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Childhood obesity: Prevention, management and new insight in pathophysiology

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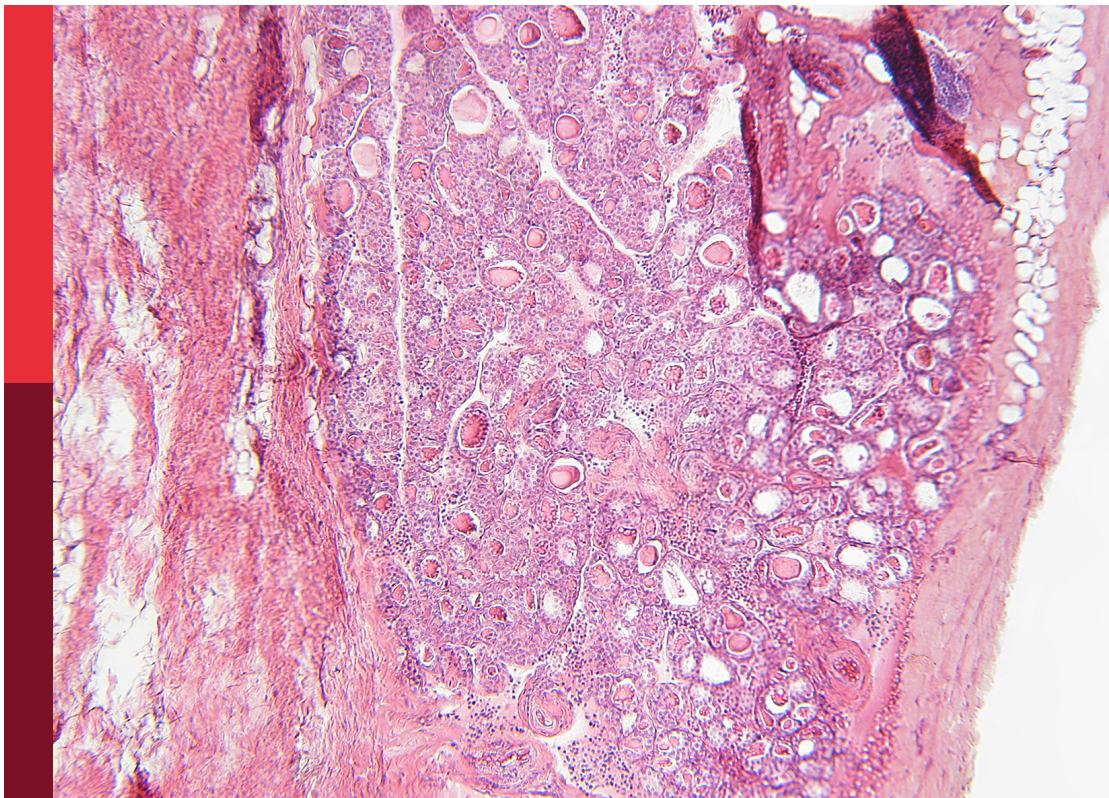

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Childhood obesity: Prevention, management and new insight in pathophysiology

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Editorial: Childhood obesity: prevention, management and new insight in pathophysiology

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Editorial on the Research Topic
Childhood obesity: prevention, management and new insight in pathophysiology

Childhood obesity (CO) has become a major health problem worldwide and its incidence is steadily increasing (1, 2). Obesity is associated with a wide spectrum of both cardio-metabolic and psychological-behavioural complications that can occur as early as childhood and adolescence and can lead to a deterioration in the quality and perspective of adult life (3).

This Research Topic includes thirteen studies evaluating different aspects of CO. Specifically, six studies, both experimental and review studies, evaluate epidemiological aspects and risk factors related to childhood obesity; four studies analyze possible biochemical markers related to the pathogenesis of obesity-related complications; three studies evaluate psychological-behavioural aspects in children and adolescents with CO.

In recent decades, lifestyle changes, mainly characterized by an increase in sedentariness and unhealthy eating habits, are among the main culprits for the growing incidence of CO. In this context, the COVID-19 pandemic has undoubtedly amplified these aspects worldwide, further accelerating the spread of CO. Yang et al. evaluated the real-world national trends of obesity prevalence of Chinese children between the years 2017-2021 to assess the impact of COVID-19 pandemic on pediatric obesity. Through the collection of hospital and parent-reported data, the authors assessed the prevalence of obesity/overweight in the pre-pandemic and pandemic periods and changes in the BMI z-score during the COVID-19 lockdown in a large number of subjects. Authors demonstrated a relatively stable trend in the prevalence of CO in the pre-pandemic period and a significant increase in the prevalence during the COVID-19 pandemic; in particular, a high increase in the BMI z-score among primary and secondary school children, especially in specific regions, was documented. The authors concluded that these data support a targeted intervention, including through mobile growth assessment based on parent-reported, to prevent the spread of obesity especially during pandemic periods in China.

The spread of CO over the years has been accompanied by a lowering of its onset age. The diagnosis of simple obesity is often made as early as the first 3-5 years of life. However, in this context, attention should always be kept on those possible cases of obesity secondary to other causes, such as genetic obesities. This is the goal of the multicenter research project by Mierzwa et al. who aimed to create a Polish database of severely obese children and adolescents and to assess the prevalence of monogenic forms of obesity in this cohort, with a focus on abnormalities in the leptin-propylanocortin pathway. Another aspect related to causes of secondary obesity is assessed by the research of Hetman et al., who evaluate the application of standard or specific growth charts for early identification of growth disorders, including obesity, in children with Down syndrome.

CO, especially severe and early-onset obesity, is known to be related to an increased risk of developing cardio-metabolic complications, such as altered glucose and lipid profile, non-alcoholic fatty liver disease (NAFLD), hypertension. Abdominal obesity is one of the factors primarily related to these complications. Liu et al. assessed the change over time in the prevalence of abdominal obesity in a U.S. population, documenting that, although there was no significant increase in the prevalence of abdominal obesity in the entire population, some ethnic groups did show a significant increase, which therefore warrant further attention.

Due to the increasing incidence of CO, NAFLD has become the most common hepatopathy in pediatric age. The diagnostic gold-standard is liver biopsy, which, however, is burdened by several limitations, including invasiveness and costs. Therefore, ultrasonography, sometimes combined with elastography, is the most widely used noninvasive method to identify hepatic steatosis, although this method also has limitations (4). Marcinkiewicz et al. in their study, using liver ultrasound, identified NAFLD in approximately 23% of subjects in a pediatric population, with a higher prevalence among subjects with glucose intolerance. Therefore, liver ultrasonography can play a role, at least a preliminary one, in diagnosis and follow-up of NALFD in CO.

In addition to the diagnosis and subsequent tailored therapeutic approach to CO, the task of the scientific community and health professionals is to identify the causes promoting the onset of CO in order to implement preventive actions to curb this growing health problem. Factors influencing the onset of CO include both environmental factors (including nutritional aspects) and, in general, the individual's lifestyle. However, the possible influence of pre-natal factors should also be considered. Several evidence showed that the origins of CO can be as early as maternal pregnancy. In a systematic review and meta-analysis, Yan et al. suggested that gestational hypertension and preeclampsia might be associated with obesity in the offspring. However, both possible prenatal (e.g., maternal factors) and postnatal confounding factors able to influence the onset of obesity and aspects related to hormonal adaptations of the fetus exposed to stressors, should always be considered.

A significant part of the scientific research concerning CO, is directed toward the investigation of new markers as predictors of obesity and its complications.

Cystatin C, a non-glycosylated protein filtered by the glomerulus, has been linked to obesity-related complications (e.g., NAFLD, vascular alterations) mainly in adult populations. In a Chinese cross-sectional study, Huo et al. analyzed the association between cystatin C and overweight or obesity in adolescence. In this study, authors suggested a potential role of serum cystatin C levels as an indicator of early obesity risk in adolescents, although further studies will be needed to confirm the preliminary results of this study.

To assess the risk of cardiovascular disease in obese children, Tumor Necrosis Factor Weak Inducer of Apoptosis (TWEAK), also known as Tumor Necrosis Factor (TNF) ligand superfamily member 12, probably involved in insulin resistance, may be used. Cluster of Differentiation 163 (CD163) is a macrophage-specific protein that has been identified as a scavenger receptor of TWEAK, promoting its degradation. CD163 is described as a strong predictor of type 2 diabetes in adults. In cardiovascular diseases, a high CD163/TWEAK ratio was found. The study by Escartin et al. is the first one to observe the evolution of these parameters in prepubertal children, supporting the hypotheses that these cytokines probably play a role in childhood obesity.

Endoplasmic reticulum (ER) stress proteins could play a role as early markers of metabolic alterations in childhood and adolescence. The results of a multicenter study from Italy (Antoniotti et al.) suggested the existence of an important link between ER stress and metabolic changes behind obesity complications even in pediatric age. These preliminary data suggest that Calreticulin (CALR) and PDIA3, two key molecules of ER stress, could be related to insulin resistance and altered lipid profile in pediatric obesity. These authors suggested that CALR and PDIA3 could be early markers of insulin resistance and dyslipidemia ER stress-related, useful to stratify patients at higher risk of further complications.

He et al. evaluated sex differences in insulin resistance-induced changes in metabolic and inflammatory markers in school-age children with overweight and obesity. These authors demonstrated an elevated white blood cell count and absolute neutrophil count in children with overweight and obesity, especially in girls, and a strong association of these parameters with HOMA-IR. Therefore, they suggested that these findings can be markers of insulin resistance.

Psychological-behavioral complications related to CO were also evaluated in this Research topic. Wojcik et al., in the original research project studied the prevalence of depressive symptoms and anxiety in adolescents with obesity and their caregivers. The results of the study showed that in childhood obesity, anxiety disorders are much more important than depressive disorders, both for patients and their parents. These authors concluded that childhood obesity, like any chronic disease, could have a significant impact on the emotional state of children and adolescents as well as the possibility of realizing interests and spending free time.

Second paper of this group is "A Brief Literature Review of the Role of Social Media in Body Image Shaping and Eating Patterns among Children and Adolescents" by Modrzejewska et al. The careful search of the literature had revealed only 8 articles for the analysis of this topic. The data analysis showed that social media may have a strong influence on the development of eating patterns and body image in children and adolescents, which in turn could be one of the risk factors for the development of obesity. Due to their great influence on youth, social media should be used as a resource for the prevention and treatment of obesity.

Finally, in a mini review Shi et al. evaluated mechanisms underlying the link between central precocious puberty and obesity, both from metabolic and cognitive-behavioral perspectives.

The studies in this Research Topic provide valuable evidence on several aspects regarding CO, providing input for further research on the topic. In conclusion, numerous factors (genetic, environmental, sociodemographic, behavioral, and perinatal) may contribute to the onset of obesity and its complications, although to date their exact role, interaction, and mechanisms involved in this process are not completely understood. Early intervention is an important strategy to prevent the onset of obesity, because there are crucial times during children's growth and development when preventive efforts or therapeutic interventions can be most effective.

Author contributions

All authors planned the outline of the editorial. DC, IB-S and MW drafted the manuscript. All authors contributed to manuscript revisions and approved the final version.

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.

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References

1. Twig G, Yaniv G, Levine H, Leiba A, Goldberger N, Derazne E, et al. Body-mass index in 2.3 million adolescents and cardiovascular death in adulthood. *N Engl J Med* (2016) 374(25):2430–40. doi: 10.1056/NEJMoa1503840

2. Skinner AC, Ravanbakht SN, Skelton JA, Perrin EM, Armstrong SC. Prevalence of obesity and severe obesity in US children, 1999-2016. *Pediatrics* (2018) 141(3). doi: 10.1542/peds.2017-3459

3. Franks PW, Hanson RL, Knowler WC, Sievers ML, Bennett PH, Looker HC. Childhood obesity, other cardiovascular risk factors, and premature death. *N Engl J Med* (2010) 362(6):485–93. doi: 10.1056/NEJMoa0904130

4. Corica D, Bottari A, Aversa T, Morabito LA, Curatola S, Alibrandi A, et al. Prospective assessment of liver stiffness by shear wave elastography in childhood obesity: a pilot study. *Endocrine* (2022) 75(1):59–69. doi: 10.1007/s12020-021-02828-5

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ORIGINAL RESEARCH
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Serum Cystatin C Levels Are Associated With Obesity in Adolescents Aged 14–17 Years

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Background: The association between serum cystatin C levels and obesity has not been fully explored in adolescents. This study aimed to explore the association between serum cystatin C levels and obesity in adolescents of different sexes.

Methods: We conducted a cross-sectional study including 481 adolescents aged 14–17 years. Cystatin C level was measured by immunoassay. Health examinations data, biochemical parameters, and questionnaire information were collected. The restricted cubic spline model analyzed the association between cystatin C levels and obesity in boys and girls.

Results: Boys exhibited significantly higher cystatin C levels than girls, with a mean level of 0.97 ± 0.10 mg/L in boys and 0.86 ± 0.09 mg/L in girls ($P < 0.001$). The restricted cubic spline model suggested that low or high cystatin C levels were associated with an increased risk of obesity in boys, whereas only higher cystatin C levels were associated with an increased risk of obesity in girls.

Conclusions: A U-shaped correlation was observed between serum cystatin C levels and the risk of obesity in boys. However, in girls, the risk of obesity showed a trend of initially increase and then decrease with increasing cystatin C levels. Longitudinal studies should be conducted to further investigate the diagnostic potential of cystatin C in the progression of early obesity in adolescents of different sexes.

Keywords: cystatin C, obesity, adolescent, boys, girls

Abbreviations: BMI, Body mass index; WC, waist circumference; WHtR, waist-to-height ratio; TG, triglycerides; HDL, high-density lipoprotein; LDL, low-density lipoprotein; TC, total cholesterol; UA, uric acid; FPG, fasting plasma glucose; SBP, systolic blood pressure; DBP, diastolic blood pressure; OR, odds ratio; CI, confidence interval; RCS, restricted cubic spline; eGFR, estimated glomerular filtration rate.

INTRODUCTION

Overweight and obesity pose a great threat to population health in various countries. Obesity is one of the significant risk factors for many noncommunicable diseases, including hypertension, dyslipidemia, and some types of cancer (1). However, a large proportion of obesity in adults stems from obesity in childhood. Weight gain during childhood and adolescence is likely to contribute to overweight and obesity for life (2). Meanwhile, overweight or obesity in childhood and adolescence can significantly increase the incidence of adult obesity, as well as obesity-related disease morbidity and early mortality (3). The morbidity rate of obesity in children increased from 4% in 1975 to more than 18% in 2016 (4). A total of 50 million girls and 74 million boys developed obesity worldwide in 2016 (5). More than 50% of adults and nearly 20% of children and adolescents developed overweight or obesity in 2019, according to the latest national data from China (6). Obesity in children also leads to many health problems, including hypertension, high total cholesterol, and impaired glucose tolerance (7). Obesity in children has become a serious public health issue in various countries around the world (8). Therefore, early detection and prompt treatment of children and adolescents with obesity are very important to avoid or delay the occurrence of obesity and chronic diseases in adults.

Cystatin C is an essential protein that is non-glycosylated and filtered by the glomerulus (9). It is considered as an indicator for evaluating renal function (10). Data suggest a potential genetic link between nonalcoholic fatty liver disease and chronic kidney disease in children (11). Studies on adults have reported a positive correlation between cystatin C level and body mass index (BMI) or waist circumference (WC) (12). Further, some data suggested that increased cystatin C levels could be regarded as an early prognostic indicator of vascular risk in children with obesity (13). Many studies have been conducted on cystatin C levels in adults, but few on the association between cystatin C level and overweight or obesity in adolescents. Currently, no normal reference value range exists for cystatin C levels in adolescents. Thus, the primary goal of this cross-sectional study was to investigate the association between cystatin C and overweight or obesity in adolescence. It used the serum cystatin C level as an indicator of early obesity in adolescents to promote early preventive intervention for overweight or obesity so as to reduce the incidence of obesity and obesity-related diseases in adults.

MATERIALS AND METHODS

Study Population

In this study, all participants were selected by random cluster sampling from a high school in Huanggu District, Shenyang City, Liaoning Province. A cross-sectional survey was conducted on health examinations, biological sample collection, and questionnaire administered to these adolescents. Thus, 481 adolescents whose ages ranged from 14 to 17 years were included.

The inclusion criteria were adolescents with complete anthropometric and biospecimen data. We excluded adolescents with kidney disease, endocrine disease, autoimmune disease, and infectious disease as well as those taking any medication by asking the medical history. Furthermore, we identified and excluded one isolated extreme value of cystatin C level (0.39 mg/L).

Health Examinations

Trained investigators performed measurements under standard conditions and uniform specifications. Anthropometric measurements were recorded using a fully automated electronic scale (equipment and methods were referenced to GB/T26343), and the instruments used were uniform and calibrated.

Height and weight were measured with the adolescents without shoes and wearing light clothes. Height was measured with the adolescents standing on the bottom plate of the human body altimeter. Weight was measured with the adolescents standing on at the center of the weighing scale. The readings were recorded in "cm" and "kg" and recorded to one decimal place. The BMI was equal to weight divided by height squared (kg/m^2), retained to one decimal place. The WC was measured with the adolescents standing naturally and with a nonelastic tape around the central level of the umbilicus from the end of exhalation. It was recorded in "cm" and read to one decimal place. The waist-to-height ratio (WHtR) was equal to WC (cm) divided by height (cm), without unit.

The blood pressure was measured using a Riva-Rocci sphygmomanometer. An L-type (32–42 cm) or XL type (42–50 cm) cuff model was used according to the upper arm circumference of children. The study participants should not engage in any strenuous exercise within 1 h before the measurement. The bladder was emptied and the participants sat still for more than 10 min before measuring blood pressure. The blood pressure was measured three consecutive times for each adolescent, with no less than 30-s intervals between each measurement. The values were averaged after removing outliers and recorded as mmHg.

Biological Sample Collection

The blood samples were collected from all adolescents on an early morning fast. The biochemical measurement analyses were performed by the Department of Laboratory Medicine of the Shengjing Hospital Affiliated to China Medical University. The cystatin C level was analyzed with an automated particle-enhanced turbidity immunoassay using the Architect I16200 automated analyzer (Architect, Shandong, China). In addition, fasting plasma glucose (FPG), triglycerides (TG), uric acid (UA), high-density lipoprotein (HDL), low-density lipoprotein (LDL), and total cholesterol (TC) were measured with a Hitachi 7600 clinical analyzer (Hitachi, Tokyo, Japan). Calculate estimated glomerular filtration rate (eGFR) using the eGFR = k * patient's length (cm)/serum creatinine (mg/dL) formula (14).

Questionnaire Administered

The surveys were conducted during face-to-face interviews at the school by trained interviewers. Structured questionnaire was administered to collect data on demographic information (name, age, sex, ethnicity, address, and number of family members), daily dietary intake (eating attitudes, dietary control, eating behaviors, and food preferences), and physical activity information (daily activities and sports activities within 7 days).

Approval was obtained from the ethics committee of Dalian Medical University. Informed consent was obtained from all adolescent and their parents.

Definitions of Obesity

The BMI of participants was defined as normal, overweight, or obesity according to the Chinese reference norms for overweight and obesity in children and adolescents (15).

Children and adolescents with abdominal obesity were identified using the WC ≥90th percentile value for age and sex according to the standard definition of overweight and obesity in children worldwide by the International Obesity Working Group (16).

The value of WHtR ≥0.5 was suggested to determine central obesity for children (17).

Hypertension was defined as a mean systolic blood pressure (SBP) and/or diastolic blood pressure (DBP) at ≥95th percentile for age, sex, and height on ≥3 times (18).

Statistical Analysis

Mean and standard deviations were used to represent normal distributions for continuous variables. Medians and interquartile ranges were used to define skewed distributions. In contrast, categorical variables were described as counts and percentages. Adolescents were analyzed stratified by sex. The means of cystatin C levels in groups with obesity and groups without obesity defined by the three criteria (BMI, WC, and WHtR) were analyzed using the *t*-test. The factors independently associated with obesity were identified by univariate logistic regression. Trend tests for cystatin C quartile levels with obesity risk were also conducted, resulting in odds ratio (OR) and 95% confidence interval (CI). ORs at other levels were calculated with binary logistic regression using the lowest quartile of cystatin C as a reference. We used restricted cubic spline (RCS) models fitted for linear regression models and logistic regression models. Knots were present at 10th, 50th, and 90th percentiles, the reference cystatin C level of the logistic regression model was the median. The RCS based on the linear regression model was used to model the association of cystatin C level and BMI-for-age Z-score (19).

Differences were considered statistically significant when the *P* value was less than 0.05. Statistical analysis was performed on R studio version 3.6.1 software (Copyright 2007 Free Software Foundation).

RESULTS

General Information

A total of 481 participants, 199 boys (41.4%) and 282 girls (58.6%), were included, aged between 14 and 17 years. The cystatin C level obeyed a normal distribution with the mean levels in all adolescents being 0.90 ± 0.11 mg/L (**Table 1**). Boys exhibited significantly higher levels of cystatin C level than girls, with a mean level of 0.97 ± 0.10 mg/L in boys and 0.86 ± 0.09 mg/L in girls ($P<0.001$). The data showed that age was not significantly different ($P = 0.330$).

As shown in **Table 2**, we defined obesity using three criteria (BMI, WC, and WHtR). We analyzed the differences in cystatin C levels between adolescents with obesity and adolescents without obesity of different sexes. The prevalence was 30.6% for overweight/obesity defined by BMI, 21.7% for abdominal obesity defined by the WC value, and 31.6% for obesity defined by the WHtR ≥0.5. In obesity defined by the BMI, higher cystatin C levels were observed in adolescents with obesity ($P = 0.005$). Girls with obesity had higher cystatin C levels compared with girls with normal BMI ($P = 0.003$). In contrast, the difference was not significant in boys ($P = 0.703$). However, in both abdominal obesity defined by the WC and obesity defined by the WHtR ≥0.5, no statistically significant differences in cystatin C levels were found between boys with obesity and boys without obesity or between girls with obesity and girls without obesity.

Univariate Analysis of Traditional Blood Biochemical Parameters Related to BMI-Defined Obesity

Table 3 shows the blood biochemical parameters of traditional risk factors for obesity in adolescents stratified by sex. In boys, TG, HDL, LDL, UA, TC, and SBP were found to be associated with the obesity risk in adolescents. However, in girls, age, TG,

TABLE 1 | Distribution of cystatin C levels in different sex and age.

Variable	Category	N (%)	CysC (mg/L)		Statistic	P
			Mean	SD		
total	——	481	0.904	0.110	——	——
sex	boys	199 (41.4)	0.965	0.104	t = 11.787	<0.001
	girls	282 (58.6)	0.860	0.091		
age	14	13 (2.7)	0.875	0.079	F = 1.145	0.330
	15	142 (29.5)	0.916	0.112		
	16	247 (51.4)	0.901	0.104		
	17	79 (16.4)	0.894	0.124		

CysC, cystatin C; SD, standard deviation.

TABLE 2 | Association between cystatin C levels and obesity as defined by three criteria: BMI, WC, and WHtR.

Variable	Category	Total					Boys					Girls				
		N (%)	CysC (mg/L)		Statistic	P	N (%)	CysC (mg/L)		Statistic	P	N (%)	CysC (mg/L)		Statistic	P
			Mean	SD				Mean	SD				Mean	SD		
BMI	Normal	334 (69.4)	0.894	0.110	t= -2.823	0.005	129 (64.8)	0.963	0.098	t= -0.382	0.703	205 (72.7)	0.851	0.094	t= -2.991	0.003
	overweight/ obesity	147 (30.6)	0.925	0.106			70 (35.2)	0.969	0.115			77 (27.3)	0.884	0.078		
WC	Normal	372 (78.3)	0.904	0.106	t=0.270	0.787	165 (83.8)	0.963	0.096	t= -0.363	0.719	207 (74.5)	0.858	0.089	t= -0.888	0.377
	obesity	103 (21.7)	0.901	0.118			32 (16.2)	0.972	0.137			71 (25.5)	0.869	0.093		
WHtR	normal	325 (68.4)	0.901	0.095	t= -0.846	0.398	135 (68.5)	0.964	0.095	t= -0.111	0.912	190 (68.3)	0.856	0.091	t= -1.252	0.212
	obesity	150 (31.6)	0.910	0.120			62 (31.5)	0.966	0.120			88 (31.7)	0.871	0.089		

CysC, cystatin C; SD, standard deviation; BMI, body mass index; WC, waist circumference; WHtR, waist to height ratio.

HDL, UA, and SBP were associated with the obesity risk ($P < 0.05$).

