazare JA, Smith J, Aschner P, Barter P, Van Gaal L, et al. Visceral, Subcutaneous Abdominal Adiposity and Liver Fat Content Distribution in Normal Glucose Tolerance, Impaired Fasting Glucose and/or Impaired Glucose Tolerance. *Int J Obes (Lond)* (2015) 39(3):495–501. doi: 10.1038/ijo.2014.163
10. Cornier MA, Despres JP, Davis N, Grossniklaus DA, Klein S, Lamarche B, et al. Assessing Adiposity: A Scientific Statement From the American Heart Association. *Circulation* (2011) 124(18):1996–2019. doi: 10.1161/CIR.0b013e318233bc6a
11. Mamtani MR, Kulkarni HR. Predictive Performance of Anthropometric Indexes of Central Obesity for the Risk of Type 2 Diabetes. *Arch Med Res* (2005) 36(5):581–9. doi: 10.1016/j.arcmed.2005.03.049
12. Nevill AM, Stewart AD, Olds T, Duncan MJ. A New Waist-to-Height Ratio Predicts Abdominal Adiposity in Adults. *Res Sports Med* (2020) 28(1):15–26. doi: 10.1080/15438627.2018.1502183
13. Tellechea ML, Aranguren F, Martinez-Larrad MT, Serrano-Rios M, Taverna MJ, Frechtel GD. Ability of Lipid Accumulation Product to Identify Metabolic Syndrome in Healthy Men From Buenos Aires. *Diabetes Care* (2009) 32(7):e85. doi: 10.2337/dc08-2284
14. Amato MC, Giordano C, Galia M, Criscimanna A, Vitabile S, Midiri M, et al. Visceral Adiposity Index: A Reliable Indicator of Visceral Fat Function Associated With Cardiometabolic Risk. *Diabetes Care* (2010) 33(4):920–2. doi: 10.2337/dc09-1825
15. Motamed N, Khonsari MR, Rabiee B, Ajdarkosh H, Hemasi GR, Sohrabi MR, et al. Discriminatory Ability of Visceral Adiposity Index (Vai) in Diagnosis of Metabolic Syndrome: A Population Based Study. *Exp Clin Endocrinol Diabetes* (2017) 125(3):202–7. doi: 10.1055/s-0042-119032
16. Wang H, Liu A, Zhao T, Gong X, Pang T, Zhou Y, et al. Comparison of Anthropometric Indices for Predicting the Risk of Metabolic Syndrome and Its Components in Chinese Adults: A Prospective, Longitudinal Study. *BMJ Open* (2017) 7(9):e016062. doi: 10.1136/bmjopen-2017-016062
17. Shin KA, Kim YJ. Usefulness of Surrogate Markers of Body Fat Distribution for Predicting Metabolic Syndrome in Middle-Aged and Older Korean Populations. *Diabetes Metab Syndr Obes* (2019) 12:2251–9. doi: 10.2147/DMSO.S217628
18. DeFronzo RA, Tobin JD, Andres R. Glucose Clamp Technique: A Method for Quantifying Insulin Secretion and Resistance. *Am J Physiol* (1979) 237(3):E214-23. doi: 10.1152/ajpendo.1979.237.3.E214
19. 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. doi: 10.1186/s12933-014-0146-3
20. Liu PJ, Lou HP, Zhu YN. Screening for Metabolic Syndrome Using an Integrated Continuous Index Consisting of Waist Circumference and Triglyceride: A Preliminary Cross-Sectional Study. *Diabetes Metab Syndr Obes* (2020) 13:2899–907. doi: 10.2147/DMSO.S259770
21. Suliga E, Ciesla E, Gluszek-Osuch M, Rogula T, Gluszek S, Koziel D. The Usefulness of Anthropometric Indices to Identify the Risk of Metabolic Syndrome. *Nutrients* (2019) 11(11):2598. doi: 10.3390/nu11112598