Tendency Test of Cystatin C Quartile Levels With Obesity Defined by BMI

Table 4 presents the associations between cystatin C quartiles and the risk of obesity in adolescents, stratified by sex. Model 1 adjusted for age in different sexes. Model 2 adjusted for age, TG, TC, HDL, LDL, UA, and SBP in boys, but it adjusted for age, TG, HDL, UA, and SBP in girls.

The boys, had a trend of increasing obesity risk with increasing cystatin C levels. In model 2, taking the cystatin C first quartile level as the reference group, the risk of obesity was reduced by 10% at the second quartile level (OR = 0.90, 95% CI: 0.34–2.39, $P = 0.826$), while it was increased by 25% at the third quartile level (OR = 1.25, 95% CI: 0.49–3.19, $P = 0.646$) and 38% at the fourth quartile level (OR = 1.38, 95% CI: 0.50–3.82, $P = 0.539$) cystatin C levels. However, this growth trend was not statistically significant ($P > 0.05$).

In contrast, in girls, obesity risk showed a decreasing trend with increasing cystatin C levels. In model 2, taking the cystatin C first quartile level as the reference, the risks of obesity were elevated by four-, three-, and two-fold in the second quartile (OR = 3.85, 95% CI: 1.46–8.13, $P = 0.006$), third quartile (OR = 2.90, 95% CI: 1.06–7.97, $P = 0.038$), and fourth quartile (OR = 2.45, 95% CI: 0.90–6.65, $P = 0.078$) cystatin C levels, respectively.

Dose-Response Association of the Cystatin C Level With Obesity Defined by BMI Based on RCS Models

We used RCS to explore the dose-response association between cystatin C level and obesity. The association between BMI-of-age Z-score and cystatin C levels on a continuous scale was U-shaped in boys (**Figure 1A**). We estimated the BMI-of-age Z-score to reach a nadir at cystatin C of 0.95mg/L, with inverse associations below and positive associations above ($P_{\text{nonlinear}} = 0.037$). However, in girls, a positive association was found between BMI-of-age Z-score and cystatin C level (**Figure 1B**). That is, BMI-of-age Z-score gradually increased with increasing cystatin C levels ($P_{\text{nonlinear}} = 0.806$).

Regarding the strong U-shaped relationship between the risk of obesity and cystatin C levels in boys, the plot showed a decreasing risk trend in the lower range of predicted cystatin C

TABLE 3 | Univariate analysis of obesity risk factors in boys with obesity and girls with obesity defined by BMI criteria.

Variable	Boys			Girls		
	OR	95% CI	*P*	OR	95% CI	*P*
age	1.142	[0.748, 1.745]	0.539	0.702	[0.495, 0.995]	0.047
TG	3.245	[1.563, 6.738]	0.002	2.623	[1.410, 4.879]	0.002
HDL	0.126	[0.036, 0.445]	0.001	0.302	[0.124, 0.738]	0.009
LDL	3.082	[1.711, 5.553]	<0.001	1.480	[0.957, 2.290]	0.078
FPG	1.277	[0.690, 2.363]	0.436	0.777	[0.356, 1.695]	0.526
UA	1.010	[1.006, 1.015]	<0.001	1.012	[1.007, 1.017]	<0.001
SBP	1.061	[1.035, 1.089]	<0.001	1.064	[1.038, 1.091]	<0.001
DBP	1.013	[0.983, 1.044]	0.385	1.022	[0.994, 1.052]	0.129
TC	1.898	[1.192, 3.021]	0.007	1.296	[0.881, 1.906]	0.188
eGFR	1.008	[0.973, 1.043]	0.660	0.993	[0.972, 1.014]	0.505

OR, odds ratio; 95% CI, 95% confidence interval; TG, triglyceride; HDL, high density lipoprotein; LDL, low density lipoprotein; FPG, fasting plasma glucose; UA, uric acid; SBP, systolic blood pressure; DBP, diastolic blood pressure; TC, total cholesterol; eGFR, estimated glomerular filtration rate.

TABLE 4 | Analysis of the strength of association between each level of cystatin C quartile by sex and obesity defined by BMI as a criterion in adolescents.

Boys					Girls				
CysC (mg/L)	Model 1		Model 2[a]		CysC (mg/L)	Model 1		Model 2[b]	
	OR (95%CI)	*P*	OR (95%CI)	*P*		OR (95%CI)	*P*	OR (95%CI)	*P*
Quartile 1 (0.67-0.90)	1.00	——	1.00	——	Quartile 1 (0.60-0.80)	1.00	——	1.00	——
Quartile 2 (0.91-0.96)	0.922 (0.397-2.143)	0.850	0.896 (0.336-2.387)	0.826	Quartile 2 (0.81-0.86)	4.374 (1.826-10.480)	0.001	3.847 (1.461-10.134)	0.006
Quartile 3 (0.97-1.04)	1.130 (0.509-2.506)	0.764	1.246 (0.487-3.187)	0.646	Quartile 3 (0.87-0.92)	3.632 (1.459-9.043)	0.006	2.904 (1.058-7.970)	0.038
Quartile 4 (1.05-1.27)	1.570 (0.675-3.653)	0.295	1.377 (0.497-3.817)	0.539	Quartile 4 (0.93-1.10)	3.396 (1.384-8.336)	0.008	2.451 (0.904-6.646)	0.078

Model 1: adjusted for age.
Model 2: a. adjusted for model 1+ TG+ TC+ HDL+ LDL+ UA and SBP.
b. adjusted for model 1+ TG+ HDL+UA and SBP.
CysC, cystatin C; UA, uric acid; TC, total cholesterol; SBP, systolic blood pressure; TG, triglyceride; HDL, high density lipoprotein; LDL, low density lipoprotein; OR, odds ratio; 95% CI, 95% confidence interval.

levels, reaching a minimum risk around 0.97 mg/L and then increasing ($P_{\text{nonlinear}} = 0.478$). Below 0.97 mg/L, the OR per standard deviation higher predicted cystatin C levels was 1.45 (0.71–1.84). Above 0.97 mg/L, the OR per standard deviation higher predicted cystatin C levels 1.12 (0.70–1.78) (**Figure 2A**). The risk of obesity showed an increasing trend with elevated cystatin C levels before the cystatin C level reached 0.91 mg/L in girls, with the highest risk of obesity (OR = 1.07, 95% CI: 0.88–

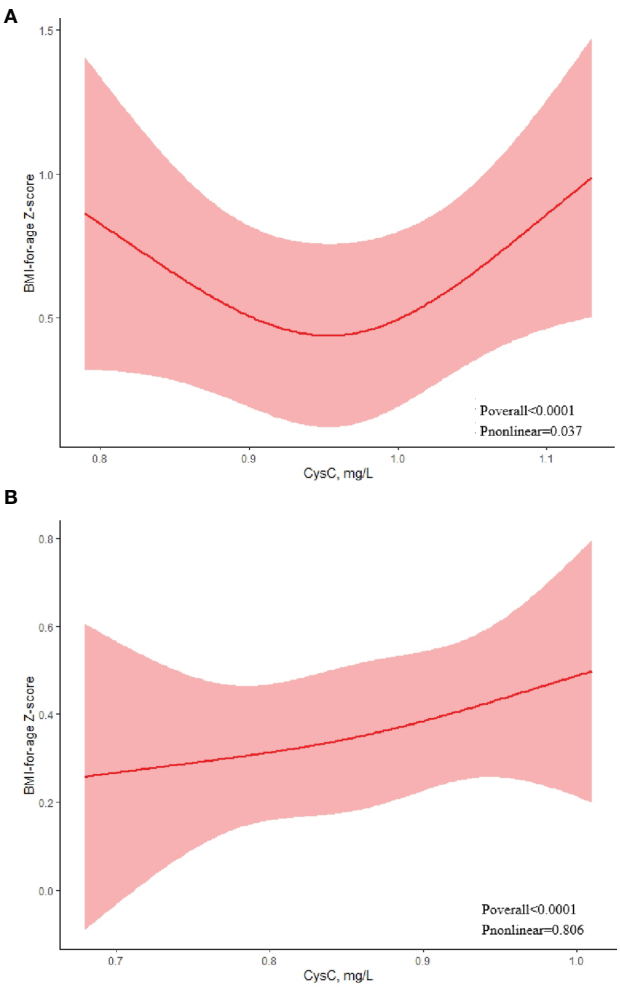

FIGURE 1 | Association between serum cystatin C and BMI-for-age Z-score using a restricted cubic spline regression model in boys **(A)** and girls **(B)**.

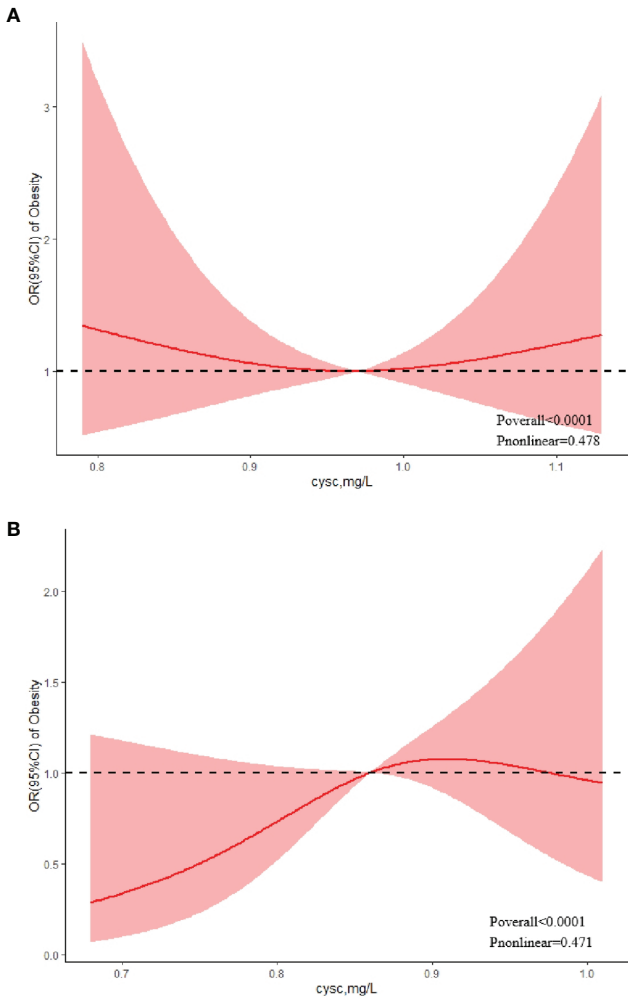

FIGURE 2 | Association between serum cystatin C and risk of obesity using a restricted cubic spline regression model in boys **(A)** and girls **(B)**.

1.31) at the cystatin C levels of 0.91 mg/L (**Figure 2B**). After that, the risk of obesity tended to decline ($P_{\text{nonlinear}} = 0.471$).

DISCUSSION

The present study proposed that the cystatin C level in adolescents was associated with obesity, particularly obesity defined by BMI. These data suggested that the cystatin C level could be considered as an early biomarker of obesity in adolescents. There was a U-shaped association between cystatin C levels and obesity in boys, and the risk of obesity showed a trend of first decrease and then increase as cystatin C levels increased, and either low or high cystatin levels were associated with an increased risk of obesity. Whereas in girls, the risk of obesity showed a trend of initially increase and then decrease with increasing cystatin C levels.

The study found that boys had higher cystatin C levels than girls, which was consistent with other findings that boys had higher cystatin C levels than girls after 1 year of age (20). The possible reason for the result might be that adolescent boys had a higher increase in muscle mass than girls (20). In addition, renal factors, hormonal factors, or other factors interfering with the determination of the cystatin C level in individual girls might also explain sex-related differences (21). Several findings suggested that individuals began to produce more cystatin C when they were around 10–13 years of age. After the age of 13 years, the individual's cystatin C production reached a peak. It then remained stable until around the age of 50 years (22). Considering the particularity of children's growth and development, and that the distribution of cystatin C did differ between boys and girls, we analyzed sex as a stratification factor in the next analysis. About 80% of adolescents with obesity still obese in adulthood, and hence early preventive interventions for obesity are warranted, however, childhood BMI is less sensitive

to predict adult obesity (23), so we need to find other more sensitive indicators for detecting obesity in adolescents. Cystatin C is an effective cathepsin-proteolytic enzyme inhibitor (24). It can be used to calculate the eGFR (25). Studies have also shown a significant association between GFR and duration of obesity (26). However, in our study, no relationship between eGFR and obesity was found. The present study showed that the elevated circulating cystatin C levels in study participants with obesity may be to counteract obesity-related inflammation (27). Elevated cystatin C levels in adolescence were also associated with lipid metabolic dysregulation and cardiometabolic risk (28). The production of cystatin C in adipose tissue can lead to an increase in the concentration of cystatin C in the blood of children with obesity (29). The studies demonstrated that cystatin C level was associated with WC, BMI, and visceral fat (30–32). However, our study only found a relationship between BMI-defined obesity and cystatin C levels. It also provided a rationale for using cystatin C as a biomarker to predict overweight and obesity in adolescents.

Our study found a U-shaped association between cystatin C levels and obesity in boys, with a decreasing risk of obesity before cystatin C values reached 0.97 mg/L and then increasing; however, in girls, the risk of obesity increased before the cystatin C value reached 0.91 mg/L and then tended to decrease. The different associations we observed between the cystatin C levels and obesity risk of different sexes were interesting. Since the age distribution of our study population was in adolescence (14 – 17 years), this difference may have resulted from different levels of pubertal development in boys and girls. To our knowledge, no study had found this U-shaped association of cystatin C levels with the risk of obesity in boys. Therefore, we hope that prospective studies with larger sample sizes will be followed to confirm the differences of different cystatin C levels with obesity risk among boys and girls. The typical reference range in adults for Cystatin cystatin C was 0.40–1.10 mg/L (33). However, no explicit reference value range exists for children and adolescents. Whether normal cystatin C values ranges for boys and girls separately need to be developed in the future for early diagnosis of obesity remains to be explored.

This analysis had several limitations. First, the adolescent population we studied was obtained by cluster sampling and is not nationally representative. Second, critical adverse factors, such as pubertal development and family history, as well as insulin resistance levels, were not considered in our analysis due to limited data. Population-based prospective studies should be carried out to overcome these limitations and comprehensively understand the cystatin C levels in adolescents with obesity and adolescents without obesity.

In conclusion, this study suggested a U-shaped association between serum cystatin C levels and obesity in boys, with low or high cystatin C levels both associated with an increased risk of obesity. However, in girls, the risk of obesity showed a trend of initially increase and then decrease with increasing cystatin C levels. Prospective studies with larger sample sizes should be conducted to evaluate the value of cystatin C levels in predicting the progression of early obesity in adolescents of different sexes.

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 Dalian Medical University Ethics Committee. Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin. Written informed consent was obtained from the minor(s)' legal guardian/next of kin for the publication of any potentially identifiable images or data included in this article.

AUTHOR CONTRIBUTIONS

Y-XH conceived data, wrote the original draft and performed the primary analysis. YL and Y-NM carried out the field data collection. WW and J-MT extracted the data and helped with the analysis. N-NW and X-FL performed the formal analysis. XC provided design ideas, controlled the analytical methods, and edited the review & editing. All authors reviewed and approved the final manuscript. XC acts as guarantor. The corresponding author attests that all listed authors meet authorship criteria and that no others meeting the criteria have been omitted.

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REFERENCES

1. WHO. Obesity: Preventing and Managing the Global epidemic. Report of a WHO Consultation. *World Health Organ Tech Rep Ser* (2000) 894:i–xii, 1-253.

2. Singh AS, Mulder C, Twisk JW, van Mechelen W, Chinapaw MJ. Tracking of Childhood Overweight Into Adulthood: A Systematic Review of the Literature. *Obes Rev* (2008) 9(5):474–88. doi: 10.1111/j.1467-789X.2008.00475.x

3. Franks PW, Hanson RL, Knowler WC, Sievers ML, Bennett PH, Looker HC. Childhood Obesity, Other Cardiovascular Risk Factors, and Premature Death. *N Engl J Med* (2010) 362(6):485–93. doi: 10.1056/NEJMoa0904130
4. 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
5. Ogden CL, Carroll MD, Lawman HG, Fryar CD, Kruszon-Moran D, Kit BK, et al. Trends in Obesity Prevalence Among Children and Adolescents in the United States, 1988-1994 Through 2013-2014. *Jama* (2016) 315(21):2292–9. doi: 10.1001/jama.2016.6361
6. *The State Council Information Office of the People's Republic of China: Press Briefing for the Report on Chinese Residents' Chronic Diseases and Nutrition 2020. (in Chinese)* . Available at: http://www.gov.cn/xinwen/2020-12/24/content_5572983.htm.
7. Jia P, Luo M, Li Y, Zheng JS, Xiao Q, Luo J. Fast-Food Restaurant, Unhealthy Eating, and Childhood Obesity: A Systematic Review and Meta-Analysis. *Obes Rev* (2021) 22 Suppl 1(Suppl 1):e12944. doi: 10.1111/obr.12944
8. Kumar S, Kelly AS. Review of Childhood Obesity: From Epidemiology, Etiology, and Comorbidities to Clinical Assessment and Treatment. *Mayo Clin Proc* (2017) 92(2):251–65. doi: 10.1016/j.mayocp.2016.09.017
9. Barreto EF, Rule AD, Voils SA, Kane-Gill SL. Innovative Use of Novel Biomarkers to Improve the Safety of Renally Eliminated and Nephrotoxic Medications. *Pharmacotherapy* (2018) 38(8):794–803. doi: 10.1002/phar.2149
10. Knight EL, Verhave JC, Spiegelman D, Hillege HL, de Zeeuw D, Curhan GC, et al. Factors Influencing Serum Cystatin C Levels Other Than Renal Function and the Impact on Renal Function Measurement. *Kidney Int* (2004) 65 (4):1416–21. doi: 10.1111/j.1523-1755.2004.00517.x
11. Di Sessa A, Guarino S, Passaro AP, Liguori L, Umano GR, Cirillo G, et al. NAFLD and Renal Function in Children: Is There a Genetic Link? *Expert Rev Gastroenterol Hepatol* (2021) 15(9):975–84. doi: 10.1080/17474124.2021.1906649
12. Muntner P, Winston J, Uribarri J, Mann D, Fox CS. Overweight, Obesity, and Elevated Serum Cystatin C Levels in Adults in the United States. *Am J Med* (2008) 121(4):341–8. doi: 10.1016/j.amjmed.2008.01.003
13. Codoñer-Franch P, Ballester-Asensio E, Martínez-Pons L, Vallecillo-Hernández J, Navarro-Ruíz A, del Valle-Pérez R, et al. Cardiometabolic Risk, and Body Composition in Severely Obese Children. *Pediatr Nephrol* (2011) 26(2):301–7. doi: 10.1007/s00467-010-1679-7
14. Schwartz GJ, Brion LP, Spitzer A. The Use of Plasma Creatinine Concentration for Estimating Glomerular Filtration Rate in Infants, Children, and Adolescents. *Pediatr Clin North Am* (1987) 34(3):571–90. doi: 10.1016/s0031-3955(16)36251-4
15. Group of China Obesity Task Force. Body Mass Index Reference Norm for Screening Overweight and Obesity in Chinese Children and Adolescents. *Zhonghua Liu Xing Bing Xue Za Zhi* (2004) 25(2):97–102.
16. 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
17. Magalhães EI, Sant'Ana LF, Priore SE, Franceschini Sdo C. [Waist Circumference, Waist/Height Ratio, and Neck Circumference as Parameters of Central Obesity Assessment in Children]. *Rev Paul Pediatr* (2014) 32 (3):273–81. doi: 10.1590/0103-0582201432320
18. 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(2 Suppl 4th Report):555–76.
19. de Onis M, Onyango AW, Borghi E, Siyam A, Nishida C, Siekmann J. Development of a WHO Growth Reference for School-Aged Children and Adolescents. *Bull World Health Organ* (2007) 85(9):660–7. doi: 10.2471/blt.07.043497
20. Liu C, Wen J, Xiang J, Ouyang X, Yang Y, Lu W, et al. Age- and Sex-Specific Reference Intervals for the Serum Cystatin C/creatinine Ratio in Healthy Children (0-18 Years Old). *J Int Med Res* (2019) 47(7):3151–9. doi: 10.1177/0300060519855575
21. Groesbeck D, Köttgen A, Parekh R, Selvin E, Schwartz GJ, Coresh J, et al. Age, Gender, and Race Effects on Cystatin C Levels in US Adolescents. *Clin J Am Soc Nephrol* (2008) 3(6):1777–85. doi: 10.2215/cjn.00840208
22. Marmarinos A, Garoufi A, Panagoulia A, Dimou S, Drakatos A, Paraskakis I, et al. Cystatin-C Levels in Healthy Children and Adolescents: Influence of Age, Gender, Body Mass Index and Blood Pressure. *Clin Biochem* (2016) 49(1-2):150–3. doi: 10.1016/j.clinbiochem.2015.10.012
23. Simmonds M, Llewellyn A, Owen CG, Woolacott N. Predicting Adult Obesity From Childhood Obesity: A Systematic Review and Meta-Analysis. *Obes Rev* (2016) 17(2):95–107. doi: 10.1111/obr.12334
24. Perlenfein TJ, Murphy RM. Expression, Purification, and Characterization of Human Cystatin C Monomers and Oligomers. *Protein Expr Purif* (2016) 117:35–43. doi: 10.1016/j.pep.2015.09.023
25. Grubb A, Horio M, Hansson LO, Björk J, Nyman U, Flodin M, et al. Generation of a New Cystatin C-Based Estimating Equation for Glomerular Filtration Rate by Use of 7 Assays Standardized to the International Calibrator. *Clin Chem* (2014) 60(7):974–86. doi: 10.1373/clinchem.2013.220707
26. Marzuillo P, Grandone A, Di Sessa A, Guarino S, Diplomatico M, Umano GR, et al. Anthropometric and Biochemical Determinants of Estimated Glomerular Filtration Rate in a Large Cohort of Obese Children. *J Ren Nutr* (2018) 28(5):359–62. doi: 10.1053/j.jrn.2018.01.001
27. Dedual MA, Wueest S, Challa TD, Lucchini FC, Aeppli TRJ, Borsigova M, et al. Obesity-Induced Increase in Cystatin C Alleviates Tissue Inflammation. *Diabetes* (2020) 69(9):1927–35. doi: 10.2337/db19-1206
28. Klisic A, Kavaric N, Ninic A. Serum Cystatin C Levels Are Associated With Triglycerides/High-Density Lipoprotein Cholesterol Ratio in Adolescent Girls Ages Between 16-19 Years Old. *Eur Rev Med Pharmacol Sci* (2020) 24 (20):10680–6. doi: 10.26355/eurrev_202010_23426
29. Dubern B, Lafarge JC, Fellahi S, Naour N, Copin N, Bastard JP, et al. Relevance of Increased Serum Cystatin C to Vascular Alterations in Obese Children. *Pediatr Obes* (2012) 7(5):374–81. doi: 10.1111/j.2047-6310.2012.00053.x
30. Magnusson M, Hedblad B, Engström G, Persson M, Nilsson P, Melander O. High Levels of Cystatin C Predict the Metabolic Syndrome: The Prospective Malmö Diet and Cancer Study. *J Intern Med* (2013) 274(2):192–9. doi: 10.1111/joim.12051
31. Servais A, Giral P, Bernard M, Bruckert E, Deray G, Isnard Bagnis C. Is Serum Cystatin-C a Reliable Marker for Metabolic Syndrome? *Am J Med* (2008) 121 (5):426–32. doi: 10.1016/j.amjmed.2008.01.040
32. Young JA, Hwang SJ, Sarnak MJ, Hoffmann U, Massaro JM, Levy D, et al. Association of Visceral and Subcutaneous Adiposity With Kidney Function. *Clin J Am Soc Nephrol* (2008) 3(6):1786–91. doi: 10.2215/cjn.02490508
33. Wu J, Wu Q, Wu M, Mao W. Serum Cystatin C Predicts Mortality in HBV-Related Decompensated Cirrhosis. *BioMed Res Int* (2019) 2019:7272045. doi: 10.1155/2019/7272045

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ORIGINAL RESEARCH
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Nationwide Trends of Pediatric Obesity and BMI z-Score From 2017-2021 in China: Comparable Findings From Real-World Mobile- and Hospital-Based Data

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Introduction: Lifestyle changes including COVID-19 lockdown cause weight gain and may change obesity trends; however, timely changes are largely unknown and monitoring measures are usually lack. This first large-scale study aimed to analyze the real-world national trends of obesity prevalence of Chinese children in the past five years, and the impact of COVID-19 pandemic on pediatric obesity development through both mobile- and hospital-based data.

Methods: This study included children aged 3 to 19 years old all over China from January 2017 to April 2021. Hospital-measured and parent-reported cases from XIGAO database were analyzed. Body mass index (BMI) z-score calculation and obesity status evaluation were made according to Chinese standards. We evaluated obesity/overweight prevalence over the past five years and the changes of BMI z-score during COVID-19 lockdown.

Results: A total of 656396 children from 31 provinces were involved, including 447481 hospital-measured cases and 208915 parent-reported cases. The obesity and overweight prevalence were 8.05% (95%CI 7.76%–8.39%) and 10.06% (95%CI 10.79%–11.55%), comparable to those of China National Nutrition Surveys during 2015–2019. Northern China had the highest obesity prevalence. Parent-reported data had higher obesity/overweight prevalence than hospital-measured data (18.3% [95%CI 17.7%–18.9%] *vs.* 21.7% [95%CI 20.7%–23.0%]). The trend of obesity prevalence remained stable with slight decrease, but COVID-19 lockdown caused a significant increase of 1.86% in 2020. Both mobile- and hospital-based data showed weight gain in the first half of 2020. High BMI z-score increase were found among primary and junior middle school children, and children in northeast area during lockdown.

Conclusion: Weight gain during COVID-19 among Chinese children had regional differences and mainly affect primary and junior middle school children, thus warrants targeted interventions. The mobile growth assessment based on parent-reported data was a feasible, efficient and timely way for obesity monitoring among Chinese children, especially during epidemic.

Keywords: pediatric obesity, body mass index, children, adolescent, China

INTRODUCTION

Obesity in children and adolescents is a challenging public health issue, affecting more than 120 million children and adolescents aged 5–19 years in 2016 (1). In the past 30 years, the national obesity prevalence of children aged 6–17 in China increased from 1.8%–2.4% in the 1990s to 7.9%–12.7% in 2015–2019 (2). Excessive calorie intake and inactivity and/or sedentary behavior increase the body mass index (BMI) and obesity prevalence of children, which not only lead to the risk of cardiovascular and metabolic diseases, but also cause psychological and social complications. The shutdown caused by COVID-19 pandemic restricted children and adolescents' outdoor physical activities and changed their living habits including diet and sleep, resulting in weight gains (3). Both studies from China regional data (4) or other countries found significant weight gain during lockdown among school-aged children and adolescents (5).

Regular growth assessment is an important way to monitor weight gain of children. Monitoring data can not only help schools and governments to introduce corresponding countermeasures, but also remind parents in time to help their children develop a healthy lifestyle. With the popularization of mobile phones and the Internet, parents can use mobiles to upload data and assess the growth and development of their children anytime and anywhere. This contactless way has become a good method of monitoring during the epidemic without going to hospitals or schools.

Population-based study about weight changes of children across China before and after the COVID-19 pandemic was limited. This study aimed to evaluate the prevalence of obesity of children and adolescents of all age groups in various regions of China in the past five years; and the impact of the COVID-19 pandemic on obesity development among children of different regions and ages in the real-world. Furthermore, we aimed to evaluate the feasibility of mobile parental-evaluation in the growth and development monitoring of Chinese children.

METHODS

Study Population

In order to evaluate the changes of growth of Chinese children and adolescents continuously in the real world, this study analyzed data from January 2017 to April 2021 from the XIGAO database, which was developed to monitor the characteristics and trend of Chinese children's growth and development. XIGAO database was developed by Shijiazhuang Xigao Technology Co., Ltd, which consisted of two parts of data, part based on hospitals and the other based on mobiles. The hospital part of the database was built in 2016, and the data began to be collected from mobiles in 2017. Therefore, this study only included data after 2017. More than 1500 hospitals in 29 provinces, autonomous regions or municipalities were involved in the database. Data of outpatient who came to monitor the growth and development were automatically extracted and uploaded from the hospital information system to the database. Furthermore, parents from 31 provinces, autonomous regions or municipalities all over China could upload data to XIGAO database on their mobiles through the QR code displayed in websites, hospitals or during the health education of schools and kindergartens.

In this study, data including sex, date of birth, date of measurement, province, height and weight were collected and analyzed. Cases who were younger than 3 years old or older than 19 years old, lack important information such as sex, height, weight and age, whose height significantly deviated from the growth curve (height SDS without ± 2SD), or who had biologically implausible BMI (BMI z-score without ± 5SD) were removed (6).

This study was approved by the ethic committee of Shijiazhuang Kid Grow Science and Technology Co. Ltd (2021 [8]). The information related to patient identification in this study was hidden and was not handed over to the researchers.

Measurements and Obesity Definition

Height was accurate to 0.1 cm, and weight was accurate to 0.1 kg. BMI was calculated as weight (kg) divided by height squared (m^2). We calculated the z-score of BMI, height and weight according to Chinese data (7, 8).

For children ≥7 years old, the diagnosis of obesity or overweight was made according to the Chinese standard (9). For children between 3–5 years old, BMI z-score ≥2 was defined as overweight and BMI z-score ≥3 was defined as obesity; while for children between 5–7 years old, BMI z-score ≥1 was defined as overweight and BMI z-score ≥2 was defined as obesity. Underweight children and adolescents were assigned to the normal category when analyzing.

Statistical Analysis

Age-, sex- and region-standardized prevalence of obesity and overweight were calculated based on 2010 China census population data (10). The trends of prevalence of obesity and overweight and BMI z-score were also calculated separately by

sex, age (3–6 years old, 7–11 years old, 12–14 years old and 15–19 years old; children <7 years old were defined as preschool children, children ≥7 years old were defined as school-aged) and region (Central, East, North, Northeast, Northwest, South and Southwest). Since the COVID-19 epidemic in China was brought under control after April 2020 and schools in various regions gradually resumed classes after May 2020 (11); therefore, in order to understand the short-term impact of the epidemic on weight gain in children and adolescents, we compared the data in 2019, the first half of 2020, and the second half of 2020 to evaluate the impact of pandemic. Student's t test and χ^2 test were used to compare continuous and categorical variables. Bland-Altman analysis was used to evaluate the consistency of hospital- and mobile-based data. A two-sided p value < 0.05 was considered statistically significant. Statistical analyses were conducted using the R software program (version 4.3).

RESULTS

Basic Characteristics

During the study period, a total of 562840 cases from hospital and 325743 participants from mobiles were enrolled in this study. Finally, a total of 656396 cases were analyzed, including 447481 hospital-measured cases and 208915 parent-reported cases (**Figure 1**), with the mean age of 7.22 ± 3.18 years. **Table 1** showed the general characteristics of the study population.

The mean BMI z-score of the study population was 0.18 ± 1.31. Girls had higher BMI z-score than that of boys (0.20 ± 1.30 *vs.* 0.15 ± 1.33, $p < 0.0001$). However, the distribution of BMI z-score was different among different age groups between boys and girls (**Figure 2E**). In age group 3-5 years, boys displayed higher BMI z-score than girls ($p < 0.0001$), while BMI z-score of girls was higher than that of boys in age group 6–8 years, 9–11 years, and 12–14 years. Among adolescents aged 15–19, there was no significant statistical difference of BMI z-score between boys and girls.

Prevalence of Obesity and Overweight

According to Chinese standard, 50534 (7.7%) and 65750 (10.0%) children and adolescents in the study population were obese and overweight, respectively. After adjusting age, sex and region, the standardized prevalence of obesity and obesity/overweight in the study population was 8.05% (95%CI 7.76%–8.39%) and 19.19% (95%CI 18.73%–19.70%).

Boys had higher standardized prevalence of obesity than girls (9.03% [95%CI 8.65%–9.46%] *vs.* 6.91% [95%CI 6.46%–7.47%]). Children in the north area of China presented the highest standardized prevalence of obesity (10.85% [95%CI 9.37%–12.73%]), and followed by the northeast area of China (9.48% [95%CI 8.98%–10.04%]); while the south (6.16% [95% 5.75%–6.61%]) and southwest (6.68% [95%CI 6.41%–7.31%]) region had the lowest obesity prevalence (**Supplementary Table S1** and **Figures 2A, B**).

The obesity prevalence changed with ages. It first increased significantly after the age of four, reached the peak at the age of eight, then slowly declined between 9–14 years old, and finally raised again at the age of 15 (**Figure 2C**). The overall change trend of the prevalence of obesity/overweight was similar to the prevalence of obesity, but it always showed a high level after the age of eight.

Trends of Obesity Prevalence and BMI z-Score

Despite the significant increase in 2020 due to COVID-19 lockdown, the prevalence of obesity and obesity/overweight among Chinese children and adolescents showed a steadily decrease trend (**Figure 2D**). Compared with 2017–2018, the prevalence of obesity and obesity/overweight in 2019 was decreased by 0.26% and 0.87% respectively. Although increased to 9.53% in 2020, with the increase of 1.86%, the prevalence of obesity returned to 7.76% in the first four months of 2021, which was close to that in 2019.

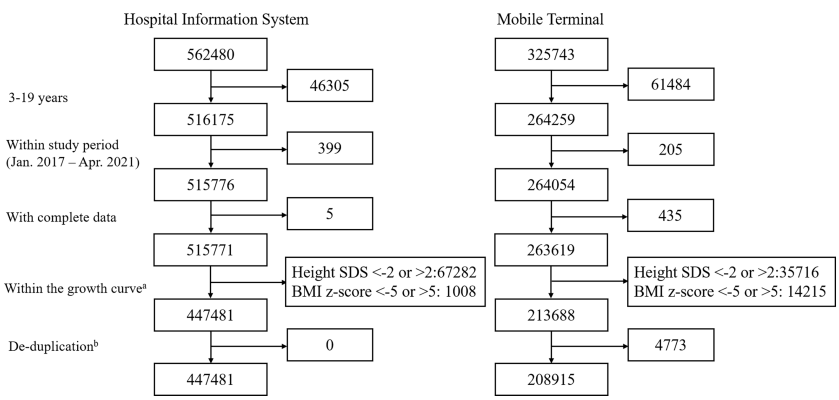

FIGURE 1 | Flow chart of sampling procedure. [a]To remove the cases that might had growth disorder, we only included the cases whose height age- and sex-specific SDS was between ±2. Then cases with BMI z-score higher than 5 or lower than -5 were excluded in order to excluded incorrect entries and weight abnormalities that might be secondary to other diseases. [b]Each child had a unique ID. Cases that repeatedly registered data in both the hospital database and the mobile database were regarded as repeated cases, and data from mobiles were excluded.

TABLE 1 | General characteristics of the study population.

	Total (n = 656396)	Female (n = 321973)	Male (n = 334423)	$P^{\#}$
Age (years)	7.22 (3.18)	7.30 (2.96)	7.24 (3.39)	< 0.0001
Age groups (years)				< 0.0001
3–6	363466 (55.4%)	167364 (52.0%)	196102 (58.6%)	
7–11	230412 (35.1%)	133112 (41.3%)	97300 (29.1%)	
12–14	53328 (8.1%)	18353 (5.7%)	34975 (10.5%)	
15–19	9190 (1.4%)	3144 (1.0%)	6046 (1.8%)	
Region*				< 0.0001
Central	61046 (9.3%)	29099 (9.0%)	31947 (9.6%)	
East	170941 (26.0%)	83280 (25.9%)	87661 (26.2%)	
North	161040 (24.5%)	77798 (24.2%)	83242 (24.9%)	
Northeast	14202 (2.2%)	6883 (2.1%)	7319 (2.2%)	
Northwest	6809 (1.0%)	3368 (1.0%)	3441 (1.0%)	
South	140042 (21.6%)	73276 (22.8%)	66766 (20.0%)	
Southwest	65651 (10.0%)	32434 (10.1%)	33217 (9.9%)	
BMI z-score	0.18 (1.31)	0.20 (1.30)	0.15 (1.33)	< 0.0001
Obesity status				< 0.0001
Obesity	50534 (7.7%)	22467 (7.0%)	28067 (8.4%)	
Overweight	65750 (10.0%)	30278 (9.4%)	35472 (10.6%)	
Obesity/Overweight	116284 (17.7%)	52745 (16.4%)	53539 (19.0%)	

Data were n (%) or mean (SD). BMI, body-mass index.
**36665 cases from mobiles didn't have exact province. #p value for difference in different sexes.*

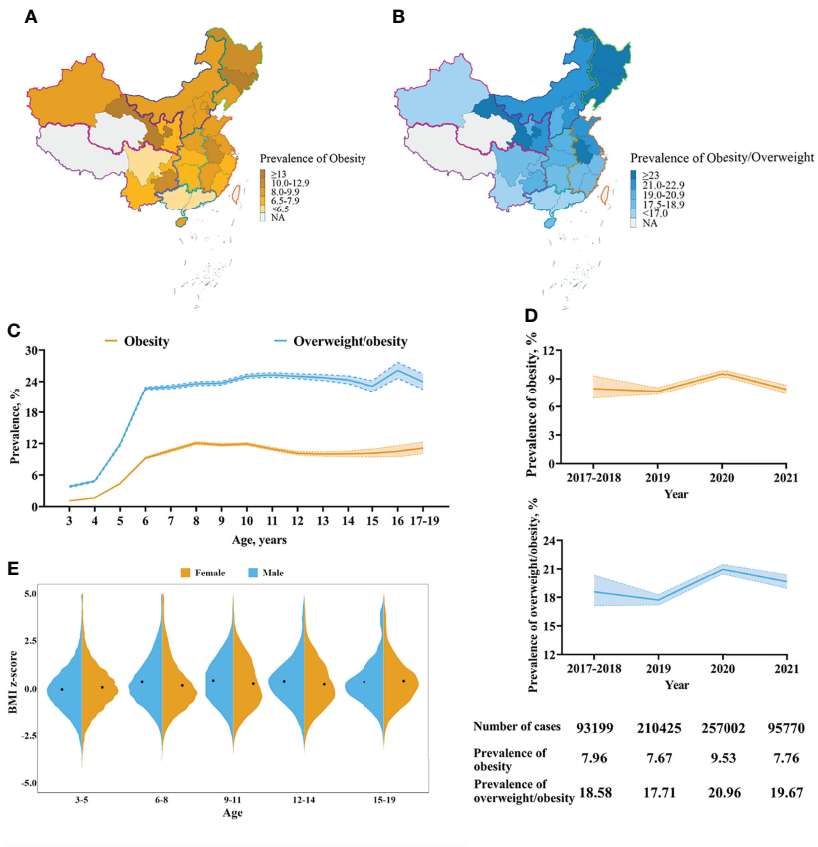

FIGURE 2 | Characteristics of the standardized prevalence of obesity and overweight and BMI z-score. **(A)** The standardized prevalence of obesity by province. **(B)** The standardized prevalence of obesity and overweight by province. The prevalence was standardized by sex and age in different provinces. This study didn't included children in Taiwan, Hong Kong and Macao. Cases in Tibet Autonomous Region and Qinghai province were limited thus were not included in the map. **(C)** Trajectories of the prevalence of obesity and overweight by age. **(D)** Trends of the prevalence of obesity and overweight. The prevalence was standardized by sex and age. The band indicated 95% CI. **(E)** BMI z-score of different sexes and ages. Black points indicated the mean of BMI z-score of different groups.

However, BMI z-score of children and adolescents increased in the past five years (**Supplementary Figure 1**). Compared with the lowest level in 2019, with 0.09 ± 1.35, the mean BMI z-score increased to 0.23 ± 1.34 in 2020 and continued to reach 0.27 ± 1.23 in the first four months of 2021. But the changes of BMI z-score had sex, age and region characteristics, and were related with COVID-19 pandemic shutdown.

For preschool children, a significant increase of the mean of BMI z-score was found in the first four months of 2021; while BMI z-score increased significantly among school-aged children and adolescents in the year of 2020, and a slight decrease was found in 2021 (**Supplementary Table S2**).

Further analyzing the influence of COVID-19 lockdown on the changes of BMI z-score, we found that children aged 7–11 years old were most effected by the lockdown with the increase of 0.26 in the mean of BMI z-score in the first half of 2020 compared with 2019, followed by adolescent aged 12–14 years, with the increase of 0.20 (**Table 2**). Preschool children only had relatively slight increase in BMI z-score in the first half of 2020 and returned to the previous level in the second half of 2020. Furthermore, children in the northeast gained the most weight in the first half of 2020, and children in the central, north and southwest regions also experienced an increase of BMI z-score more than 0.20 (**Figure 3**). Notably, the BMI z-score of both preschool and school-aged children in the north area of China increased constantly, even after the year of 2020 (**Supplementary Table S3**).

The Accuracy of Parent-Reported Data

To analyze the accuracy of parent-reported data, we compared the duplicate cases between hospitals and mobiles within 30 days. Finally, 4309 cases uploaded data both from hospitals and mobiles within 30 days. When reviewing these data, 3980 (92.3%) cases reported the same BMI through hospitals and mobile terminals, and the differences of BMI of 4096 cases were within 0.20 (95.1%). Furthermore, we found 27 cases (0.6%) whose parent-reported weight value was nearly two times higher than hospital-measured weight value, indicating parents might mistake the unit when uploading the data. There was no significant difference between the BMI z-score of hospital- and mobile-based data according to Bland-Altman analysis (p = 0.62) (**Supplementary Figure 2**). Since the proportion of possible errors and bias was low, the parent-reported data from the mobiles was reliable.

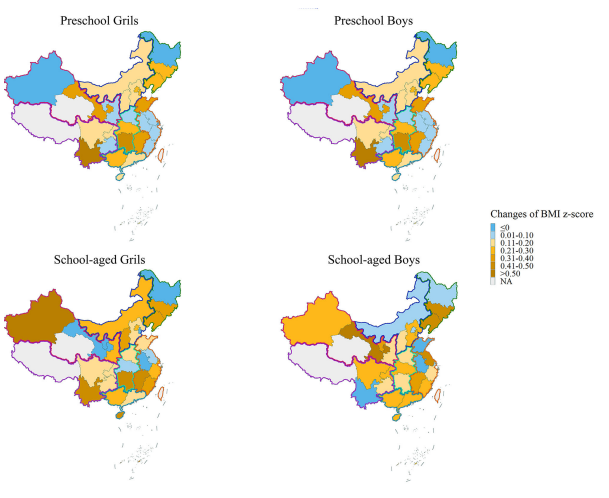

FIGURE 3 | BMI z-score changes during COVID-19 lockdown by province. The changes of the mean BMI z-score in the first half of 2020 and that in 2019.

The Characteristics of Parent-reported Measurements in BMI Monitoring

31.8% cases of this study were parent-reported data obtained from the mobiles; therefore, we could evaluate the characteristics of parent-reported data (**Supplementary Table S4**). Parents of preschool children were more inclined to participate in the BMI monitoring study, accounting for 73.9% of the total mobile-based data. Compared with the hospital measurement data, parent-reported data had higher standardized obesity and obesity/overweight prevalence (6.9% [95% CI 6.7%–7.3%] *vs.* 11.8% [95% CI 10.9%–13.0%]; 18.3% [95% CI 17.7%–18.9%] *vs.* 21.7% [95% CI 20.7%–23.0%]). Furthermore, parent-reported data had higher BMI z-score among both preschool and school-aged children (0.07 ± 1.42 *vs.* 0.05 ± 1.21, p = 0.0020; 0.43 ± 1.57 *vs.* 0.30 ± 1.25, p < 0.0001). However, both mobile-based data and hospital-based data could timely reflect the changes of BMI during pandemic (**Supplementary Table S5**).

DISCUSSION

This first large-scale study evaluated the changing trend of obesity prevalence and BMI of children and adolescents in different regions of China in the past five years using mobile- and hospital-based data. From 2017 to 2021, the obesity and overweight prevalence of Chinese children and adolescents remained at a stable level and had a slight downward trend; though the lockdown in 2020 led to a significant increase. The COVID-19 epidemic in 2020 had an impact on obesity and weight gain, but there were regional and age differences.

In the past decade, the prevalence of obesity and overweight in children and adolescents in China increased from 15.4% in 2010, to 20.4% in 2014 and 24.7%–25.8% in 2017–2019 (12–14). In our study, the prevalence of obesity and overweight was 19.19%, which was slightly lower compared with that Zhang et al. (12) and 2014 Chinese National Survey on Students' Constitution and Health (15) found. Since our study included more provinces in the south and southwest area of China and preschool children aged 3–6 years old, their relatively low prevalence of obesity and overweight resulted in a lower overall prevalence than previous studies.

The prevalence of obesity had region-, age- and sex-specific characteristics. Consist with previous studies, Chinese boys and children in northern region of China had higher prevalence of obesity, while children in south and southwest regions of China had lower prevalence (12, 16, 17). Furthermore, even in the same province, there are differences in obesity in different cities, calling

TABLE 2 | BMI z-score changes during COVID-19 shutdown by different ages and regions.

	2019	Jan.-Jun. 2020	Jul. – Dec. 2020	Jan.-Apr. 2021
Age groups (years)				
3–6	0.03 (1.36)	0.12 (1.33)	0.01 (1.29)	0.15 (1.24)
7–11	0.20 (1.34)	0.46 (1.32)	0.39 (1.36)	0.38 (1.23)
12–14	0.19 (1.26)	0.39 (1.35)	0.32 (1.35)	0.32 (1.18)
15–19	0.34 (1.28)	0.45 (1.35)	0.38 (1.27)	0.41 (1.09)
Region				
Central	0.13 (1.31)	0.35 (1.32)	0.24 (1.30)	0.29 (1.16)
East	0.17 (1.30)	0.35 (1.33)	0.31 (1.34)	0.34 (1.19)
North	0.17 (1.34)	0.38 (1.31)	0.21 (1.30)	0.50 (1.22)
Northeast	0.13 (1.40)	0.50 (1.40)	0.30 (1.34)	0.33 (1.29)
Northwest	0.11 (1.23)	0.24 (1.36)	0.33 (1.28)	0.38 (1.26)
South	-0.09 (1.37)	0.08 (1.30)	0.02 (1.33)	0.07 (1.25)
Southwest	-0.02 (1.28)	0.22 (1.29)	0.12 (1.25)	0.14 (1.15)

Data were mean (SD).

for more targeted policies for the controlling of pediatric obesity in China (18). As for the difference between different ages, we found that the obesity prevalence increased significantly after the age of four, while it was relatively low between the ages of 12–15, but then increased again. The increase of obesity prevalence in adolescents older than 16 years old was consistent with the findings among Chinese adults that young adult men had relatively high obesity prevalence (19).

Remarkably, our study found that the prevalence of obesity among Chinese children had entered a stage of stable or even with a slight decline, comparing to the annual growth rate of 0.58% and 0.63% in obesity and overweight respectively in 2010–2014 (17). A decrease of 0.26% in obesity prevalence was found in 2019 compared with 2017–2018. Although experienced a significant increase in 2020, the prevalence of obesity in the first four months of 2021 was 7.79%, which was decreased significantly comparing with the prevalence in 2020, with a decrease of 1.77%. Due to the limited research period and the special impact of the COVID-19 epidemic, we cannot conclude that the obesity problem of Chinese children has been controlled. The study of Yuan et al. (20) among Chinese children and the study of adult population in China both found a slowdown in the rise of obesity prevalence (19), suggesting an optimistic trend of obesity problem among all age groups in China. However, the overall prevalence is still at high point and long-term trend needs to be observed. Furthermore, the huge population of obese and overweight children and adolescents still needs more attention.

Although some studies have focused on the weight gain of children in China during the COVID-19 epidemic (4, 21, 22), there is still a lack of comprehensive national analysis of multiple age groups. To reduce the impact of going out restrictions on the assessment of children's growth and development, we innovatively use mobiles as a data-source for this nationwide survey. Consist with previous regional studies (4, 5), COVID-19 lockdown led to weight gains among Chinese children, but also had age and region differences.

The age difference in weight gain might be related to the growth characteristics and lifestyles of children of different ages. We found that children and adolescents aged 7–14 years old, mainly children in primary schools and junior middle schools, had the most BMI z-score gain. For school-aged children, the lower the grade, the more time spent in school activities, including physical education classes and extracurricular activities; therefore, the exercise time of primary and junior middle school students was greatly affected by the lockdown. High school students had less activity time at school than younger children in China, so they might be less affected; while preschool children had a lot of non-school activity time, so they were almost unaffected. Findings of other regional studies in China and the United States were consistent with our results (4, 22, 23).

Regional differences in weight gain during COVID-19 lockdown might be affected by various factors such as the severity of the epidemic and differences in local lifestyles. Children in northeast, central, north and southwest area showed a significant increase in BMI z-score in the first half of 2020 and a decline in the second half of the year, which was consistent with the development trend of the epidemic in China. However, the weight of children in the northwest and north increased continuously even after the epidemic, and the mean BMI z-score reached 0.5 in northern China. This suggested that the north and northwest regions are the focus of attention of obesity monitoring in the future.