22. Curtin LR, Mohadjer LK, Dohrmann SM, Montaquila JM, Kruszan-Moran D, Mirel LB, et al. The National Health and Nutrition Examination Survey: Sample Design, 1999-2006. *Vital Health Stat 2* (2012) 155):1–39.
23. Patel CJ, Pho N, McDuffie M, Easton-Marks J, Kothari C, Kohane IS, et al. A Database of Human Exposomes and Phenomes From the US National Health and Nutrition Examination Survey. *Sci Data* (2016) 3:160096. doi: 10.1038/sdata.2016.96
24. Bawadi H, Abouwatfa M, Alsaeed S, Kerkadi A, Shi Z. Body Shape Index Is a Stronger Predictor of Diabetes. *Nutrients* (2019) 11(5):1018. doi: 10.3390/nu11051018
25. Chobanian AV, Bakris GL, Black H, Cushman WC, Green LA, Izzo JL Jr, et al. The Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure: The Jnc 7 Report. *JAMA* (2003) 289(19):2560–72. doi: 10.1001/jama.289.19.2560
26. Kahn HS. The "Lipid Accumulation Product" Performs Better Than the Body Mass Index for Recognizing Cardiovascular Risk: A Population-Based Comparison. *BMC Cardiovasc Disord* (2005) 5:26. doi: 10.1186/1471-2261-5-26
27. Simental-Mendia LE, Rodriguez-Moran M, Guerrero-Romero F. The Product of Fasting Glucose and Triglycerides as Surrogate for Identifying Insulin Resistance in Apparently Healthy Subjects. *Metab Syndr Relat Disord* (2008) 6(4):299–304. doi: 10.1089/met.2008.0034
28. Carroll MD, Kruszon-Moran D, Tolliver E. Trends in Apolipoprotein B, Non-High-Density Lipoprotein Cholesterol, and Low-Density Lipoprotein Cholesterol for Adults Aged 20 and Over, 2005-2016. *Natl Health Stat Rep* (2019) 127):1–16.
29. Grundy SM, Cleeman JI, Daniels SR, Donato KA, Eckel RH, Franklin BA, et al. Diagnosis and Management of the Metabolic Syndrome: An American Heart Association/National Heart, Lung, and Blood Institute Scientific Statement: Executive Summary. *Crit Pathw Cardiol* (2005) 4(4):198–203. doi: 10.1097/00132577-200512000-00018
30. Perona JS, Schmidt Rio-Valle J, Ramirez-Velez R, Correa-Rodriguez M, Fernandez-Aparicio A, Gonzalez-Jimenez E. Waist Circumference and Abdominal Volume Index Are the Strongest Anthropometric Discriminators of Metabolic Syndrome in Spanish Adolescents. *Eur J Clin Invest* (2019) 49(3):e13060. doi: 10.1111/eci.13060
31. Cristine Silva K, Santana Paiva N, Rocha de Faria F, Franceschini S, Eloiza Piore S. Predictive Ability of Seven Anthropometric Indices for Cardiovascular Risk Markers and Metabolic Syndrome in Adolescents. *J Adolesc Health* (2020) 66(4):491–8. doi: 10.1016/j.jadohealth.2019.10.021
32. Rico-Martin S, Calderon-Garcia JF, Sanchez-Rey P, Franco-Antonio C, Martinez Alvarez M, Sanchez Munoz-Torrero JF. Effectiveness of Body Roundness Index in Predicting Metabolic Syndrome: A Systematic Review and Meta-Analysis. *Obes Rev* (2020) 21(7):e13023. doi: 10.1111/obr.13023
33. Moon K, Sung SH, Chang YK, Park IK, Paek YM, Kim SG, et al. [The Association Between Apolipoprotein E Genotype and Lipid Profiles in Healthy Woman Workers. *J Prev Med Public Health* (2010) 43(3):213–21. doi: 10.3961/jpmph.2010.43.3.213