Conducting family self-assessment was a good way for monitoring weight changes and providing timely interventions based on changes, especially during the epidemic, when the assessment carried out in schools and clinics was restricted. Our study has confirmed the feasibility of using mobiles and the Internet to conduct growth and development assessments for children and adolescents across China. By entering the basic information and anthropometry data, parents can obtain the results of their children's developmental assessment within a few seconds, avoiding complicated calculations and table lookups that need to be completed by pediatricians. In the future, the online assessment method can be widely promoted as an effective assessment method for children's growth and development, especially during epidemics and in remote areas. The main concern about the online assessment was the inaccuracy of parent- and self-reported data, which might have bias due to reporting behaviors and systematic measurement differences. Zhou et al. (24) raised concerns about the accuracy of self-reported weight and height of Chinese adolescents, and He et al. (25) found that parent-reported BMI tended to overestimate the

prevalence of obesity in children and adolescents. A study of the accuracy of parental measurements of height and weight during COVID-19 found lower BMI at home than in clinics (26). Although the accuracy analysis of this study suggested that the parent-reported data was reliable, we found that the BMI z-score of mobile-based data was higher than those of hospital-based data in this study, especially among school-aged children. Although there was statistical difference among preschool children, the difference was only 0.02. In addition to the possible errors caused by clothing and food intake during the home measurement process, parents of obese children were more inclined to participate in related assessments might be the reason for this difference. Even with bias, a slight overestimation of BMI in obesity assessment helped parents of children at risk of overweight to pay more attention on their diets and lifestyles, and seek help from pediatricians in time. Therefore, parent-reported growth assessment through mobile was a feasible, efficient and timely way with synchronous response, thus it is worthy of being widely promoted. In the future, better health education and measurement teaching will help parents measure their children's height and weight more accurately.

One of the limitations of this study was that all cases from mobiles were voluntarily participated. Therefore, younger children accounted for a large proportion of the study population. However, when calculating the trend of BMI, children were divided into different age groups, and when calculating the obesity prevalence, we standardized the prevalence according to age, sex and region, so it would not affect the results. In addition, we found that the obesity prevalence of mobile-based data was higher than that of the hospital-based data and other researches (2, 12, 13). The reason for this phenomenon might be that parents of overweight children paid more attention to the assessment of their children's growth and development and were more inclined to participate in such surveys. Furthermore, this large-scale epidemiological survey didn't collect information about lifestyles, such as dietary or physical activities, which were important factors affecting BMI. In the future, we will expand the information collected in the database to better investigate possible factors affecting BMI changes.

CONCLUSIONS

In conclusion, the prevalence of obesity and overweight of Chinese children and adolescents showed a slight downward trend though increased in 2020 due to the COVID-19 lockdown. The high obesity prevalence in northern China and the continuous increase of BMI z-score in north and northwest regions called for more region-specific ways for pediatric obesity management. The assessment through mobiles based on parent-reported anthropometry data was a good way for obesity monitoring, especially during epidemics. High BMI z-score increase among primary and junior middle school children suggested more indoor sports education for these children especially during the pandemic.

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 ethic committee of Shijiazhuang Kid Grow Science and Technology Co. Ltd. Written informed consent from the participants' legal guardian/next of kin was not required to participate in this study in accordance with the national legislation and the institutional requirements.

AUTHOR CONTRIBUTIONS

YY and ZX conceptualized and designed the study, completed the statistical analyses, drafted the initial manuscript, and reviewed and revised the manuscript. MZ and SZ developed the database and supervised the data collection. FL supervised the data collection, contributed to the design of the study, and reviewed and revised the manuscript. ZP and CS contributed to interpretation of the data and extensive revision of the manuscript. JY, JH and TQ assisted with data interpretation and reviewed and revised the manuscript. All authors approved the final manuscript as submitted and agreed to be accountable for all aspects of the work.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2022.859245/full#supplementary-material

REFERENCES

1. Bentham J, Di Cesare M, Bilano V, Bixby H, Zhou B, Stevens GA, et al. 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) 390:2627–42. doi: 10.1016/S0140-6736(17)32129-3
2. Pan XF, Wang L, Pan A. Epidemiology and Determinants of Obesity in China. *Lancet Diabetes Endocrinol* (2021) 9:373–92. doi: 10.1016/S2213-8587(21)00045-0
3. Pietrobelli A, Pecoraro L, Ferruzzi A, Heo M, Zoller T, Antoniazzi F, et al. Effects of COVID-19 Lockdown on Lifestyle Behaviors in Children With Obesity Living in Verona, Italy: A Longitudinal Study. *Obes (Silver Spring)* (2020) 28:1382–5. doi: 10.1002/oby.22861.Effects
4. Hu J, Liu J, Wang J, Shen M, Ge W, Shen H, et al. Unfavorable Progression of Obesity in Children and Adolescents Due to COVID-19 Pandemic: A School-Based Survey in China. *Obesity* (2021)29:1907–15. doi: 10.1002/oby.23276
5. Chang T, Chen Y, Chen W, Chen C, Hsu W, Chou Y. Weight Gain Associated With COVID-19 Lockdown in Children and Adolescents : A Systematic Review and Meta-Analysis. *J Nutrients* (2021) 13:3668. doi: 10.3390/nu13103668
6. Spinelli A, Buoncristiano M, Kovacs VA, Yngve A, Spiroski I, Obreja G, et al. Prevalence of Severe Obesity Among Primary School Children in 21 European Countries. *Obes Facts* (2019) 12:244–58. doi: 10.1159/000500436
7. Hui L, Cheng-ye J, Xin-nan Z, Ya-qin Z. Body Mass Index Growth Curves for Chinese Children and Adolescents Aged 0 to 18 Years. *Chin J Pediatr* (2009) 47:493–8. doi: 10.3760/cma.j.issn.0578-1310.2009.07.004
8. Hui L, Cheng-ye J, Xin-nan Z, Ya-qin Z. [Height and Weight Standardized Growth Charts for Chinese Children and Adolescents Aged 0-18 Years]. *Chin J Peidatr* (2009) 47:487–92. doi: 10.3760/cma.j.issn.0578-1310.2009.07.003
9. Group of China Obesity Task Force. [Body Mass Index Reference Norm for Screening Overweight and Obesity in Chinese Children and Adolescents]. *Chin J Epideminol* (2004) 25:97–102. doi: 10.3760/j.issn:0254-6450.2004.02.003
10. National Bureau of Statisitcs. *Tabulation on the 2010 Population Census of the People's Republic of China*. Available at: http://www.stats.gov.cn/tjsj/pcsj/rkpc/6rp/indexch.htm.
11. WHO Coronavirus (COVID-19). Available at: https://covid19.who.int/region/wpro/country/cn.
12. Zhang L, Chen J, Zhang J, Wu W, Huang K, Chen R, et al. Regional Disparities in Obesity Among a Heterogeneous Population of Chinese Children and Adolescents. *JAMA Netw Open* (2021) 4:1–9. doi: 10.1001/jamanetworkopen.2021.31040
13. Dong Y, Lau PWC, Dong B, Zou Z, Yang Y, Wen B, et al. Trends in Physical Fitness, Growth, and Nutritional Status of Chinese Children and Adolescents: A Retrospective Analysis of 1·5 Million Students From Six Successive National Surveys Between 1985 and 2014. *Lancet Child Adolesc Heal* (2019) 3:871–80. doi: 10.1016/S2352-4642(19)30302-5
14. Zhu Z, Tang Y, Zhuang J, Liu Y, Wu X, Cai Y, et al. Physical Activity, Screen Viewing Time, and Overweight/Obesity Among Chinese Children and Adolescents: An Update From the 2017 Physical Activity and Fitness in China - The Youth Study. *BMC Public Health* (2019) 19:1–8. doi: 10.1186/s12889-019-6515-9
15. Dong Y, Jan C, Ma Y, Dong B, Zou Z, Yang Y, et al. Economic Development and the Nutritional Status of Chinese School-Aged Children and Adolescents From 1995 to 2014: An Analysis of Five Successive National Surveys. *Lancet Diabetes Endocrinol* (2019) 7:288–99. doi: 10.1016/S2213-8587(19)30075-0
16. Dong Y, Ma Y, Dong B, Zou Z, Hu P, Wang Z, et al. Geographical Variation and Urban-Rural Disparity of Overweight and Obesity in Chinese School-Aged Children Between 2010 and 2014: Two Successive National Cross-Sectional Surveys. *BMJ Open* (2019) 9:1–9. doi: 10.1136/bmjopen-2018-025559
17. Shuo W, Yanhui D, Zhenghe W, Zhiyong Z, Health A. Trends in Overweight and Obesity Among Chinese Children of 7- 18 Years Old During 1985- 2014. *Chin J Prev Med* (2017) 51:300–5. doi: 10.3760/cma.j.issn.0253-9624.2017.04.005
18. Qin W, Wang L, Xu L, Sun L, Li J, Zhang J, et al. An Exploratory Spatial Analysis of Overweight and Obesity Among Children and Adolescents in Shandong, China. *BMJ Open* (2019) 9:1–8. doi: 10.1136/bmjopen-2018-028152
19. Wang L, Zhou B, Zhao Z, Yang L, Zhang M, Jiang Y, et al. Body-Mass Index and Obesity in Urban and Rural China: Findings From Consecutive Nationally Representative Surveys During 2004–18. *Lancet* (2021) 398:53–63. doi: 10.1016/S0140-6736(21)00798-4
20. Yuan JN, Jin BH, Si ST, Yu YX, Liang L, Wang CL, et al. [Changing Prevalence of Overweight and Obesity Among Chinese Children Aged 6-15 From 2009-2019]. *Zhonghua Er Ke Za Zhi = Chin J Pediatr* (2021) 59:935–41. doi: 10.3760/cma.j.cn112140-20210523-00441
21. Qiu N, He H, Qiao L, Ding Y, Ji S, Guo X, et al. Sex Differences in Changes in BMI and Blood Pressure in Chinese School-Aged Children During the COVID-19 Quarantine. *Int J Obes* (2021) 45:2132–6. doi: 10.1038/s41366-021-00871-w
22. Wen J, Zhu L, Ji C. Changes in Weight and Height Among Chinese Preschool Children During COVID-19 School Closures. *Int J Obes* (2021) 45:2269–73. doi: 10.1038/s41366-021-00912-4
23. Lange SJ, Kompaniyets L, Freedman DS, Kraus EM, Porter R, Blanck HM, et al. Longitudinal Trends in Body Mass Index Before and During the COVID-19 Pandemic Among Persons Aged 2–19 Years — United States, 2018–2020. *MMWR Morb Mortal Wkly Rep* (2021) 70:1278–83. doi: 10.15585/mmwr.mm7037a3
24. Zhou X, Dibley MJ, Cheng Y, Ouyang X, Yan H. Validity of Self-Reported Weight, Height and Resultant Body Mass Index in Chinese Adolescents and Factors Associated With Errors in Self-Reports. *BMC Public Health* (2010) 10. doi: 10.1186/1471-2458-10-190
25. He J, Fan X. How Accurate is Using Parent-Reported Height and Weight for Screening Children and Adolescents for Overweight and Obesity? Meta-Analyses at Both Population and Individual Levels. *Child Obes* (2018) 14:302–15. doi: 10.1089/chi.2018.0062
26. Tenenbaum A, Shefer-Averbuch N, Lazar L, Yakobovitch-Gavan M, Phillip M, Oron T. Growth Assessment of Children During the COVID-19 Pandemic —Can We Rely on Parental Measurements? *Acta Paediatr Int J Paediatr* (2021) 110:3040–5. doi: 10.1111/apa.16034

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Trends in Abdominal Obesity and Central Adiposity Measures by Dual-Energy X-Ray Absorptiometry Among US Children: 2011–2018

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Objective: Previous studies that have reported trends on abdominal obesity among US children were usually based on anthropometric assessments. However, little is known about the recent trends in central adiposity measures by DXA and abdominal obesity since 2011–2012.

Study Design: A serial cross-sectional analysis of US population-weighted data among children from NHANES 2011 to 2018 was conducted.

Results: Between 2011–2012 and 2017–2018, there was a relatively stable trend among children aged 8–19 years in trunk fat and trunk fat percentage. During the same time periods, there were no significant changes in prevalence of abdominal obesity by waist circumference (18.6 vs. 21.1%) among those aged 2–19 years, and abdominal obesity by WHtR (34.1 vs. 36.2%) among those aged 6–19 years. However, a significant increase trend among boys aged 2–19 years was found in prevalence of abdominal obesity by waist circumference (16.1–22.7%; $P = 0.004$). For Mexican American youth and non-Hispanic Asian boys, there is a significant increase in mean trunk fat percentage and waist circumference.

Conclusion: Between 2011–2012 and 2017–2018, there have been no significant changes in central adiposity measured by DXA and prevalence of abdominal obesity among US children. Our study further supports that there is an urgent need to improve their lifestyle to reduce abdominal obesity for US children, especially for Mexican American youth and non-Hispanic Asian boys.

Keywords: trunk fat, trunk fat percentage, central adiposity measures, dual-energy x-ray absorptiometry, abdominal obesity, children

INTRODUCTION

Abdominal obesity is strongly associated with several risk factors for metabolic syndrome, diabetes, and cardiovascular disease than overall obesity (1). Among children and adolescents, many analyses about abdominal obesity based on waist circumference or waist-to-height ratio (WHtR) have been published (2). Dual-energy X-ray absorptiometry (DXA) is the most widely accepted method of

objectively measuring body composition and suitable for assessing body composition in children and adolescents with good feasibility and reasonable accuracy. However, little is known about the recent trends in central adiposity measures by DXA and abdominal obesity among children and adolescents since 2011–2012. From 2011–12 to 2017–18, the NHANES DXA examination provides nationally representative data on body composition measured using whole body DXA scans (3). Moreover, some studies showed that Asian individuals were shown to have a different body fat percentage and fat-free mass than other racial or ethnic groups (4, 5). Since the 2011–2012 NHANES cycle, non-Hispanic Asian individuals were differentiated in the released data and can be analyzed as a separate racial or ethnic category in the present study.

In addition, the intra-abdominal adipose tissue depot is considered mainly responsible for the adverse metabolic effects of a central fat pattern (6). Although DXA cannot discriminate between intra-abdominal and subcutaneous fat, studies in children (7) indicated strong associations between trunk fat mass measured by DXA and intra-abdominal fat measured by computed tomography or magnetic resonance imaging.

Therefore, this study aimed to examine the most recent national estimates and trends in measures of central body fat by DXA and abdominal obesity using data from NHANES 2011–2018, including waist circumference, WHtR, trunk fat, and trunk fat percentage by race or ethnicity among US children and adolescents.

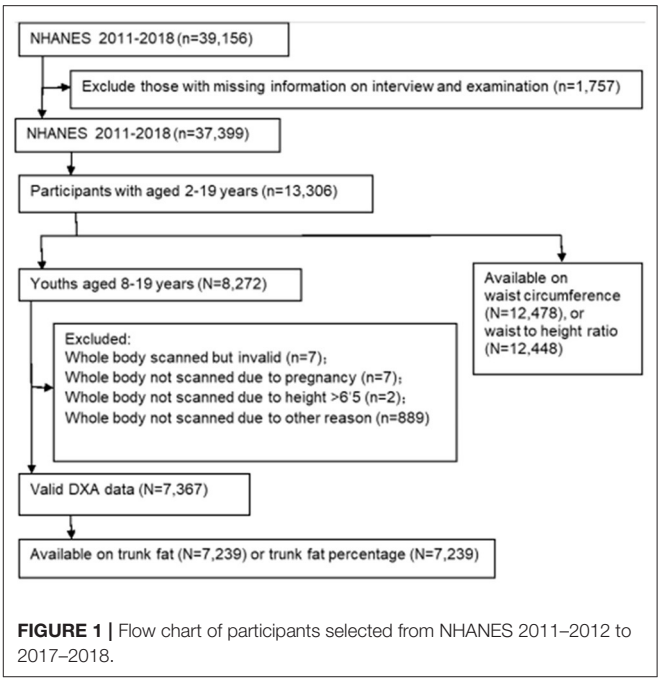

FIGURE 1 | Flow chart of participants selected from NHANES 2011–2012 to 2017–2018.

METHODS

Study Design

NHANES uses a stratified multistage probability sampling design to obtain a representative sample of the US non-institutionalized civilian population and is a cross-sectional survey designed to monitor the health and nutritional status (8). Participants were interviewed at home to collect sociodemographic, family, and medical information. They also received a detailed medical examination in the mobile examination center. From 2011, the NHANES restarted to provide nationally representative data on body composition by performing whole body DXA scans. NHANES procedures were approved by a human subject review board, and written informed consent, assent, or both was obtained from all participants. The overall examination response rate has gradually decreased from 69.5% in 2011–12 to 48.8% in 2017–2018. We included children and adolescents aged 2–19 years who attended an examination during any NHANES cycle from 2011–2012 to 2017–2018. All available data were analyzed in all available NHANES cycles from 2011 to 2018. If relevant variables were missing, individuals from particular analyses were excluded. We included all available NHANES cycles for waist circumference, WHtR, trunk fat, and trunk fat percentage from 2011–2012 to 2017–2018. **Figure 1** indicates a flowchart of participants.

Anthropometric Measurements

To ensure comparability of anthropometric measures of survey participants, body measurements were obtained using the standardized methods and equipment of the Centers for Disease Control and Prevention throughout the NHANES surveys. Waist circumference was measured above the iliac crest using a steel tape. WHtR was defined as the ratio of waist (cm) and height (cm).

DXA Measurements

Trunk fat and trunk fat percentage were determined for participants aged 8–59 years by whole body DXA. Whole body scans were performed on the Hologic Discovery model A densitometers with low radiation exposure (<20 μSv). All scan data were analyzed using Hologic APEX version 4.0 software with the NHANES BCA option.

Other Variables

Information about age, gender, and race/ethnicity was obtained using standardized questionnaires during in-person interviews. Participants self- or parent-reported their race and ethnicity from provided categories and coded as Mexican, other Hispanic, non-Hispanic white, non-Hispanic black, non-Hispanic Asian, or other races since 2011. In this study, other Hispanic and other races were combined to create other racial/ethnic groups. Race or ethnicity was categorized as Mexican American, non-Hispanic white, non-Hispanic black, non-Hispanic Asian, and other race/ethnic groups.

Statistical Analysis

According to the NHANES analytic guidelines, all analyses used the NHANES examination sample weights to account for differential probabilities of oversampling, non-response, and non-coverage (9). This study estimated mean trunk fat and trunk fat percentage among children and adolescents aged 8–19 years. Also, we evaluated age-adjusted mean waist circumference and

TABLE 1 | Trends in mean trunk fat (95% CI) by race or ethnicity among US children aged 8–19 y, NHANES 2011–2012 to 2017–2018.

Variable	Mean trunk fat (kg)				Absolute Increase[a]	*P* for trend[b]
	2011–12 (*n* = 1,828)	2013–14 (*n* = 1,996)	2015–16 (*n* = 1,907)	2017–18 (*n* = 1,508)		
Overall	7.05 (6.65–7.44)	7.28 (6.75–7.81)	7.07 (6.58–7.56)	7.14 (6.87–7.41)	0.09	0.50
Age, y						
8–11	4.71 (4.30–5.13)	4.79 (4.32–5.25)	4.53 (4.18–4.88)	4.85 (4.41–5.30)	0.14	0.78
12–19	8.17 (7.62–8.73)	8.51 (7.77–9.24)	8.35 (7.79–8.92)	8.39 (8.10–8.68)	0.22	0.53
Gender						
Boys	6.34 (5.85–6.83)	6.59 (6.14–7.05)	6.42 (5.80–7.05)	6.54 (6.24–6.84)	0.20	0.36
Girls	7.78 (7.24–8.32)	8.04 (7.28–8.79)	7.79 (7.15–8.43)	7.81 (7.40–8.22)	0.03	0.86
RACE/ETHNICITY						
Mexican American						
All	7.53 (7.02–8.03)	8.01 (7.40–8.63)	9.00 (8.43–9.58)	8.17 (7.29–9.05)	0.64	**0.03**
Boys	7.02 (6.42–7.63)	7.15 (6.54–7.75)	8.89 (7.74–10.05)	7.53 (6.46–8.61)	0.51	**0.04**
Girls	8.08 (7.33–8.83)	8.95 (8.10–9.79)	9.13 (8.18–10.08)	8.84 (7.58–10.11)	0.76	0.22
Non-hispanic white						
All	6.95 (6.31–7.59)	7.19 (6.41–7.96)	6.66 (6.07–7.26)	6.87 (6.38–7.37)	−0.08	0.71
Boys	6.09 (5.35–6.82)	6.69 (6.02–7.36)	5.94 (5.23–6.66)	6.42 (5.89–6.95)	0.33	0.23
Girls	7.80 (6.94–8.66)	7.73 (6.57–8.90)	7.47 (6.55–8.39)	7.39 (6.70–8.08)	−0.41	0.47
Non-hispanic black						
All	6.85 (6.17–7.53)	6.75 (6.07–7.43)	6.75 (5.82–7.69)	6.99 (6.12–7.87)	0.14	0.48
Boys	6.03 (5.42–6.63)	5.51 (4.92–6.10)	5.46 (4.42–6.51)	5.86 (4.66–7.06)	−0.17	0.91
Girls	7.76 (6.70–8.82)	8.08 (7.09–9.07)	8.16 (6.86–9.46)	8.16 (7.11–9.22)	0.40	0.22
Non-hispanic Asian						
All	6.17 (5.60–6.75)	6.04 (5.59–6.49)	5.85 (5.25–6.46)	6.83 (6.14–7.52)	0.66	0.17
Boys	5.68 (4.82–6.54)	5.95 (5.08–6.81)	5.12 (4.43–5.81)	6.79 (6.00–7.59)	1.11	0.12
Girls	6.74 (6.04–7.44)	6.13 (5.57–6.70)	6.75 (6.02–7.48)	6.86 (5.96–7.76)	0.12	0.48
Other						
All	7.50 (5.99–9.02)	7.85 (7.04–8.65)	7.32 (6.48–8.15)	7.07 (6.08–8.07)	−0.43	0.64
Boys	7.33 (4.64–10.01)	6.86 (5.82–7.891)	7.08 (5.81–8.35)	6.38 (5.29–7.47)	−0.95	0.70
Girls	7.70 (6.91–8.48)	8.95 (8.07–9.84)	7.58 (6.84–8.31)	8.00 (6.41–9.60)	0.30	0.70

[a]*Absolute increase between NHANES 2011–2012 and NHANES 2017–2018.* [b]*Time trends in mean trunk fat from 2011–2012 to 2017–2018 were examined with a multiple linear regression model, with adjustment for age, gender, and race or ethnicity, when applicable. Bold values are the statistically significant p-values.*

WHtR, and age-adjusted prevalence of abdominal obesity, overall and stratified by gender and race or ethnicity, among those aged 2–19 years. Data for children and adolescents were age-standardized to the 2000 US census using age groups 2–5, 6–11, and 12–19 years (10).

Furthermore, DXA was conducted among participants aged 8–59 years. Data for age groups of 8 years were not available from the census 2,000 population data (10), and we could not obtain age-adjusted data among age groups of 8 years. Therefore, the estimates in measures of central body fat by DXA, such as trunk fat and trunk fat percentage, were unadjusted and analyzed in the following two age groups: 8–11 and 12–19 years.

The 90th percentiles of waist circumference by age and gender in NHANES III were used to estimate prevalence of abdominal obesity (11). The WHtR value of 0.5 was also used as a cutoff point of abdominal obesity for youth aged 6–19 years (12, 13). In children aged 2–5 years, a WHtR value of 0.5 may overestimate the prevalence of abdominal obesity; thus, this cutoff value was not suitable to children aged 2–5 years in the present study. Linear regression models were performed to investigate linear trends over time with survey periods as a continuous independent variable in models, with adjustment for age, gender, and race or ethnicity, when applicable.

All statistical analyses were performed using survey modules of SAS software version 9.4 (SAS Institute, Cary, NC). A two-sided *p*-value of.05 was used to estimate statistical significance.

RESULTS

The present study included 13,306 children and adolescents aged 2–19 years from NHANES 2011–2018 (**Appendix Table 1**). Among those, 7,367 children and adolescents aged 8–19 years had valid measurement of body composition by DXA. There are different sample sizes for different measurement methods: 7,239 for trunk fat and trunk fat percentage, 12,478 for waist circumference, and 12,448 for WHtR (**Figure 1**).

TABLE 2 | Trends in mean trunk fat percentage (95% CI) by race or ethnicity among US children aged 8–19 y, NHANES 2011–2012 to 2017–2018.

Variable	Mean trunk fat percentage (%)				Absolute Increase[a]	*P* for trend[b]
	2011–12 (*n* = 1,828)	2013–14 (*n* = 1,996)	2015–16 (*n* = 1,907)	2017–18 (*n* = 1,508)		
Overall	25.6 (24.9–26.3)	26.1 (25.2–27.0)	25.9 (25.0–26.9)	26.2 (25.7–26.7)	0.6	0.18
Age, y						
8–11	26.0 (25.0–27.0)	26.4 (24.8–28.1)	26.2 (25.2–27.3)	26.2 (24.7–27.8)	0.2	0.77
12–19	25.4 (24.6–26.3)	26.0 (24.9–27.2)	25.8 (24.8–26.8)	26.2 (25.8–26.5)	0.8	0.12
Gender						
Boys	22.1 (21.3–22.8)	23.0 (22.3–23.7)	22.8 (21.6–24.0)	23.1 (22.4–23.8)	1.0	0.08
Girls	29.3 (28.4–30.3)	29.6 (28.3–31.0)	29.5 (28.5–30.6)	29.8 (28.8–30.7)	0.5	0.60
RACE/ETHNICITY						
Mexican American						
All	27.8 (26.6–29.0)	28.5 (27.5–29.4)	30.0 (29.3–30.6)	29.3 (27.8–30.8)	1.5	**0.04**
Boys	24.7 (23.5–25.9)	25.1 (24.2–26.0)	27.0 (25.9–28.1)	26.1 (24.0–28.1)	1.4	0.08
Girls	31.5 (30.0–33.1)	32.4 (31.5–33.2)	33.6 (32.2–35.0)	33.0 (31.5–34.5)	1.5	0.13
Non-hispanic white						
All	25.2 (24.2–26.3)	25.6 (24.4–26.8)	25.1 (23.9–26.4)	25.0 (23.9–26.2)	−0.2	0.81
Boys	21.5 (20.4–22.7)	22.9 (21.9–23.9)	22.2 (20.7–23.7)	22.2 (20.8–23.5)	0.7	0.70
Girls	28.9 (27.5–30.3)	28.7 (26.8–30.6)	28.5 (27.0–30.0)	28.3 (26.8–29.8)	−0.6	0.54
Non-hispanic black						
All	23.9 (22.8–25.0)	24.5 (23.3–25.6)	23.9 (22.5–25.4)	25.1 (23.5–26.8)	1.2	0.30
Boys	20.4 (19.5–21.4)	20.2 (19.3–21.1)	19.5 (17.5–21.5)	20.7 (18.4–23.0)	0.3	0.98
Girls	27.8 (26.1–29.5)	29.2 (27.4–31.0)	28.7 (26.5–30.8)	30.0 (28.9–31.2)	2.2	0.07
Non-hispanic Asian						
All	25.4 (24.2–26.7)	25.5 (23.8–27.2)	24.9 (23.5–26.3)	27.3 (26.3–28.2)	1.9	0.08
Boys	22.1 (20.4–23.9)	23.2 (21.0–25.3)	21.4 (19.4–23.5)	25.4 (24.1–26.8)	3.3	**0.04**
Girls	29.3 (26.9–31.6)	27.9 (26.5–29.2)	29.1 (28.0–30.1)	29.1 (27.7–30.4)	−0.2	0.67
Other						
All	26.9 (24.8–29.0)	27.4 (25.6–29.2)	27.3 (26.0–28.6)	27.1 (25.3–29.0)	0.2	0.72
Boys	23.4 (20.7–26.0)	23.4 (21.3–25.5)	24.5 (22.6–26.4)	23.8 (21.5–26.1)	0.4	0.67
Girls	30.8 (29.3–32.3)	31.6 (30.5–32.8)	30.4 (28.9–31.9)	30.8 (28.8–32.9)	0.0	0.80

[a]Absolute increase between NHANES 2011–2012 and NHANES 2017–2018. [b]Time trends in mean trunk fat percentage from 2011–2012 to 2017–2018 were examined with a multiple linear regression model, with adjustment for age, gender, and race or ethnicity, when applicable. Bold values are the statistically significant p-values.

In the sample of valid measurement of body composition by DXA (**Appendix Table 2**), the weighted mean age was 13.43 years (standard error, 0.06); 3,574 participants were girls (weighted proportion, 48.09%); 1,526 were of Mexican American ancestry (15.42%), 1,963 were of non-Hispanic white ancestry (53.21%), 1,869 were of non-Hispanic black ancestry (13.95%), and 746 were of non-Hispanic Asian ancestry (4.50%) (**Appendix Table 2**). Population characteristics and sample sizes are slightly different across survey cycles (**Appendix Tables 1, 2**).

Trunk Fat

Among children aged 8–19 years, the unadjusted mean trunk fat remained unchanged across surveys between 2011 and 2018 ($P = 0.50$) and by age, gender, and racial or ethnic group, except for the Mexican American group, whose mean trunk fat significantly increased ($P = 0.03$) (**Table 1**). Moreover, all girls aged 8–19 years had significantly higher mean trunk fat than all boys aged 8–19 years.

Trunk Fat Percentage

Among US youth aged 8–19 years, the unadjusted mean trunk fat percentage remained stable across surveys from 2011 to 2018 ($P = 0.18$) and by age and gender (**Table 2** and **Figure 2**). However, all girls aged 8–19 years had significantly higher mean trunk fat percentage than all boys aged 8–19 years.

In the subgroup analyses by race/ethnicity, a linear increase trend in mean trunk fat percentage for youth aged 8–19 years was observed in the Mexican American group ($P = 0.04$) and non-Hispanic Asian boys ($P = 0.04$) from 2011 to 2018, but not in all other race/ethnicity groups.

Waist Circumference

Among children aged 2–19 years, age-adjusted mean waist circumference held steady across surveys from 2011 to 2018 ($P = 0.61$) and by age, gender, and racial or ethnic group, except for the Mexican American group ($P = 0.03$) and non-Hispanic Asian

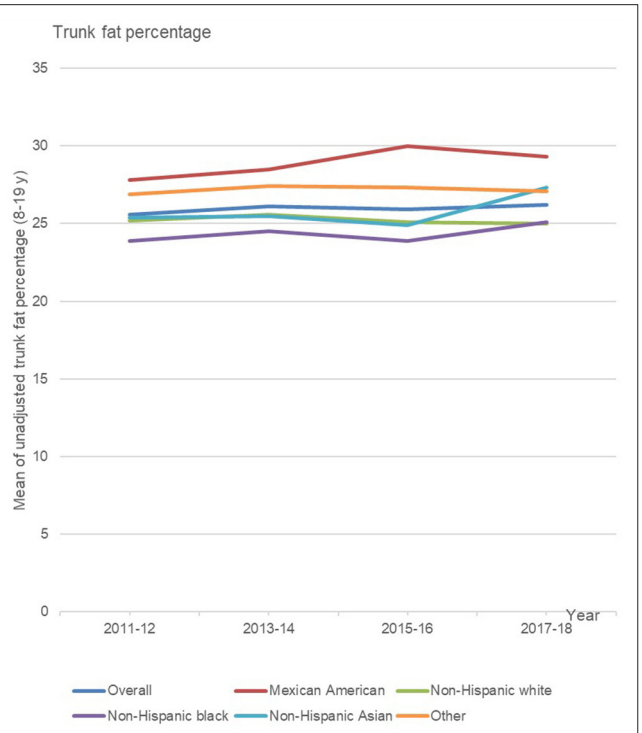

FIGURE 2 | Trends in mean trunk fat percentage by race or ethnicity among US children aged 8–19 y, NHANES 2011–2012 to 2017–2018.

boys ($P = 0.02$), whose age-adjusted mean waist circumference significantly increased (**Appendix Table 3**).

Waist-to-Height Ratio

Between 2011–2012 and 2017–2018, age-adjusted mean WHtR remained stable across surveys among children aged 2–19 years ($P = 0.46$) and by age, gender, and racial or ethnic group, except for the Mexican American group ($P = 0.02$), whose age-adjusted mean WHtR significantly increased (**Appendix Table 4**).

Pearson Correlation Analysis

There was a high correlation between waist circumference and trunk fat in both sexes in 2011–2012 and 2017–2018 (all $r \geq 0.8$). The correlation between WHtR and trunk fat in boys and girls also was statistically significant in 2011–2012 and 2017–2018 ($P < 0.01$) (**Appendix Table 5**).

Abdominal Obesity (by Waist Circumference)

For children aged 2–19 years, the overall age-adjusted prevalence of abdominal obesity showed no significant changes from 18.6% (95% CI, 15.5–21.6%) in 2011–2012 to 21.1% (95% CI, 18.9–23.3%) in 2017–2018 ($P = 0.25$) (**Table 3** and **Figure 3**). However, there was an increasing linear trend in prevalence of abdominal obesity among all boys aged 2–19 years ($P = 0.004$), but not among girls during 2011–2018.

In the subgroup analyses by race/ethnicity, prevalence of abdominal obesity for those aged 2–19 years marginally increased in Mexican American boys ($P = 0.06$), non-Hispanic white boys ($P = 0.06$), and non-Hispanic Asian boys ($P = 0.06$), but not in all other race/ethnicity groups from 2011 to 2018 (**Table 3**; **Figure 3**).

Abdominal Obesity (by WHtR)

For youths aged 6–19 years, age-adjusted prevalence of abdominal obesity did not significantly change from 2011 to 2018 ($P = 0.25$) and by age, gender, and racial or ethnic group, except for non-Hispanic Asian boys, whose WHtR marginally increased ($P = 0.06$) (**Appendix Table 6**).

DISCUSSION

From 2011–2012 to 2017–2018, there were no significant changes among children aged 8–19 years in mean trunk fat and trunk fat percentage, or among those aged 2–19 years in mean waist circumference and WHtR. During the same time periods, prevalence of abdominal obesity by waist circumference among children aged 2–19 years, and abdominal obesity by WHtR among those aged 6–19 years remained stable. Among the Mexican American group, there was a significant increasing trend in mean trunk fat and trunk fat percentage among children aged 8–19 years, and mean waist circumference and WHtR among children aged 2–19 years from 2011 to 2018. For non-Hispanic Asian boys, there is a significant increasing trend among children aged 8–19 years in mean trunk fat percentage and among those aged 2–19 years in mean waist circumference during 2011–2018.

This study provided the recent national estimates and trends of central body fat measured by DXA (trunk fat and trunk fat percentage) and anthropometry (waist circumference and WHtR). Some studies found that there are strong correlations between trunk fat mass evaluated by DXA and intra-abdominal fat evaluated by computed tomography or magnetic resonance imaging (7). Although body mass index is the most frequently used method to evaluate obesity, it does not always reflect true body fatness. The health complications related to obesity are associated with increased body fat deposition rather than with body weight *per se*. In the present study, for Mexican American youth and non-Hispanic Asian boys, there is an increasing trend in mean trunk fat percentage among those aged 8–19 years and in mean waist circumference among children aged 2–19 years during 2011–18. This study found that there was a high correlation between waist circumference and trunk fat in both sexes in 2011–2012 and 2017–2018. In preschool-aged children, waist circumference can identify effectively between children with low and high levels of trunk fat mass, as measured by DXA (14).

Moreover, this study provided the most recent national estimates and trends of abdominal obesity among US children and adolescents. In 2017 to 2018, overall prevalence of abdominal obesity is still high, being 21.1% (defined by waist circumference) in participants aged 2–19 years and 36.2% (defined by WHtR) in those aged 6–19 years. For boys aged 2–19 years, there is an increasing trend in prevalence of abdominal obesity by waist circumference. This suggested that central fatness among boys

TABLE 3 | Trends in age-adjusted prevalence of abdominal obesity by waist circumference among US children aged 2–19 y, NHANES 2011–2012 to 2017–2018.

Variable	Age-adjusted prevalence of abdominal obesity by waist circumference (%)				Absolute Increase[a]	*P* for trend[b]
	2011–12	2013–14	2015–16	2017–18		
Overall	18.6 (15.5–21.6)	20.2 (17.6–22.7)	19.2 (16.2–22.2)	21.1 (18.9–23.3)	2.5	0.25
Age, y						
2–7	14.6 (12.4–16.9)	17.7 (14.5–20.8)	16.7 (13.0–20.4)	18.4 (14.1–22.6)	3.8	0.15
8–11	20.0 (13.9–26.2)	20.7 (15.5–25.8)	17.5 (13.5–21.6)	22.1 (18.1–26.1)	2.1	0.74
12–19	20.7 (16.4–25.0)	21.8 (17.1–26.5)	21.8 (17.5–26.2)	22.4 (19.2–25.7)	1.7	0.51
Gender						
Boys	16.1 (13.0–19.2)	18.8 (15.7–21.9)	19.0 (14.7–23.4)	22.7 (19.6–25.8)	6.6	**0.004**
Girls	21.1 (17.3–24.9)	21.7 (18.6–24.8)	19.3 (16.6–22.0)	19.4 (15.5–23.4)	−1.7	0.39
RACE/ETHNICITY						
Mexican American						
All	24.6 (22.4–26.7)	26.5 (22.2–30.8)	27.0 (24.2–29.7)	27.4 (22.1–32.6)	2.8	0.32
Boys	24.1 (20.0–28.2)	23.4 (18.7–28.1)	27.7 (22.3–33.2)	28.7 (23.1–34.2)	4.6	0.06
Girls	25.3 (22.2–28.3)	29.7 (23.5–36.0)	26.3 (19.8–32.8)	25.8 (18.7–32.9)	0.5	0.86
Non-hispanic white						
All	17.4 (12.0–22.7)	19.3 (15.6–22.9)	16.6 (13.3–19.9)	19.1 (15.9–22.2)	1.7	0.83
Boys	13.5 (8.7–18.3)	18.3 (13.5–23.0)	16.9 (11.8–22.0)	20.8 (15.7–25.9)	7.3	0.06
Girls	21.5 (14.6–28.5)	20.4 (15.7–25.0)	16.2 (12.1–20.4)	17.2 (10.7–23.6)	−4.3	0.22
Non-hispanic black						
All	18.2 (15.3–21.1)	15.3 (13.1–17.4)	18.6 (12.6–24.5)	21.3 (16.1–26.5)	3.1	0.19
Boys	15.9 (13.4–18.4)	11.7 (7.8–15.5)	14.2 (7.8–20.5)	16.2 (10.7–21.7)	0.3	0.75
Girls	20.6 (15.4–25.9)	19.2 (16.4–21.9)	23.1 (15.5–30.7)	26.5 (20.2–32.8)	5.9	0.09
Non-hispanic Asian						
All	7.9 (4.5–11.3)	9.8 (6.2–13.5)	9.5 (4.9–14.1)	11.2 (8.2–14.3)	3.3	0.14
Boys	9.1 (5.6–12.5)	14.3 (6.2–22.4)	8.8 (3.4–14.3)	17.0 (12.2–21.8)	7.9	0.06
Girls	6.7 (1.3–12.2)	5.3 (2.7–8.0)	9.9 (4.2–15.6)	5.3 (2.6–8.0)	−1.4	0.91
Other						
All	20.4 (16.0–24.9)	24.8 (17.6–31.9)	24.1 (18.5–29.8)	23.3 (16.9–29.8)	2.9	0.59
Boys	21.0 (16.3–25.6)	23.6 (14.0–33.2)	26.1 (18.4–33.8)	29.5 (21.2–37.7)	8.5	0.10
Girls	19.8 (12.5–27.1)	26.0 (17.8–34.3)	21.9 (17.1–26.8)	17.2 (9.4–24.9)	−2.6	0.44

[a]Absolute increase between NHANES 2011–2012 and NHANES 2017–2018. [b]Time trends in age-adjusted prevalence of abdominal obesity by waist circumference from 2011–2012 to 2017–2018 were examined with a multiple linear regression model, with adjustment for age, gender, and race or ethnicity, when applicable. Bold values indicates that P values.

were increasing more rapidly than among girls. Considering the importance of body fat distribution for health at later life (6), there is an urgent need to improve their lifestyle to reduce obesity for US children and adolescents, especially for Mexican American youth and non-Hispanic Asian boys.

Furthermore, this study showed that girls aged 8–19 years had significantly higher mean trunk fat percentage than boys aged 8–19 years. Our results are consistent with another study by Taylor et al. (14) in preschool-aged children. That shows that girls stored proportionately more of their body fat in the truncal region. Nevertheless, several studies reported no difference (15, 16). Whether there were significant gender differences in mean trunk fat percentage at this age, it is still uncertain. In addition, a study among Asian adults showed that women had higher prevalence of central obesity than men, but linear trends in the prevalence of central obesity could not be evaluated in that study (17). Gender differences in regional adiposity should be further confirmed by future research.

Differences in obesity prevalence in children by race/ethnicity origin have been published in the United States (18). Some studies indicates that Asians may have more body fat than whites, especially when body mass indexes are lower (5). The present study reported prevalence of abdominal obesity in non-Hispanic Asian youth and found that in 2011–12, 9.1% of non-Hispanic Asian boys were abdominal obese, which increased to 17.0% in 2017–18. Also, among US adults, a previous study indicated that age-adjusted mean waist circumference was significantly increased in non-Hispanic Asian men (19). Although Asian children is generally considered healthy, they are now at a great risk of abdominal obesity especially in non-Hispanic Asian boys.

The main strength of this study was the nationally representative nature of US children and adolescents. The trends for central obesity in NHANES 1999–2004 and 2005–2012 were previously reported, but we reported the trends for 4 recent survey cycles (2011–12, 2013–14, 2015–16, and 2017–18). In addition, data from NHANES 2011–18 provides an opportunity

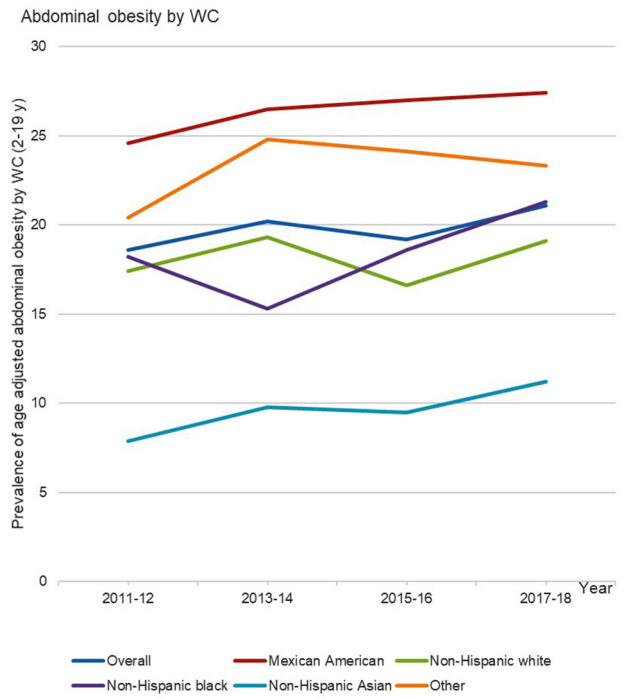

FIGURE 3 | Trends in age-adjusted prevalence of abdominal obesity by waist circumference among US children aged 2–19 y, NHANES 2011–2012 to 2017–2018.

to separately assess the trends in central adiposity measures and abdominal obesity in non-Hispanic Asians and other races or ethnicities.

This study has several limitations. We assessed central adiposity using waist circumference, WHtR, trunk fat and trunk fat percentage rather than directly measured visceral fat. Additionally, there are still no commonly agreed cut-offs to identify children with excess adiposity using DXA adiposity measures such as trunk fat and trunk fat percentage.

CONCLUSION

This nationally representative study provides useful information of the most recent national estimates and trends in measures of central body fat by DXA, including trunk fat and trunk fat percentage, and abdominal obesity among US children and adolescents from NHANES 2011–2018. Non-Hispanic Asian boys and Mexican American youth had a significant increase in measures of abdominal and trunk fat accumulation, which warrants further attention.

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/s.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by NHANES procedures were approved by a human subject review board. Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

AUTHOR CONTRIBUTIONS

JL, GZ, and XW contributed to the conception, design of the study, and critically revised the manuscript. YT and NJ were main responsible for data collection. JL, YT, and NJ contributed to the statistical analysis and interpretation of the data. JL, YZ, and GZ assisted drafting the manuscript and literature research. All gave final approval and agreed to submit the work.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fped.2022.903413/full#supplementary-material

REFERENCES

1. Ragino YI, Stakhneva EM, Polonskaya YV, Kashtanova EV. The role of secretory activity molecules of visceral adipocytes in abdominal obesity in the development of cardiovascular disease: a review. *Biomolecules.* (2020) 10:18. doi: 10.3390/biom10030374
2. Li C, Ford ES, Mokdad AH, Cook S. Recent trends in waist circumference and waist-height ratio among US children and adolescents. *Pediatrics.* (2006) 118:2006–1062. doi: 10.1542/peds.2006-1062
3. Tinsley GM, Smith-Ryan AE, Kim Y, Blue MNM, Nickerson BS, Stratton MT, et al. Fat-free mass characteristics vary based on sex, race, and weight status in US adults. *Nutr Res.* (2020) 81:58–70. doi: 10.1016/j.nutres.2020.07.002
4. Wulan SN, Westerterp KR, Plasqui G. Dietary and 24-h fat oxidation in Asians and whites who differ in body composition. *Am J Clin Nutr.* (2012) 95:1335–41. doi: 10.3945/ajcn.111.031369
5. Deurenberg P, Deurenberg-Yap M, Guricci S. Asians are different from caucasians and from each other in their body mass index/body fat per cent relationship. *Obes Rev.* (2002) 3:141–6. doi: 10.1046/j.1467-789X.2002.00065.x

6. Goran MI, Gower BA. Relation between visceral fat and disease risk in children and adolescents. *Am J Clin Nutr.* (1999) 70:149S–156S. doi: 10.1093/ajcn/70.1.149s
7. Goran MI, Gower BA, Treuth M, Nagy TR. Prediction of intra-abdominal and subcutaneous abdominal adipose tissue in healthy pre-pubertal children. *Int J Obes Relat Metab Disord.* (1998) 22:549–58. doi: 10.1038/sj.ijo.0800624
8. Johnson CL, Paulose-Ram R, Ogden CL, Carroll MD, Kruszon-Moran D, Dohrmann SM, et al. National health and nutrition examination survey: analytic guidelines, 1999-2010. *Vital Health Stat.* (2013) 2:1–24. Available online at: https://pubmed.ncbi.nlm.nih.gov/25090154/
9. Chen TC, Clark J, Riddles MK, Mohadjer LK, Fakhouri T. National health and nutrition examination survey, 2015-2018: sample design and estimation procedures. *Vital Health Stat.* (2020) 1–35. Available online at: https://pubmed.ncbi.nlm.nih.gov/33663649/
10. Klein RJ, Schoenborn CA. Age adjustment using the 2000 projected U.S. population. *Healthy People.* (2001) 1–10. doi: 10.1037/e583772012-001 Available online at: https://pubmed.ncbi.nlm.nih.gov/11676466/
11. Cook S, Weitzman M, Auinger P, Nguyen M, Dietz WH. Prevalence of a metabolic syndrome phenotype in adolescents: findings from the third national health and nutrition examination survey, 1988-1994. *Arch Pediatr Adolesc Med.* (2003) 157:821–7. doi: 10.1001/archpedi.157.8.821
12. McCarthy HD, Cole TJ, Fry T, Jebb SA, Prentice AM. Body fat reference curves for children. *Int J Obes.* (2006) 30:598–602. doi: 10.1038/sj.ijo.0803232
13. Mushtaq MU, Gull S, Abdullah HM, Shahid U, Shad MA, Akram J. Waist circumference, waist-hip ratio and waist-height ratio percentiles and central obesity among Pakistani children aged five to twelve years. *BMC Pediatr.* (2011) 11:1471–2431. doi: 10.1186/1471-2431-11-105
14. Taylor RW, Williams SM, Grant AM, Ferguson E, Taylor BJ, Goulding A. Waist circumference as a measure of trunk fat mass in children aged 3 to 5 years. *Int J Pediatr Obes.* (2008) 3:226–33. doi: 10.1080/17477160802030429
15. Arfai K, Pitukcheewanont PD, Goran MI, Tavare CJ, Heller L, Gilsanz V. Bone, muscle, and fat: sex-related differences in prepubertal children. *Radiology.* (2002) 224:338–44. doi: 10.1148/radiol.2242011369
16. Nagy TR, Gower BA, Trowbridge CA, Dezenberg C, Shewchuk RM, Goran MI. Effects of gender, ethnicity, body composition, and fat distribution on serum leptin concentrations in children. *J Clin Endocrinol Metab.* (1997) 82:2148–52. doi: 10.1210/jc.82.7.2148
17. Liu X, Yang C, Boucher NL, Rothberg AEJBPH. Prevalence and change of central obesity among US Asian adults: NHANES 2011–2014. *BMC Public Health*. (2017) 17:678. doi: 10.1186/s12889-017-4689-6
18. Ogden CL, Carroll MD, Lawman HG, Fryar CD, Kruszon-Moran D, Kit BK, et al. Trends in obesity prevalence among children and adolescents in the United States, 1988-1994 through 2013-2014. *JAMA.* (2016) 315:2292–9. doi: 10.1001/jama.2016.6361
19. Liu B, Du Y, Wu Y, Snetselaar LG, Wallace RB, Bao WJB. Trends in obesity and adiposity measures by race or ethnicity among adults in the United States 2011-18: population based study. *BMJ.* (2021) 372:n365. doi: 10.1136/bmj.n365

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ORIGINAL RESEARCH
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New Insights in Cytokines in Childhood Obesity: Changes in TWEAK and CD163 After a 2-Year Intervention Program in Prepubertal Children With Obesity

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Objective: Obesity is characterized by a low-grade inflammatory state in adipose tissue. Tumor Necrosis Factor Weak Inducer of Apoptosis (TWEAK) and Cluster of Differentiation 163 (CD163) are cytokines potentially involved in the pathogenesis of obesity. Little is known about them in children. The aim of this study was to observe serum levels of TWEAK and CD163 in prepubertal children with obesity compared to lean, and to evaluate its changes after a 2-year intervention program in children with obesity.