34. Cao C, Liu Q, Yang L, Zheng X, Lan P, Koyanagi A, et al. Handgrip Strength Is Associated With Suicidal Thoughts in Men: Cross-Sectional Analyses From Nhanes. *Scand J Med Sci Sports* (2020) 30(1):92–9. doi: 10.1111/sms.13559
35. Hu G, Jia G, Tang S, Zheng P, Hu L. Association of Low-Level Blood Lead With Serum Uric Acid in U.S. Adolescents: A Cross-Sectional Study. *Environ Health* (2019) 18(1):86. doi: 10.1186/s12940-019-0524-0
36. Li X, Li L, Yang L, Yang J, Lu H. No Association Between Serum Uric Acid and Lumbar Spine Bone Mineral Density in Us Adult Males: A Cross Sectional Study. *Sci Rep* (2021) 11(1):15588. doi: 10.1038/s41598-021-95207-z
37. Fleming E, Singhal A. Chronic Disease Counseling and Screening by Dental Professionals: Results From Nhanes, 2011-2016. *Prev Chronic Dis* (2020) 17: E87. doi: 10.5888/pcd17.200152
38. Jaddoe VW, de Jonge LL, Hofman A, Franco OH, Steegers EA, Gaillard R. First Trimester Fetal Growth Restriction and Cardiovascular Risk Factors in School Age Children: Population Based Cohort Study. *BMJ* (2014) 348:g14. doi: 10.1136/bmj.g14
39. Kahn HS. The Lipid Accumulation Product Is Better Than Bmi for Identifying Diabetes: A Population-Based Comparison. *Diabetes Care* (2006) 29(1):151–3. doi: 10.2337/diacare.29.1.151
40. Hosseinpanah F, Barzin M, Mirbolouk M, Abtahi H, Cheraghi L, Azizi F. Lipid Accumulation Product and Incident Cardiovascular Events in a Normal Weight Population: Tehran Lipid and Glucose Study. *Eur J Prev Cardiol* (2016) 23(2):187–93. doi: 10.1177/2047487314558771
41. Zhou C, Zhan L, Yuan J, Tong X, Peng Y, Zha Y. Comparison of Visceral, General and Central Obesity Indices in the Prediction of Metabolic Syndrome in Maintenance Hemodialysis Patients. *Eat Weight Disord* (2020) 25(3):727–34. doi: 10.1007/s40519-019-00678-9
42. Motamed N, Razmjou S, Hemmasi G, Maadi M, Zamani F. Lipid Accumulation Product and Metabolic Syndrome: A Population-Based Study in Northern Iran, Amol. *J Endocrinol Invest* (2016) 39(4):375–82. doi: 10.1007/s40618-015-0369-5
43. Taverna MJ, Martinez-Larrad MT, Frechtel GD, Serrano-Rios M. Lipid Accumulation Product: A Powerful Marker of Metabolic Syndrome in Healthy Population. *Eur J Endocrinol* (2011) 164(4):559–67. doi: 10.1530/EJE-10-1039
44. Li R, Li Q, Cui M, Yin Z, Li L, Zhong T, et al. Clinical Surrogate Markers for Predicting Metabolic Syndrome in Middle-Aged and Elderly Chinese. *J Diabetes Investig* (2018) 9(2):411–8. doi: 10.1111/jdi.12708
45. Pekgor S, Duran C, Berberoglu U, Eryilmaz MA. The Role of Visceral Adiposity Index Levels in Predicting the Presence of Metabolic Syndrome and Insulin Resistance in Overweight and Obese Patients. *Metab Syndr Relat Disord* (2019) 17(5):296–302. doi: 10.1089/met.2019.0005

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Advantages of publishing in Frontiers

OPEN ACCESS
Articles are free to read for greatest visibility and readership

FAST PUBLICATION
Around 90 days from submission to decision

HIGH QUALITY PEER-REVIEW
Rigorous, collaborative, and constructive peer-review

TRANSPARENT PEER-REVIEW
Editors and reviewers acknowledged by name on published articles

Frontiers
Avenue du Tribunal-Fédéral 34
1005 Lausanne | Switzerland

Visit us: www.frontiersin.org
Contact us: frontiersin.org/about/contact

REPRODUCIBILITY OF RESEARCH
Support open data and methods to enhance research reproducibility

DIGITAL PUBLISHING
Articles designed for optimal readership across devices

FOLLOW US
@frontiersin

IMPACT METRICS
Advanced article metrics track visibility across digital media

EXTENSIVE PROMOTION
Marketing and promotion of impactful research

LOOP RESEARCH NETWORK
Our network increases your article's readership