Methods: Case-control study with a prospective follow-up of cases for 2 years in a referral pediatric endocrine outpatient centre. Seventy-three prepubertal children with obesity, and forty-seven age- and gender-matched lean controls were studied. Sixty-two cases finished the program. Anthropometric parameters, Homeostatic Model Assessment for Insulin Resistance (HOMA-IR), lipid profile, and concentrations of TWEAK and CD163 were determined. Children with obesity were re-evaluated after a 2-year intervention program consisting of diet and exercise. Weight loss was considered if z-score Body Mass Index (BMI) decreased at least 0.5 Standard Deviations (SD).

Results: We observed higher CD163 levels in children with obesity compared to controls. No significant differences were observed in TWEAK and CD163/TWEAK ratio at baseline. After the 2-year intervention program, TWEAK levels were higher and CD163/TWEAK ratio was lower in children with weight loss than those without weight loss. CD163 decreased in both groups.

Conclusion: TWEAK and CD163 seem to have a role in the pathogenesis of obesity in prepubertal children.

Keywords: TWEAK, CD163, childhood, obesity, prepubertal

INTRODUCTION

The prevalence of childhood obesity has increased dramatically in recent decades, becoming a global public health problem (1). Pathological conditions associated with obesity, such as diabetes, hyperlipidemia, or hypertension (2), have been observed mainly in adults. As the prevalence of childhood obesity increases, these conditions are becoming more common among infants and teenagers (3–8).

Obesity is associated with a chronic inflammatory state in adipose tissue (9–11). In this context, we can observe altered levels of molecules involved in the regulation of inflammation, called cytokines (12–14).

Research on cytokines has increased during the last years in order to identify risk and protective factors of cardiovascular disease and to generate potential target treatments (15, 16).

Tumor Necrosis Factor Weak Inducer of Apoptosis (TWEAK), also known as Tumor Necrosis Factor (TNF) ligand superfamily member 12, is a cytokine involved in multiple biological functions (12, 17, 18). It seems to play a protective role in the regulation of obesity and insulin resistance (15, 19, 20). It was observed that serum levels of soluble TWEAK are lower in adults with severe obesity, and they increase in these patients after a significant weight loss (21).

Cluster of Differentiation 163 (CD163) is a macrophage-specific protein that increases in the context of inflammation (22). *In vitro*, it has been identified as a scavenger receptor of TWEAK, promoting its degradation (23). Serum levels of CD163 are higher in obesity and insulin resistance, being a strong predictor of type 2 diabetes in adults (24). In cardiovascular diseases, a high CD163/TWEAK ratio was found (25).

Little is known about TWEAK and CD163 in children. The main objective of the study was to evaluate if a significant weight loss in children with obesity may induce changes in these parameters after a 2-year lifestyle intervention program. In addition, the investigation aimed to evaluate serum levels of TWEAK, CD163, and CD163/TWEAK ratio in prepubertal children with obesity compared to lean children.

PATIENTS AND METHODS

Seventy-three prepubertal children with obesity were included, from an outpatient long-term intervention program over a period of 2 years. Inclusion criteria were the presence of obesity as defined by body mass index (BMI) > 2 standard deviation (SD) scores for age and sex upon Spanish normative charts (26), aged between 6 and 10 years and prepubertal according to Tanner staging (27, 28). Controls were forty-seven prepubertal healthy children attending for preoperative blood tests before minor surgery. They were matched by age and gender. Exclusion criteria were the presence of endocrinopathies, obesity-associated syndrome, and any infectious or inflammatory disease in the past 10 days, or taking medication that affected weight, lipid metabolism, or arterial blood pressure (BP). Written informed consent was obtained from all patients' parents, and all investigations followed the Helsinki Declaration. The study was approved by the Ethics Committee of our Institution (reference code 2004104).

Baseline Clinical Evaluation

Detailed medical, personal, and family history of obesity and cardiometabolic risk was obtained from all subjects, including birth weight and length for gestational age. A complete physical examination was performed. Height was measured by a Harpenden stadiometer to the nearest 0.1 cm and body weight by balance scale to the nearest 0.1 kg. Waist circumference was measured with a tape at the middle point between the last rib and the superior iliac crest, adjusted to the nearest 0.1 cm, and compared with an age and sex-reference population (29). All measurements were performed for duplication by the same investigator, with the patient in light clothes and without shoes. The mean of the two determinations was used for calculations. Adiposity was evaluated by BMI (calculated as weight in kilograms divided by the square of height in meters). Pubertal development was assessed by direct physical examination according to Tanner staging. Blood pressure was measured by triplicate with the Critikon Dinamap 8100 automatic system (Johnson-Johnson Company, Tampa, FL, USA), with an appropriate sized cuff and after at least ten minutes resting in supine position. The lowest BP value was recorded and evaluated using the percentiles of the International Task Force for Blood Pressure (30).

Obesity degree was fixed with z-BMI using the LMS method (31). We also applied the obesity criteria by Cole (International Obesity Task Force) (32) to our BMI data.

Baseline Metabolic Evaluation

Blood samples were obtained after a 12-hours overnight fast. Concentrations of soluble TWEAK, soluble CD163, alanine aminotransferase (ALT), uric acid, glucose, insulin, cholesterol and triglycerides were determined. Likewise, a standard 2-hours oral glucose tolerance test was performed in subjects with obesity.

Samples were stored at -80°C until their analysis. Plasma glucose was measured by the glucose hexokinase method, insulin by an electrochemiluminescent method, cholesterol and its fractions by cholesterol esterase/oxidase, and triglycerides by lipase/glycerol kinase (Roche Diagnostics, Mannheim, Germany). Intra- and inter-assay coefficient of variation (CV) values were 1.9–2.1% for glucose and 2.6–2.8% for insulin, respectively.

Plasma soluble TWEAK was measured by an enzyme-linked immunoassay (ELISA) technique with a sensitivity of 0.02 pg/mL (intra-assay CV <10%, inter-assay CV <9%). Plasma soluble CD163 was measured by ELISA with a sensitivity of 0.1 ng/mL (intra-assay CV <6%, inter-assay CV <7%). Impaired glucose metabolism was defined according to the American Diabetes Association criteria (33). Insulin resistance was evaluated using the Homeostatic Model Assessment for Insulin Resistance $\left[HOMA\ IR = \frac{fasting\ insulin\ (\mu U/mL) * fasting\ glucose\ (\mu mol/L)}{22{,}5}\right]$ (34).

Intervention and Follow-Up

After the baseline evaluation, 73 prepubertal children with obesity started a lifestyle intervention program that included a balanced norm caloric diet adjusted by age and a personally adapted exercise program. The diet contained 30% of energy intake from fat, 15% from protein, and 55% from carbohydrate (5% as sugar). A plan of 30–45 minutes of moderate exercise three times a week was negotiated. Television and video games were limited to a maximum of 2 hours a day. Follow-up visits were scheduled every 4 months.

After 2 years, a clinical and metabolic evaluation was performed with the same parameters as in the baseline evaluation. We considered a significant weight loss if the z-score of BMI had decreased at least 0.5 SD (35).

Statistical Analysis

Data were expressed as mean ± SD for quantitative variables and as percentages for categorical variables. Logarithmic transformation before the analysis was used when variables did not follow a normal distribution. Student's t-test or Mann-Whitney U test were used for comparing differences between groups. Paired t-test or Wilcoxon test was used to compare variables before and after the intervention program. Bivariate correlations were evaluated with Pearson's and Spearman's coefficients as appropriate. Furthermore, some multivariate linear regression models were used. TWEAK, CD163 or CD163/TWEAK ratio as dependent variables and age, sex, weight status (BMI), HOMA-IR and lipid profile, as independent variables. A p value < 0.05 was considered significant. Analyses were performed with SAS v9.4, SAS Institute Inc., Cary, NC, USA.

RESULTS

73 children with obesity and 47 controls were included in the study. Characteristics of subjects are shown in **Table 1**. Comparing cases and controls, CD163 levels were higher in children with obesity. No statistically significant differences were found in TWEAK and CD163/TWEAK ratio.

After a 2-year follow-up of the cases, 62 completed the study and 11 dropped out (15%). 31 patients achieved a significant weight loss. Comparing patients with and without weight loss after the intervention program, TWEAK decreased in both groups after the intervention program. However, we observed that this parameter was higher in patients who achieved weight loss, before and after the intervention program. Serum levels of CD163 decreased after 2 years in both groups, and the decrease was more pronounced in patients with weight loss (**Figure 1**). CD163/TWEAK ratio was higher in children without weight loss, and it increased after 2 years in this group (**Figure 2**). Data are shown in **Table 2**.

TABLE 1 | Baseline Subjects Characteristics.

	With obesity (n = 73)	Control (n = 47)	*p*
Sex	37 girls/36 boys	16 girls/31 boys	0.073
Age (years)	8.03 ± 1.08	7.74 ± 1.35	0.22
BMI (Kg/m^2)	26.5 ± 3.07	16.2 ± 1.41	<0.001
z-BMI (SD)	4.76 ± 1.67	-0.27 ± 0.74	<0.001
Birth Weight (gr)	3337 ± 599.5	3194 ± 539.9	0.28
Birth Length (cm)	50.0 ± 2.61	50.1 ± 1.60	0.3
Familiar diabetes 2	35 (47%)	11 (23%)	0.01
Father BMI (Kg/m^2)	28.6 ± 3.93	25.2 ± 2.31	0.003
Mother BMI (Kg/m^2)	29.1 ± 7.83	23.1 ± 3.22	0.001
Acanthosis	23 (31%)	0 (0%)	<0.001
Waist (cm)	81.3 ± 8.5	57.5 ± 6.05	0.035
SBP (mmHg)	109.2 ± 14.5	104.1 ± 9.60	0.026
DBP (mmHg)	63.4 ± 8.32	64.1 ± 9.25	0.672
Glucose (mg/dL)	83.7 ± 7.78	83.3 ± 5.04	0.804
Insulin (μU/mL)[a]	10 (5-15)	4 (2-5)	<0.001
HOMA-IR[a]	1.97 (1.02-3.14)	0.80 (0.40-1.08)	<0.001
Cholesterol (mg/dL)	155.5 ± 27.9	157.8 ± 31.4	0.621
Triglycerides(mg/dL)[a]	69 (52.5-87.5)	48 (40-54)	<0.001
LDLc (mg/dL)	93.5 ± 24.1	82.6 ± 24.7	0.032
HDLc (mg/dL)	51.3 ± 12.8	65.5 ± 14.7	<0.001
Uric acid (mg/dL)	4.12 ± 0.85	3.41 ± 0.626	<0.001
ALT (U/L)	20.1 ± 6.68	19.2 ± 6.97	0.479
TWEAK (pg/mL)	1479.1 ± 1347.7	1184.9 ± 771.8	0.244
CD163 (ng/mL)	210.2 ± 48.9	187.2 ± 31.6	0.012
CD163/TWEAK ratio	0.14 ± 0,07	0.15 ± 0,10	0.255

Data are presented as mean ± SD or percentages.

ALT, alaninetransferase; BMI, body mass index;CD163, Cluster of differentiation 163; DBP, diastolic blood pressure; HDLc, high density lipoprotein cholesterol; HOMA-IR, homeostatic model insulin resistance index; LDLc, low density lipoprotein cholesterol; SBP, systolic blood pressure; TWEAK, Tumor Necrosis Factor weak inducer of apoptosis.

[a]*Median (interquartile range).*

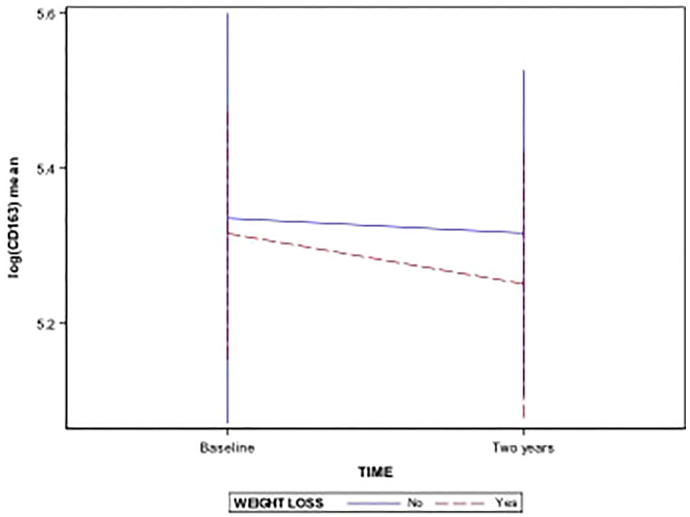

FIGURE 1 | CD163 decreased after 2 years in both groups, and the decrease was more pronounced in patients with weight loss.

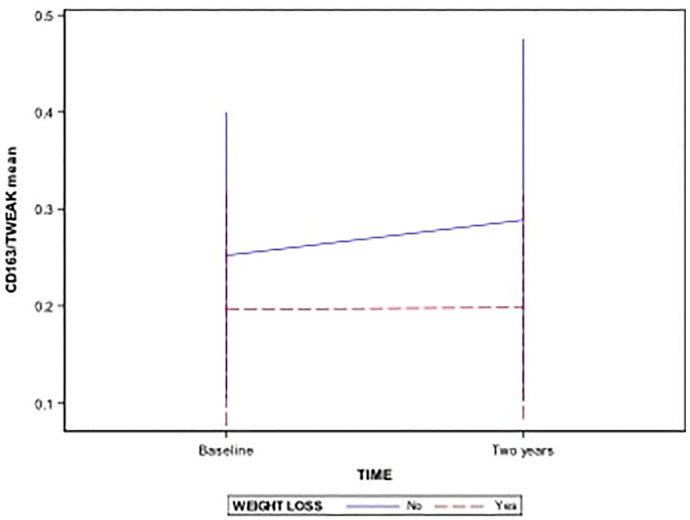

FIGURE 2 | CD163/TWEAK ratio was higher in children without weight loss, and it increased after 2 years in this group.

To further understand the relationship between TWEAK and CD163 and the rest of the variables, we carried out some bivariate correlation analysis with each of them. Age, sex, HOMA, and lipid profile were included as independent variables. No significant correlation among all these variables was found.

Forty-nine percent of the patients began puberty (assessed by Tanner stage) during the follow-up phase. After adjusting the regression model by Tanner, results were not modified.

DISCUSSION

TWEAK and CD163 have been linked to obesity and associated cardiovascular diseases in adults (15). However, there are still few data about these cytokines in children.

Our investigation did not find statistically significant differences in TWEAK or CD163/TWEAK ratio of children with obesity compared to controls. A possible explanation for this might be that this cytokine is involved in processes such as

TABLE 2 | Baseline and after 2-years follow up characteristics according to weight loss in children with obesity.

Sex	Without weight loss (n = 31)		With weight loss (n = 31)		
	16 girls/15 boys		16 girls/15 boys		
	Baseline	Two years	Baseline	Two years	p^a
Age (years)	7.6 ± 1.0	9.9 ± 1.1 [d]	8.4 ± 1.0[c]	10.4 ± 1.1 [d]	
BMI (Kg/m^2)	26.0 ± 2.7	28.8 ± 3.4 [d]	26.5 ± 2.8	25.9 ± 3.3 [d]	<0.001
z-BMI(SD)	4.68 ± 1.63	5.0 ± 1.7 [d]	4.6 ± 1.3	3.3 ± 1.3 [d]	<0.001
Waist (cm)	78.4 ± 7.1	87.5 ± 10.6[d]	81.5 ± 6.4 [c]	83.6 ± 8.2	0.002
SBP (mmHg)	111.8 ± 16.1	111 ± 8.8	107 ± 12.4	108 ± 11.4	0.773
DBP (mmHg)	64.1 ± 8.6	69.5 ± 9.6	61.9 ± 8.8	61.7 ± 9.1	0.252
Glucose (mg/dL)	83.9 ± 6.7	88.5 ± 7.2[d]	84.2 ± 7.57	88.4 ± 8.87 [d]	0.829
Glucose 120min	110 ± 21.5	112 ± 16.7	112 ± 16.7	110 ± 14.9	0.394
Insulin (μU/mL)[b]	9 (5-13.2)	15 (10-20.2) [d]	10 (5-13)	11 (8-14)	0.374
HOMA[b]	1.9 (1.0-2.7)	3.3 (2.0-4.6) [d]	1.9 (0.9-2.7)	2.5 (1.8-3.1) [d]	0.371
Cholesterol (mg/dL)	155 ± 32.1	160 ± 30.1	157 ± 25.5	161 ± 28.1	0.907
Triglycerides (mg/dL) [b]	62 (53-76)	77 (61-89) [d]	72 (44-15)	75 (58-108)	0.437
LDLc (mg/dL)	92.1 ± 29.1	92.2 ± 28.6	95.8 ± 19.6	94.8 ± 20.3	0.685
HDLc (mg/dL)	55.1 ± 12.4	53.2 ± 12.9	49.8 ± 13.5	50.7 ± 15.2	0.506
Uric acid (mg/dL)	4.26 ± 0.80	4.68 ± 0.87 [d]	3.83 ± 0.82 [c]	4.12 ± 0.75	0.932
TWEAK (pg/mL)	1081.5 ± 699.6	1017.7 ± 720.7	1792.3 ± 1637.0	1535.9 ± 1370.4	0.032
CD163 (ng/mL)	215.0 ± 61.36	208.3 ± 49.2	206.2 ± 36.1	193.4 ± 34.1	0.334
CD163/TWEAK ratio	0.252 ± 0.148	0.289 ± 0.187	0.197 ± 0.120	0.198 ± 0.115	0.032

Data are presented as mean ± SD.

[a]Mean of % of change between both groups. % of change= (end value - initial value)/initial value x 100.

[b]Median (interquartile range).

[c]Baseline comparison between those with and without weight loss (independent t-test) (p<0.05).

[d]Two year paired t-test (p<0.05).

BMI, body mass index; CD163, Cluster of differentiation 163; DBP, diastolic blood pressure; HDLc, high density lipoprotein cholesterol; HOMA-IR, homeostatic model insulin resistance index; LDLc, low density lipoprotein cholesterol; SBP, systolic blood pressure; TWEAK, Tumor Necrosis Factor Weak Inducer of Apoptosis.

cell proliferation and differentiation (36), present in childhood growth. The protective role of TWEAK in obesity is not demonstrated in prepubertal children, although some studies observed an anti-inflammatory role of this cytokine in obese adults (25, 37).

Regarding CD163, this cytokine is higher in adults with obesity, and known as a biomarker of insulin resistance (23). Carolan et al. observed elevated CD163 in children with obesity, suggesting that it could be a biomarker to prioritize lifestyle intervention in childhood (38). In our study, CD163 serum levels were also higher in children with obesity compared to lean.

The present study is the first to observe a favorable effect of a 2-year lifestyle intervention program in prepubertal children who achieved a significant weight loss compared with those who did not. We observed expected higher levels of TWEAK, lower CD163/TWEAK ratio, and a more pronounced decrease of CD163 in the group of patients who achieved weight loss.

This study is the first one to observe the evolution of these parameters in prepubertal children, supporting the hypotheses that these cytokines may play a role in childhood obesity. One study found similar results in adults with severe obesity after weight loss achieved by bariatric surgery (25). Kazankov et al. observed a decrease in CD163 after 10 weeks of a lifestyle intervention in children with a mean age of 12 years (39).

This research has some limitations. It has a weak external validity because all subjects were only from one center. However, inclusion criteria for subjects are enough to guarantee a strong internal validity in our results. Unfortunately we don't have estradiol, testosterone levels or TWEAK evolution of control group after two years of follow-up.

Our findings show that TWEAK and CD163 may be involved in the pathogenesis of obesity in prepubertal children. Childhood is a necessary stage for the prevention of consequences derived from obesity. This study opens an interesting line of research aimed at this objective.

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 CEIC from FUNDACIÓ INSTITUT D'INVESTIGACIÓ I INNOVACIÓ PARC TAULÍ (I3PT).

Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

AUTHOR CONTRIBUTIONS

AC, RC, and JG-C conceived the study. RC obtained informed consent and clinical variables. JV analyzed samples. MF and RE performed literature search. All authors were involved in writing the paper and had final approval of the submitted and published versions.

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REFERENCES

1. Bendor CD, Bardugo A, Pinhas-Hamiel O, Afek A, Twig G. Cardiovascular Morbidity, Diabetes and Cancer Risk Among Children and Adolescents With Severe Obesity. *Cardiovasc Diabetol* (2020) 19(1):79. doi: 10.1186/s12933-020-01052-1
2. Pasquali R, Casanueva F, Haluzik M, van Hulsteijn L, Ledoux S, Monteiro MP, et al. European Society of Endocrinology Clinical Practice Guideline: Endocrine Work-Up in Obesity. *Eur J Endocrinol* (2020) 182(1):G1–G32. doi: 10.1530/EJE-19-0893
3. Thota P, Perez-Lopez FR, Benites-Zapata VA, Pasupuleti V, Hernandez AV. Obesity-Related Insulin Resistance in Adolescents: A Systematic Review and Meta-Analysis of Observational Studies. *Gynecol Endocrinol* (2017) 33 (3):179–84. doi: 10.1080/09513590.2016.1273897
4. Weiss R, Dziura J, Burgert T, Tamborlane W, Taksali S, Yeckel C, et al. Obesity and the Metabolic Syndrome in Children and Adolescents. *N Engl J Med* (2004) 350(23):2362–74. doi: 10.1056/NEJMoa031049
5. Sinha R, Fisch G, Teague B, Tamborlane W, Banyas B, Allen K, et al. Prevalence of Impaired Glucose Tolerance Among Children and Adolescents With Marked Obesity. *N Engl J Med* (2002) 346(11):802–10. doi: 10.1056/NEJMoa012578
6. Wiegand S, Maikowski U, Blankenstein O, Biebermann H, Tarnow P, Grüters A. Type 2 Diabetes and Impaired Glucose Tolerance in European Children and Adolescents With Obesity – A Problem That Is No Longer Restricted to Minority Groups. *Eur J Endocrinol* (2004) 151(2):199–206. doi: 10.1530/eje.0.1510199
7. Lobstein T, Jackson-Leach R. Estimated Burden of Paediatric Obesity and Co-Morbidities in Europe. Part 2. Numbers of Children With Indicators of Obesity-Related Disease. *Int J Pediatr Obes* (2006) 1(1):33–41. doi: 10.1080/17477160600586689
8. Yusuf S, Hawken S, Ounpuu S, Dans T, Avezum A, Lanas F, et al. Effect of Potentially Modifiable Risk Factors Associated With Myocardial Infarction in 52 Countries (The INTERHEART Study): Case-Control Study. *Lancet* (2004) 364 (9438):937–52. doi: 10.1016/S0140-6736(04)17018-9
9. Karczewski J, Śledzińska E, Baturo A, Jończyk I, Maleszko A, Samborski P, et al. Obesity and Inflammation. *Eur Cytokine Netw* (2018) 29(3):83–94. doi: 10.1684/ecn.2018.0415
10. Rohm TV, Meier DT, Olefsky JM, Donath MY. Inflammation in Obesity, Diabetes, and Related Disorders. *Immunity* (2022) 55(1):31–55. doi: 10.1016/j.immuni.2021.12.013
11. Khafagy R, Dash S. Obesity and Cardiovascular Disease: The Emerging Role of Inflammation. *Front Cardiovasc Med* (2021) 8. doi: 10.3389/fcvm.2021.768119
12. Escoté X, Gómez-Zorita S, López-Yoldi M, Milton-Laskibar I, Fernández-Quintela A, Martínez JA, et al. Role of Omentin, Vaspin, Cardiotrophin-1, TWEAK and NOV/CCN3 in Obesity and Diabetes Development. *Int J Mol Sci* (2017) 18(8):1770. doi: 10.3390/ijms18081770
13. Blaszczak AM, Jalilvand A, Hsueh WA. Adipocytes, Innate Immunity and Obesity: A Mini-Review. *Front Immunol* (2021) 12. doi: 10.3389/fimmu.2021.650768
14. Barakat B, Almeida MEF. Biochemical and Immunological Changes in Obesity. *Arch Biochem Biophys* (2021) 708:108951. doi: 10.1016/j.abb.2021.108951
15. Vendrell J, Chacón MR. TWEAK: A New Player in Obesity and Diabetes. *Front Immunol* (2013) 4. doi: 10.3389/fimmu.2013.00488
16. Corripio R, Gónzalez-Clemente JM, Pérez-Sánchez J, Näf S, Gallart L, Nosàs R, et al. Weight Loss in Prepubertal Obese Children is Associated With a Decrease in Adipocyte Fatty-Acid-Binding Protein Without Changes in Lipocalin-2: A 2-Year Longitudinal Study. *Eur J Endocrinol* (2010) 163 (6):887–93. doi: 10.1530/EJE-10-0408
17. Yepes M, Winkles JA. Inhibition of TWEAK Activity as a New Treatment for Inflammatory and Degenerative Diseases. *Drug News Perspect* (2006) 19 (10):589–95. doi: 10.1358/dnp.2006.19.10.1068005
18. Tiller G, Fischer-Posovszky P, Laumen H, Finck A, Skurk T, Keuper M, et al. Effects of TWEAK (TNF Superfamily Member 12) on Differentiation, Metabolism, and Secretory Function of Human Primary Preadipocytes and Adipocytes. *Endocrinology* (2009) 150(12):5373–83. doi: 10.1210/en.2009-0488
19. Chacón MR, Richart C, Gómez JM, Megía A, Vilarrasa N, Fernández-Real JM, et al. Expression of TWEAK and its Receptor Fn14 in Human Subcutaneous Adipose Tissue. Relationship With Other Inflammatory Cytokines in Obesity. *Cytokine* (2006) 33(3):129–37. doi: 10.1016/j.cyto.2005.12.005
20. Díaz-López A, Chacón MR, Bulló M, Maymó-Masip E, Martínez-González MA, Estruch R, et al. Serum sTWEAK Concentrations and Risk of Developing Type 2 Diabetes in a High Cardiovascular Risk Population: A Nested Case-Control Study. *J Clin Endocrinol Metab* (2013) 98(8):3482–90. doi: 10.1210/jc.2013-1848
21. Maymó-Masip E, Fernández-Veledo S, Garcia España A, Vázquez-Carballo A, Tinahones FJ, García-Fuentes E, et al. The Rise of Soluble TWEAK Levels in Severely Obese Subjects After Bariatric Surgery may Affect Adipocyte-Cytokine Production Induced by Tnfα. *J Clin Endocrinol Metab* (2013) 98 (8):E1323–33. doi: 10.1210/jc.2012-4177
22. Etzerodt A, Moestrup SK. CD163 and Inflammation: Biological, Diagnostic, and Therapeutic Aspects. *Antioxid Redox Signal* (2013) 18(17):2352–63. doi: 10.1089/ars.2012.4834
23. Bover LC, Cardó-Vila M, Kuniyasu A, Sun J, Rangel R, Takeya M, et al. A Previously Unrecognized Protein-Protein Interaction Between TWEAK and CD163: Potential Biological Implications. *J Immunol* (2007) 178(12):8183–94. doi: 10.4049/jimmunol.178.12.8183
24. Fjeldborg K, Christiansen T, Bennetzen M, J Møller H, Pedersen SB, Richelsen B. The Macrophage-Specific Serum Marker, Soluble CD163, is Increased in Obesity and Reduced After Dietary-Induced Weight Loss. *Obes (Silver Spring)* (2013) 21(12):2437–43. doi: 10.1002/oby.20376
25. Maymó Masip E. *Estudio De La Citocina Tweak Y De Su Receptor Fn14 En El Contexto De La Obesidad. [Thesis]*. [Tarragona]: Universitat Rovira i Virgili (2015).
26. Hernández M, Castellet J, Narvaiza JL, Rincón JM, Ruiz I, Sánchez E, et al. *Curvas Y Tablas De Crecimiento. 1988 Ed*. I Desarrollo, editor. Madrid: Fundación F Orbegozo (1988). p. 33.
27. Marshall WA, Tanner JM. Variations in Pattern of Pubertal Changes in Girls. *Arch Dis Child.* (1969) 44(235):291–303. doi: 10.1136/adc.44.235.291
28. Marshall WA, Tanner JM. Variations in the Pattern of Pubertal Changes in Boys. *Arch Dis Child.* (1970) 45(239):13–23. doi: 10.1136/adc.45.239.13
29. Serra Majem L, Ribas Barba L, Aranceta Bartrina J, Pérez Rodrigo C, Saavedra Santana P, Peña Quintana L. [Childhood and Adolescent Obesity in Spain. *Result EnKid Study (1998-2000)] Med Clin (Barc)* (2003) 121(19):725–32. doi: 10.1016/s0025-7753(03)74077-9

30. Adolescents NHBPEPWGoHBPiCa. The Fourth Report on the Diagnosis, Evaluation, and Treatment of High Blood Pressure in Children and Adolescents. *Pediatrics* (2004) 114(2 Suppl 4th Report)::555–76.
31. Cole TJ. The LMS Method for Constructing Normalized Growth Standards. *Eur J Clin Nutr* (1990) 44(1):45–60.
32. 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
33. Association AD. Diagnosis and Classification of Diabetes Mellitus. *Diabetes Care* (2004) 27 Suppl 1:S5–S10.
34. 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
35. Reinehr T, Andler W. Changes in the Atherogenic Risk Factor Profile According to Degree of Weight Loss. *Arch Dis Child.* (2004) 89(5):419–22. doi: 10.1136/adc.2003.028803
36. Vendrell J, Maymó-Masip E, Tinahones F, García-España A, Megia A, Caubet E, et al. Tumor Necrosis-Like Weak Inducer of Apoptosis as a Proinflammatory Cytokine in Human Adipocyte Cells: Up-Regulation in Severe Obesity is Mediated by Inflammation But Not Hypoxia. *J Clin Endocrinol Metab* (2010) 95(6):2983–92. doi: 10.1210/jc.2009-2481
37. Vázquez Carballo A. *Papel Protector Del TWEAK Sobre La Resistencia a La Insulina Asociada a La Obesidad: Efecto Sobre La Migración Celular. [Thesis]*. Madrid: Universidad Complutense de Madrid (2014).
38. Carolan E, Hogan AE, Corrigan M, Gaotswe G, O'Connell J, Foley N, et al. The Impact of Childhood Obesity on Inflammation, Innate Immune Cell Frequency, and Metabolic microRNA Expression. *J Clin Endocrinol Metab* (2014) 99(3):E474–8. doi: 10.1210/jc.2013-3529
39. Kazankov K, Møller HJ, Lange A, Birkebaek NH, Holland-Fischer P, Solvig J, et al. The Macrophage Activation Marker Scd163 is Associated With Changes in NAFLD and Metabolic Profile During Lifestyle Intervention in Obese Children. *Pediatr Obes* (2015) 10(3):226–33. doi: 10.1111/ijpo.252

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The best tool for the assessment of developmental disorders in children with down syndrome: comparison of standard and specialized growth charts - cross sectional study

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Down Syndrome (DS) is a chromosomal abnormality associated with a spectrum of cognitive and physical disabilities. Children with DS are exposed to both lower and excess body weight and follow distinct growth-curve patterns that deviate significantly from those of children without chromosomal defects. Anthropometric parameters are assessed in the pediatric population with the use of growth charts. The study is based on data from 411 children and adults with DS from Poland. Detailed information concerning children and online survey results were also analyzed. Centiles and standard deviation scores (SDS) of obtained anthropometric parameters were aligned with the data using the LMS method. The study aims to identify which type of growth chart (standard vs specialized) is a leading tool for earlier detection of developmental disorders in DS. The results obtained in the two types of growth charts differed. The advantage of the specialized growth charts over the standard ones cannot be unequivocally determined. Only the combination of both tools allows to detect the development disorders early in the broadest possible way.

KEYWORDS

Down syndrome, growth charts, childhood malnutrition, childhood disability, obesity

Introduction

Down Syndrome (DS, also referred to as trisomy 21) is one of the most common chromosomal abnormalities among live-born neonates and is associated with a spectrum of cognitive and physical disabilities, such as congenital heart disease, hypothyroidism, gastrointestinal disorders, and obstructive sleep apnea (1). The occurrence of DS in 95%

of cases is related to meiotic non-disjunction causing trisomy of chromosome 21. The other types of trisomy are Robertsonian translocation and mosaic type (2). DS occurs in every 700-1000 live births (3) and its prevalence estimates between 6.1 to 13.1 per 10 000 people (4). It is predicted that 94.4% of children with DS born in 2000 will survive up to 2020, 90.8% up to 2030, and 76.3% up to 2050 (4). Early identification of developmental disorders can improve the quality of life in the future. To extend lifespan and improve the quality of life, the development of children with DS should be controlled with the use of optimal and appropriate tools.

The most often used parameters to evaluate the child's growth are anthropometric data such as body height and body weight. Body weight, as a single measure, is not sufficient to assess the nutrition of a given individual, therefore in this study nutritional status is analyzed with Body Mass Index (BMI) - a statistical index used to estimate the body fat content. It is worth remembering that this method is not ideal, but it may be the first step in assessing excess body fat. In the pediatric population, the proper assessment of BMI should be conducted on BMI growth charts.

During the first two years of life, children with DS are characterized by reduced body weight (5, 6), which may result from suction/swallowing disorders associated with muscle hypotonia and dysfunctions in the oral motor system (7). In underweight children the weight for the height it's a good measurement tool for controlling them. After the second year of life, the occurrence of overweight and obesity in children with DS is more frequent than in the general population (the prevalence of obesity at the level of 30-50%) (8–10), thereby increased BMI is common in DS (11).

Statural growth, as an indicator of development, often represents a child's health status. The growth retardation of children with DS commences prenatally (12). Morris et al. (6) demonstrated that for gestations up to 38 weeks the median birth weight of newborns with DS is similar to that of babies without DS, however, after 38 weeks their median birth weight rises slower than in unaffected babies. Other researchers also indicate a decreased birth weight in children with DS (13). After birth, the growth velocity is most reduced between 6 months and 3 years of age (14). Short stature is a phenotype of DS and can be influenced by genetic components and other factors, such as comorbidities. Styles et al. (15) compared developmental patterns in terms of body weight, height, and head circumference in children with DS compared to children without DS. Appreciable skewness was noted for body weight, which indicates the difference in the initial weight of children with DS compared to those without DS (15).

Growth charts constitute crucial tools used to assess the growth and nutritional status of children. Currently, various growth charts have been developed and adapted to racial and ethnic backgrounds or a given disease that may interfere with the proper development of a child. The most commonly used DS-specialized growth charts in the US are based on work done in 1988 (14). A great number of countries have constructed DS-specialized charts (5, 16, 17-19). Since these specialized growth charts were developed, concerns have been raised regarding their usefulness. Children with DS follow distinct growth-curve patterns that deviate significantly from those of children without chromosomal defects, therefore the use of specialized growth charts appears to be a superior method in development evaluation. The study aims to identify which type of growth chart (standard vs. specialized) is a leading tool for the earlier detection of developmental disorders in DS.

Material and methods

Design and participants

A cross-sectional study design was based on data from 411 people with DS: 386 (94%) children and 25 (6%) adults; 188 (46%) girls and 223 (54%) boys, aged 0.17 months – 36.72 years (median: 4.85) from Poland recruited from general pediatric practices and parents' interest groups. Inclusion criteria were patients with a diagnosis of DS. There were no exclusion criteria. The study was conducted in the years 2020-2021 in Wroclaw (Poland) as a part of the doctoral dissertation carried out at the Wroclaw Medical University. The ethical approval on the research protocol and consent form was obtained from the Bioethics Committee, Wroclaw Medical University (approval number KB 674/2020). The study was carried out in accordance with the Declaration of Helsinki. Administrative approvals were obtained from each institute to access the participants' data. Written informed consent was obtained from the parents of the participants prior to data collection and anthropometric measurements.

Data collection

The data were derived using two approaches between January 2020 - June 2021: by retrospectively examining medical records (20%) available at health clinics (additionally an online/telephone interview with parents or guardians was conducted to confirm the data and by obtaining the consent of data usage) and by actively recruiting participants (80%). Active recruitment and examination of retrospective medical records were conducted among children from all over Poland. All parents were invited to an online survey as an additional part of the study. After written informed consent was obtained, actively recruited children underwent an anthropometric examination in a pediatric clinic in Wroclaw (Poland) with collecting the anthropometric parameters such as body height and weight. For telephone calls, written consent was obtained by sending the consent form online. Then, the parent was asked to return the signed consent by post (original document) or online

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(scan of the document). Body Mass Index (BMI) was calculated using the formula: weight/height^2 (kg/m^2). Trained personnel (consisted of 3 people: two doctors (including authors) and a nurse) obtained measurements following standardized techniques (20), discussed prior to the research initiation. The design of the study (Collection Data part) is represented in Figure 1.

Specific data and online questionnaire

Specific data were collected from 200-300 participants depending on the parameter. Detailed information concerning parents of children with DS (Table 1), the perinatal period (Table 2), comorbidities (Table 3), and L-thyroxine treatment were obtained. In addition, an online 4-questions questionnaire concerning the topic of growth charts and their usage in medical offices was conducted (Table 4). Two hundred eighty-one parents (including those taking part in the main part of the study) answered the survey.

The ranges for assessing the parents' BMI: underweight [15.0, 18.5); healthy weight [18.5, 25.0); overweight [25.0, 30.0), and obesity ≥30The ranges for assessing the birth weight: high birth weight - greater than 4200g; normal weight - 2500g–4200g; low birth weight - less than 2500g; very low birth weight - less than 1500g; and extremely low birth weight less than 1000g.

TABLE 1 Parents' basic characteristics.

		Mother	Father
Age during pregnancy [years]	Average	32.41 ± 5.91	33.92 ± 3.43
	Median	32	33
	Min.	17	20
	Max.	54	58
Body weight [kg]	Average	68.40 ± 12.87	87.27 ± 13.75
	Median	65	86
	Min.	43	57
	Max.	110	130
Body height [cm]	Average	165.64 ± 5.73	179.59 ± 6.91
	Median	165	180
	Min.	147	159
	Max.	178	198
BMI [kg/m^2]	Average	24.89 ± 4.5	27.02 ± 3.77
	Median	24.17	26.87
	Min.	17.92	18.93
	Max.	39.40	37.55

Anthropometric measurements

Body weight: body weight (kg) was measured using the same electronic digital scales model (OMRON BF-515) with light clothes and barefoot for older children and without clothing or diapers for infants and toddlers (to the nearest 0.1kg for children >3 years and 0.05kg for children <3 years). Body height: length (to nearest 0.1 cm) was measured on an infant length board (SECA 234) for infants and toddlers unable to stand unsupported (in the supine position). For all others, height (to nearest 0.1 cm) was measured with a stadiometer (SECA 264). The trained personnel controlled the correct body posture of the child during the measurement: straight back, both feet on the ground, back of the body pressed against the wall. The same devices were used for all measurements, without changing the conditions. Birthdate information were extracted from the family or children's questionnaire. Body weight, body height,

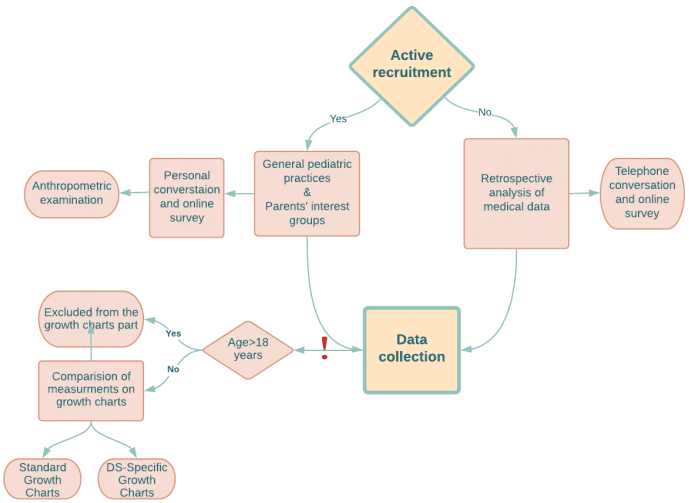

FIGURE 1
The design of the study (Data Collection).

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TABLE 2 Characteristics of postpartum parameters in the group of breastfed-children (85 children; 18 babies were born ≤36hbd).

	Min.	Max.	Median	Average
Birth weight [g]	1490	3970	2800	2840 ± 600
Birth length [cm]	41	59	51	51.29 ± 0.03
Week of pregnancy	31	42	38	37.55 ± 0.00
Apgar score 1'	0	10	9	8.25 ± 0.00

TABLE 3 The prevalence of selected common comorbidities in children with DS reported by parents.

Comorbidities	N	[%]
Hypothyroidism	142	52.98
Vision defects	112	41.79
Cardiac defects	64	23.88
Hearing problems	59	22.01
Immunodeficiency	34	12.69
Others	25	9.33
Lipid disorders	19	7.08
Autoimmune diseases	18	6.71
Malignancy	4	1.49
Hyperthyroidism	3	1.11
Hypertension	2	0.74

*N- the amount of participants (DS) with chosen comorbidity.

and BMI were expressed in the standard deviation score (SDS) value using the LMSgrowth Calculator (21)- a Microsoft Excel add-in. Centiles and SDS were fitted into the data using the LMS method (22). The LMS method summarizes the changing distribution of weight, height, head circumference, and BMI according to age by three curves representing the median (M), coefficient of variation (S), and skewness (L), the latter expressed as a Box-Cox power. The method assumes that the data in each age group can be rendered normally and distributed by applying a suitable power transformation (23). SDS indicates how many standard deviations an observation is above or below the mean independently of age and sex, which is a useful way of putting data from different sources onto the same scale (Equation (1)).

With the use of this statistics tool, it is possible to analyze the variability of the observed parameter over a certain period in a group of patients, especially those of developmental age.

$$x \quad SDS = \frac{x \quad value - x \quad value \quad for \quad 50th \quad centile}{\frac{1}{2}x(x \quad value \quad for \quad 50th \quad centile - x \quad value \quad for \quad 3rd \quad centile)} \quad (1)$$

$$x - height/weight/BMI \quad (1)$$

Growth charts and data analysis

To standardize the data, the British (24) growth charts included in the LMSgrowth Calculator were used for the calculations and taken as reference for the population of children without DS (population growth charts). DS-specific growth charts were used as the reference point for the population of children with DS (15). Three ranges were assumed (in percentile (PC)): <3rd, 3rd-97th, and> 97th, where 3rd-97th means a wide range of the norm (Table 6). However, it should be remembered that values >90th PC should be considered as overweight and that further calculations are related to obesity (>97th PC). Data from people over the age of eighteen (6%) were not considered in the comparison of growth charts. However, their parents were included in the online questionnaire part, mainly referring to their earlier experiences.

Statistical analysis

The data were processed using Statistica v. 13.3. The data were checked for normality using the Shapiro-Wilk test. Non-parametric statistical tests were applied. The Mann-Whitney U test was used for non-parametric data. Spearman's rank correlation (r) was performed to investigate the specific data (such as perinatal period, parents' physical status, L-thyroxine treatment, all affecting the current body height, body weight, and BMI of the child). The chi-squared test was used for data distribution. Descriptive statistics are presented as median/mean ± SD/ percentages. P-values <0.05 were considered significant developmental disorders in DS.

TABLE 4 Internet survey (questions with answers).

No.	Question	Answers	Results [%]
1	Have you ever heard about specialized growth charts for children with DS?	Yes	73.3
		No	26.7
2	Which type of charts is more often used by clinicians?	Standard	81.8
		Specific	13.2
3	Have any of the clinicians used specialized charts at least once?	Yes	28.2
		No	71.8
4	Are specialized growth charts important for you as a parent?	Yes	93.2
		No	6.8

Results

The study is based on data from 411 people with DS. Two hundred and fifty-five people from the study group have simple meiotic non-disjunction trisomy of chromosome 21; 12 are mosaic type; 10 have Robertsonian translocation. In the remaining cases of the questions about the type of mutation, the parents did not provide an answer, did not know the answer to the question, or never tested the child for a given mutation. The mean birth weight was 2898.02 ± 513g (median 2800 g); average birth length 0.52m ± 0.04m (median 0,51). The average age of delivery (weeks) was 37.7 ± 2.17 weeks.

Parents data

Table 1 presents the parents' basic characteristics. Fathers: One hundred thirty-six fathers (69%) have a BMI ≥25 (overweight) of which 31% corresponding to a BMI ≥30 (obesity). The average BMI value is27.02 ± 3.77. Mothers: eighty-four mothers (42%) have a BMI ≥25 (overweight) of which 26% corresponding to a BMI ≥30 (obesity). The average BMI value is 24.89 ± 4.58. There is no correlation between the current weight of the child and the parents' weight.

Child's birth weight

Data on birth weight were collected from 266 children. Fifty-nine babies were born ≤36hbb and were treated as premature babies. Among preterm babies, the mean birth weight was 2872.96 ± 484g (median 2800.00g; min.1490g; max. 3970). Among full-term babies (≥39hbd), the mean birth weight was 2967.97 ± 444g (median 2800g; minimum 2004g; maximum 4500g). Taking into account the entire group of 266 children, the mean birth weight was 2898.02 ± 513g (median 2800 g; min. 1490 g; max. 4500g). A positive correlation (low correlation) (rs = 0.152555) is found between the baby's birth weight and their current body weight.

Breastfeeding

Out of 109 children whose parents answered the question about breastfeeding, 85 (78%; girls: 36; 18 babies were born ≤ 36hbd) were breastfed. Max. duration of breastfeeding: 40 months; min. 0.5 months (median 9 months; average 11.06 ± 0.52months). Among those who were breastfed, 39 pregnancies were completed by natural childbirth. There were complications during childbirth in 15 cases. The median duration of pregnancy was 38 weeks (average 37.55 ± 0.00). The most common reasons for not breastfeeding were: lack of suckling reflex in the child and/or lack of lactation in the mother. There was no correlation between breastfeeding and body weight, body height, and BMI of the child. The median Apgar score among children fed breast milk after birth was 9 (average 8.25 ± 0.00). Table 3 presents basic characteristics of postnatal parameters in the group that was breastfed.

L-thyroxine therapy

There were 265 responses related to L-thyroxine therapy (L-thyroxine was taken by 169 children, 64%). There is no correlation between L-thyroxine intake and body weight or BMI. However, a statistically significant difference was identified for the body height growth charts readings. Smaller spread of values concerned children taking L-thyroxine. This means that when comparing the group of children taking the L-thyroxine and those not taking L-thyroxine, the children taking the medicine were within the wide normal range (3rd PC- 97th PC) more often.

Comorbidities

The data relating to selected comorbidities (Table 3) were collected from 198 participants. The three most common comorbidities in the study group are hypothyroidism affecting 52.98%; vision defects affecting 41.79%; and cardiac defects affecting 23.88%.

Growth charts

Table 5 shows percentiles and corresponding values of the SDS and their interpretation in relation to anthropometric parameters. A graphic representation of Table 6 is shown in Figures 2–4. Comparing the results obtained on two types of growth charts (standard vs. specialized): body weight - results outside the norm 30% vs. 11%; body height - 39% vs. 27%; and BMI - 28% vs. 21%. More results beyond the norm (under 3rd PC and above 97th PC) were obtained using standard growth charts.

Body weight

Using standard growth charts to assess body weight in a child with DS instead of the specific charts, 19 percentage points (p.p.) fewer children were considered in the range of normal body weight (70% vs. 89%, $p<0.02$), 21 p.p. more children had body weight deficiency (27% vs. 6%, $p<0.0001$); 2 p.p. fewer children had excess body weight (3% vs. 5%) (Figure 2). The statistically significant difference was observed in groups with weight deficiency and normal body weight.

TABLE 5 Percentiles and corresponding values of the standard deviation score and their interpretation in relation to anthropometric parameters.

Percentile (PC)	Standard deviation score (SDS)	Body weight	Body height	BMI
<3rd	<-1.88	Underweight	Growth deficiency	Underweight
3rd-10th	≥-1.88,<-1.66	Normal	Normal	Normal
10th-90th	≥-1.66, ≤1.66	Normal	Normal	Normal
90th-97th	>1.66, ≤1.88	Overweight	Normal	Overweight
>97th	>1.88	Obesity	High growth	Obesity

Body height

Using standard growth charts to assess body height in a child with DS instead of the specialized growth charts, 12 p.p. fewer (61% vs. 73%) children can be included in the normal range; 12 p.p. more (35% vs. 23%; p<0.005) children is above 97th PC (Figure 3). There is no difference in groups <3rd PC (4% vs. 4%) The statistically significant difference was observed in group with body height >97th PC.

Body mass index

Using standard growth charts to assess BMI in a child with DS instead of the specialized ones, 7 p.p. fewer children were considered in the range of normal BMI (72% vs. 79%), 5 p.p. more children had BMI <3rd PC (7% vs. 2%); and 2 p.p. more children had BMI >97th PC (21% vs. 79% p<0.005) (Figure 4). The statistically significant difference was observed only in group with BMI>97th PC.

Online survey

Two hundred and eighty-one people (parents of DS people) answered the questions from the Internet survey. The survey deals with the topic of growth charts and their application in clinical practice by doctors. The condition for completing the survey was answering all the questions. The results of the online survey are shown in Table 4.

Discussion

In this study we compared the assessment of children and adolescents' development in terms of body weight, body height, and BMI, using growth charts for the standard population and the sub-population of people with DS. The main objective was to identify which type of growth chart is the best tool for earlier detection of developmental disorders in DS. The results obtained on the two types of growth charts differed. Due to numerous comorbidities, disease phenotype, and social conditions, people with DS can be considered as a vulnerable population that requires systematic monitoring of their health status. Advances in medical care and increased access to knowledge have improved the health and well-being of individuals with DS. Currently, the illusion created in society is that the number of people with DS is decreasing. However, children with DS, one of the most common chromosomal abnormalities, will continue to be born, and with the current medical knowledge their lives may be longer, better and healthier. Monitoring of the child's health by doctors and parents should be performed with the use of appropriate assessment tools, of which the simplest and most common are growth charts. The challenge is to choose the kind of growth charts for the assessment of a given parameter so that the obtained results have a real impact on clinical decisions. The original hypothesis assumed that DS-specialized growth charts are chief tools in the comprehensive assessment (body weight, body height, BMI) of the developmental disorders in a child with DS. The obtained results, combined with clinical knowledge and experience, appear to contradict this hypothesis.

Body weight and BMI

Monitoring the child's development from the earliest stages of life with the use of optimal tools gives a chance to improve their quality of life in the future. Early health intervention can

TABLE 6 Distribution of data (body height, body weight, BMI) in both types of growth charts- standard and specialized growth charts.

		Body weight		Body height		BMI	
PC	Classification	StandardN (%)	DSN (%)	StandardN (%)	DSN (%)	StandardN (%)	DSN (%)
<3rd	Under	108 (27)	23 (5)	141 (35)	90 (23)	83 (21)	76 (19)
3rd-97th	Norm	280 (70)	350 (89)	246 (61)	288 (73)	290 (72)	306 (79)
>97th	Over	13 (3)	19 (6)	14 (4)	14 (4)	28 (7)	7 (2)

*PC- percentile, DS- Down Syndrome specialized growth charts, N- amount of individuals.

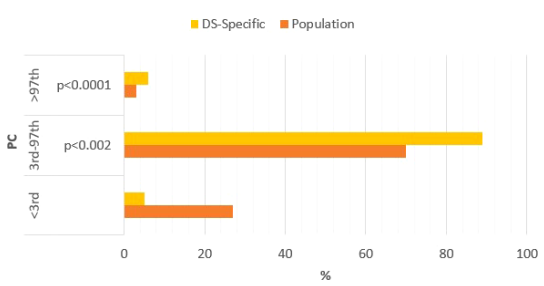

FIGURE 2
Percentage comparison of the number of individuals qualified for the given body weight categories on the standard growth charts and DS-specialized growth charts.

FIGURE 4
Percentage comparison of the number of individuals qualified for the given BMI categories on the standard growth charts and DS-specialized growth charts.

meaningfully affect adulthood. The problem of weight disorders among people with DS is very complex and challenging, concerning mainly the rapid transformation between undernutrition in the first period of life and excessive weight gain in later years. Therefore, depending on the age, this population is exposed to both deficiency and excess body weight and all the associated health consequences. As mentioned earlier, children with DS are characterized by a lower birth weight than children without chromosomal abnormalities (5, 6). However, in adolescence and adulthood, due to numerous comorbidities and the characteristics of the syndrome itself, people with DS are exposed to excessive body weight. Systematic nutritional evaluations since the day the baby is born throughout later years of life is essential. A higher obesity rate compared to the general population is observed among adolescents and adults with DS (25), therefore prevention and early treatment are principal aspects. DS has traditionally been considered as an "atheromafree" condition (26), however, the recent studies appear to contradict this thesis (27, 28). BMI, as based on body height and body weight, is a superior indicator of body nutrition. Both BMI and body weight are assessed using growth charts to detect body weight disorders. Hatch-Stein et al. (17) observed that for individuals with DS, the 85th percentile on standard growth charts is a better indicator of excess adiposity than the 85th percentile on the DS-specific BMI growth charts and claimed that standard charts should be the preferred method for early identification of obesity in children with DS. The results obtained in our study appear to confirm this. The percentage of out-of-normal results was analyzed, yielding a higher results percentage for body weight and BMI when using standard growth charts instead of the specialized ones. The use of specialized growth charts can deceptively reassure parents and lulls doctors into a false sense of a child's security. Since DS from adolescence is predisposed to excess body weight and has an increased risk of cardiovascular disease, the assessment of their BMI should be more rigorous, hence we recommend using standard growth charts. The percentage of children with DS with excess body weight is increased by genetic predisposition, low physical activity, and a high-calorie diet. Poor knowledge of healthy foods has been described in children and adolescents with DS (29). Increasing physical activity should be carried out wisely. Due to the cardiological burden in the group of people with DS, it may be necessary to assess the body's efficiency and consult a cardiologist before increasing physical activity. Introducing a balanced diet and regular meals should not be neglected. All the above-mentioned activities should be carried out with comprehensive care of i.e. the physician, nutritionists, psychologist, or trainer. A tremendous role in the whole process is played by parents and guardians. The whole family should be characterized by proper nutrition and activity patterns. Our study indicates that parents also face the problem of being overweight and obesity, which is particularly illustrated by the high BMI of the fathers (however, there is no correlation

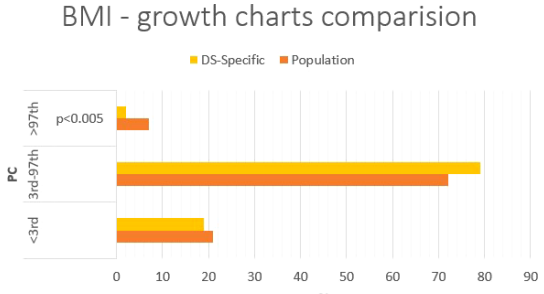

FIGURE 3
Percentage comparison of the number of individuals qualified for the given body height categories on the standard growth charts and DS-specialized growth charts.

between the current weight of the child and the parents' weight.). Fortunately, the awareness of parents who are trying to limit flour products and products with high sugar and saturated fat content in their children's diet is growing. Nevertheless, this challenging task is becoming very difficult to implement as the child grows older. However, it is not only the excess body weight that is a problem. In the study group, 1/5 of children were born with low birth weight. Low body weight in the first stage of a child's development may result from both maternal and child factors. On the side of maternal factors, there are, among others, problems with lactation, a lack of willingness to breastfeed, stress, and being overwhelmed by a new life situation related to childbirth (30–32). Breastfeeding in DS children is possible and preferred. A chance for its success can be obtained with the appropriate support of the family and competent health professionals. Frequent feeding problems in DS are the lack or very weak suckling reflex, prematurity, and defects in the digestive tract (33). Heart defects, which cause great and quick fatigue in newborns, contribute to the weak sucking reflex. Similar problems were confirmed in specific data collected in our study. Feeding difficulties, slow weight gain, and its deficiency may result in a slow and significantly impeded psychomotor development of a child. Introducing new products to a child's diet should take into account not only the type of product but also its texture (e.g., small pieces, mousses). Reduced feeding abilities with the increased risk of dysphagia and aspiration are predominant in the first years of life (32). If a child with DS is found to undergo weight loss and/or slow weight gain referring the child for a video-fluoroscopic swallow assessment and the diagnosis of contributing diseases (e.g., heart defects, celiac disease, gastrointestinal defects (Hirschsprung's disease, duodenal atresia, and others)) should be considered (32). Very important is the detection of disorders associated with both in deficiency and excess body weight and the approach of steps designed to fix these disorders.

Body height

People with DS are characterized by different patterns of growth compared to children without DS. The greatest impairment can be observed between 6 months and 3 years of age and in the puberty period, when they reach their final height (at age 15-16 years) (34). Furthermore, a shorter and earlier puberty spike related to the earlier achievement of the target height (girls: average of 9.5 years old, boys: average of 11 years old) is observed (35, 36). The assessment of body height using standard growth charts may be unfavorable as short stature is the phenotype of DS. However, the final growth of children with DS depends both on the characteristics of trisomy 21 and on the genetic potential transmitted by parents. The administration of growth hormone (GH) therapy in children with DS is associated with numerous controversies. Palloti et al. (37) observed in attempts of 3-year GH treatment an average improvement in the final height (boys by 5.16 cm; girls by 7.35cm). On the contrary, the other study shows that early treatment with GH does not affect the improvement of final height, but has a positive effect on psychomotor development and increases head circumference (38). In GH therapy a very problematic issue concerns the high risk of cancer, especially in the presence of the Philadelphia chromosome. Administration of GH could increase this risk of proliferative processes, consequently, the legitimacy of its administration should be considered. DS-specific growth charts used to assess a child's body heigh may provide valuable data to parents resulting in perceiving their child within the normal range. This can avoid unnecessary deliberations on the supply of growth hormone, the action of which, as presented above, may also have negative consequences. Additionally, there are risks of diseases that may result in delaying the rate of growth and achieving final growth, such as celiac disease or hypothyroidism. DS-specific charts were created based on data from people with Down syndrome, the presence of child measurements below the lower limit of normal is a signal for medical intervention, hence we recommend using population growth charts to assess the body height in DS.

L-thyroxine therapy

Many reports suggest that L-thyroxine therapy in the first years of life (also in children without diagnosed hypothyroidism) may result in better psychomotor development, support the child's physical therapy, and reduce thyroid immunization (39, 40). Thyroid diseases are one of the most common comorbidities among the population of people with DS (41). Most people at various stages of their lives are at risk of developing hypothyroidism. Among our study group, hypothyroidism was the most common accompanying disease in DS. In the study group, children taking L-thyroxine were within the normal range more often. On this basis, it can be concluded that the supply of L-thyroxine may support the proper growth of children with DS (40).

Comorbidities

Children with DS suffer from many comorbidities that may have nutritional implications and consequences. At the same time, thyroid disease is one of the most common accompanying diseases in DS. Obesity, as a civilization disease, very frequently affects people with DS. Obesity is also known to be associated with type 2 diabetes, cardiovascular disease, metabolic syndrome and some types of neoplastic processes. Complications of obesity

and related diseases can cause and intensify neurodegenerative processes (42).

Parental outcomes

It is well known that the older age of the mother is associated with Down's syndrome in children (43). According to the data (2011–2015), the average age at birth of the first child was 25.5 years for men and 23.1 years for women (44). In the study population the average age of mothers and fathers was: 32.41 ± 5.91 and 33.92 ± 3.43 years (Table 1). It is worth remembering that, paradoxically, taking into account the entire population, more children with DS are born to young mothers as they are of reproductive age. This can of course change over time as more and more women choose to have offspring later in life.

Online survey

The results of the online survey appear to be disturbing. So far, no growth charts adapted to the population of individuals with DS have been officially developed in Poland. Nevertheless, access to many other DS-specialized growth charts, including those developed by the CDC (5), is quick, simple and common. Despite that, as the survey results indicate, specific growth charts are not used in everyday medical practice in Poland although parents are aware of their existence. There can be many reasons for that state. First, many physicians are unaware of the existence of specialized growth charts. Secondly, in everyday practice, it is easier and more efficient to use standard growth charts. What's more, standard growth charts are available in every child's health booklet (parents should have it with them at every medical visit), which makes it effortless and faster to apply the child's data on the charts. Additionally, many doctors do not believe that it is necessary to use specialized charts to assess the development of children with DS. The usage of DS-specialized growth charts has certain consequences mainly regarding the previously mentioned issue of BMI assessment and the apparent dormancy of the parent's vigilance, while activities related to reducing the child's weight should already be taken. The optimal solution seems to be the assessment of a child on both types of growth charts to fully control their development at every stage. Fortunately, many parents are printing specialized growth charts and having them with them or pasting them into a child's health book. However, the results obtained only on specialized growth charts (body weight, BMI in some cases) may cause the parent to perceive the body weight as healthy even though it may require early intervention. Nevertheless, DS-specialized growth charts should be implemented into pediatric departments as an important and additional tool to properly assess the development of children and adolescents with DS. When extending the scope of research on this topic it would be worth expanding the research group to include people from the medical community.

Advantages and limitations of the current study

This study should be interpreted in light of its limitations. First, the data we obtained from medical records (20%) could be the results of measurements performed without the use of standardized techniques, and this could lead to measurement errors. Part of the study population came from a pediatric endocrinological health clinic, which means that they are treated for endocrine reasons. At the same time, most children with Down's syndrome are burdened with comorbidities, so it would be difficult to single out a group for example without heart defects and thyroid problems. A sample of children attending a medical office was used, not a random sample from the target population. Our research, in order to be more valuable, could also be expended by measurement of the head circumference. As the standard growth charts, the 1990 British growth charts were used. This choice was dictated by the high detail of the mentioned growth charts and their coherence with the LMSgrowth Microsoft Excel add-in used. This study also has some strengths. First, the sample size is large. Second, the data for youth aged 4 months–36 years covers almost the full range of development. Thanks to medical data collected from parental groups our research group includes children from all over Poland, not just from one region. The gathered group of over 400 children covers the spectrum of children with many DS-typical diseases.

Conclusions

There is no single comprehensive tool for the assessment of the developmental disorders in DS. The differences between the results obtained using standard growth charts and specialized ones were identified, however, they are ambiguous in the clinical meaning. It is both type of growth charts that are capable of detecting development disorders early in the broadest possible way. The findings of our study can be valuable for healthcare professionals, parents, and guardians in drawing attention to the need for complex monitoring of developmental disorders in people with DS. Accurate assessment of anthropometric indicators of the development may enable to improve the quality of life and to extend the period of a healthy lifespan.

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 Bioethics Committee, Wroclaw Medical University (approval number KB 674/2020). Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

Author contributions

MH: term, Conceptualization, Investigation, Resources, Data Curation, Writing - Original Draft, Project administration, Visualization. HM: Validation, Formal analysis, Data Curation, EB: Methodology, Resources, Writing - Review and Editing, Supervision, Project administration. All authors contributed to the article and approved the submitted version.

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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. Roizen NJ, Magyar CI, Kuschner, Sulkes SB, Druschel C, Wijngaarden E, et al. A community cross-sectional survey of medical problems in 440 children with down syndrome in new york state. *J Pediatr* (2014) 164(4):871–5. doi: 10.1016/j.jpeds.2013.11.032

2. Kuzubski W, Liberski P. Neurologia podręcznik dla studentów medycyny. (2014). Wydanie II; Warszawa, PZWL, 208–10.

3. Presson AP, Partyka G, Jensen KM, Devine OJ, Rasmussen SA, McCabe LL, et al. Current estimate of down syndrome population prevalence in the united states. *J Pediatr* (2013) 163(4):1163–8. doi: 10.1016/j.jpeds.2013.06.013

4. de Graaf G, Vis JC, Haveman M, Hochstenbach R, Engelen J, Gerssen-Schoorl K, et al. Assessment of prevalence of persons with down syndrome: A theory-based demographic model. *J Appl Res Intellect Disabil* (2011) 24(3):247–62. doi: 10.1111/j.1468-3148.2010.00593

5. Zemel BS, Pipan M, Stallings, et al. Growth charts for children with down syndrome in the united states. *Pediatrics* (2015) 136(5):e1204–11. doi: 10.1542/peds.2015-1652

6. Morris JK, Cole TJ, Springett, Dennis J. Down syndrome birth weight in England and Wales: Implications for clinical practice. *Am J Med Genet Part A* (2015) 167A(12):3070–5. doi: 10.1002/ajmg.a.37366

7. Weisz B, David AL, Chitty L, Peebles D, Pandya P, Patel P, et al. Association of isolated short femur in the mid-trimester fetus with perinatal outcome. *Ultrasound Obstetric Gynecol Off J Int Soc Ultrasound Obstetric Gynecol* (2008) 31(5):512–6. doi: 10.1002/uog.5349

8. Hermon C, Alberman E, Beral V, Swerdlow AJ. Mortality and cancer incidence in persons with down's syndrome, their parents and siblings. *Ann Hum Genet* (2001) 65(Pt 2):167–76. doi: 10.1017/S0003480001008508

9. Day SM, Strauss DJ, Shavelle RM, Reynolds RJ. Mortality and causes of death in persons with down syndrome in California. *Dev Med Child Neurol* (2005) 47 (3):171–6. doi: 10.1017/s0012162205000319

10. van Gameren-Oosterom HB, van Dommelen P, Schönbeck Y, Oudesluys-Murphy AM, van Wouwe JP, Buitendijk SE. Prevalence of overweight in Dutch children with down syndrome. *Pediatrics* (2012) 130(6):e1520–6. doi: 10.1542/peds.2012-0886

11. Hsieh K, Rimmer JH, Heller T. Obesity and associated factors in adults with intellectual disability. *J Intellect Disabil Res JIDR* (2014) 58(9):851–63. doi: 10.1111/jir.12100

12. Kurjak A, Kirkinen P. Ultrasonic growth pattern of fetuses with chromosomal aberrations. *Acta Obstetric Gynecol Scandinavic* (1982) 61(3):223–5. doi: 10.3109/00016348209156561

13. Ergaz-Shaltiel Z, Engel O, Erlichman I, Naveh Y, Schimmel MS, Tenenbaum A. Neonatal characteristics and perinatal complications in neonates with down syndrome. *Am J Med Genet* (2017) 173(5):1279–86. doi: 10.1002/ajmg.a.38165. Part A.

14. Cronk C, Crocker AC, Pueschel SM, Shea AM, Zackai E, Pickens G, et al. Growth charts for children with down syndrome: 1 month to 18 years of age. *Pediatrics* (1998) 81(1):102–10. doi: 10.1046/j.1365-2788.1996.800800.x

15. Styles ME, Cole TJ, Dennis J, Preece M. A. new cross sectional stature, weight, and head circumference references for down's syndrome in the UK and republic of Ireland. *Arch Dis Childhood* (2002) 87(2):104–8. doi: 10.1136/adc.87.2.104

16. Cremers MJ, van der Tweel I, Boersma B, Wit JM, Zonderland M. Growth curves of Dutch children with down's syndrome. *J Intellect Disabil Res JIDR* (1996) 40(Pt 5):412–20. doi: 10.1046/j.1365-2788.1996.800800.x

17. Hatch-Stein JA, Zemel BS, Prasad D, Kalkwarf HJ, Pipan M, Magge SN, et al. Body composition and BMI growth charts in children with down syndrome. *Pediatrics* (2016) 138(4):e20160541. doi: 10.1542/peds.2016-0541

18. Bertapelli F, Agiovlasitis S, Machado MR, do Val Roso R, Guerra-Junior G. Growth charts for Brazilian children with down syndrome: Birth to 20 years of age. *J Epidemiol* (2017) 27(6):265–73. doi: 10.1016/j.je.2016.06.009

19. Aburawi EH, Nagelkerke N, Deeb A, Abdulla S, Abdulrazzaq YM. National growth charts for united Arab Emirates children with down syndrome from birth to 15 years of age. *J Epidemiol* (2015) 25(1):20–9. doi: 10.2188/jea.JE20130081

20. Lohman T, Roche AF, Martorell R. *Anthropometric standardization reference manual*. Champaign, IL: Human Kinetics Books (2008).

21. Pan H, Cole TJ. *LMSgrowth, a Microsoft excel add-in to access growth references based on the l method, version 2.69.* (2010). Available at: http://www.healthforallchildren.com/shop-base/software/lmsgrowth

22. Cole TJ. The LMS method for constructing normalized growth standards. *Eur J Clin Nutr* (1990) 44(1):45–60.

23. Box GEP, Cox DR. An analysis of transformation. *J R Stat Soc* (1964) 26:211–52. Series B (Methodological).

24. Cole TJ. Growth monitoring with the British 1990 growth reference. *Arch Dis Childhood* (1997) 76(1):47–9. doi: 10.1136/adc.76.1.47

25. Seron BB, Silva RA, Greguol M. Effects of two programs of exercise on body composition of adolescents with down syndrome. *Rev Paulist Pediatr Orgao Oficial Da Sociedade Pedia Sao Paulo* (2014) 32(1):92–8. doi: 10.1590/s0103-05822014000100015

26. Murdoch JC, Rodger JC, Rao SS, Fletcher CD, Dunnigan MG. Down's syndrome: an atheroma-free model? *Br Med J* (1997) 2(6081):226–8. doi: 10.1136/bmj.2.6081.226

27. Raina T, McGrath E, Gunn J. Myocardial infarction in a patient with down syndrome: a case report and review of the literature. *Clin Cardiol* (2011) 34(2):87–9. doi: 10.1002/clc.20864

28. Hill DA, Gridley G, Cnattingius S, Mellemkjaer L, Linet M, Adami HO, et al. Mortality and cancer incidence among individuals with down syndrome. *Arch Internal Med* (2003) 163(6):705–11. doi: 10.1001/archinte.163.6.705

29. Jobling A, Cuskelly M. Young people with down syndrome: a preliminary investigation of health knowledge and associated behaviours. *J Intellect Dev Disabil* (2006) 31(4):210–8. doi: 10.1080/13668250600999186

30. Pisacane A, Toscano E, Pirri I, Continisio P, Andria G, Zoli B, et al. Down syndrome and breastfeeding. *Acta Paedia (Oslo Norway 1992)* (2003) 92(12):1479–81. doi: 10.1080/08035250310007024

31. Flores-Lujano J, Perez-Saldivar ML, Fuentes-Pananá, Gorodezky C, Bernaldez-Rios R, Del Campo-Martinez MA, et al. Breastfeeding and early infection in the aetiology of childhood leukaemia in down syndrome. *Br J Cancer* (2009) 101(5):860–4. doi: 10.1038/sj.bjc.6605244

32. Nordstrøm M, Retterstøl K, Hope S, Kolset SO. Nutritional challenges in children and adolescents with down syndrome. *Lancet Child Adolesc Health 2020* (2020) 4(6):455–64. doi: 10.1016/S2352-4642(19)30400-6

33. Frazier JB. & Friedman, b. swallow function in children with down syndrome: a retrospective study. *Dev Med Child Neurol* (1996) 38(8):695–703. doi: 10.1111/j.1469-8749.1996.tb12139.x

34. Myrelid A, Gustafsson J, Ollars B, Annerén G. Growth charts for down's syndrome from birth to 18 years of age. *Arch Dis Childhood* (2002) 87(2):97–103. doi: 10.1136/adc.87.2.97

35. Kimura J, Tachibana K, Imaizumi K, Kurosawa K, Kuroki Y. Longitudinal growth and height velocity of Japanese children with down's syndrome. *Acta Paedia (Oslo Norway 1992)* (2003) 92(9):1039–42. doi: 10.1080/08035250310004306

36. Van Gameren-Oosterom HB, Van Dommelen P, Oudesluys-Murphy AM, Buitendijk SE, Van Buuren S, Van Wouwe JP. Healthy growth in children with down syndrome. *PLoS One* (2021) 7(2):e31079. doi: 10.1371/journal.pone.0031079

37. Pallotti S, Giuliano S, Giambi C. Disturbo della crescita nella sindrome de down: terapia con ormone della crescita [Growth disorders in down's syndrome: growth hormone treatment]. *Minerv Endocrinol* (2002) 27(2):59–64.

38. Myrelid Å., Bergman S, Elfvik Strömberg M, Jonsson B, Nyberg F, Gustafsson J, et al. Late effects of early growth hormone treatment in down syndrome. *Acta Paedia (Oslo Norway 1992)* (2010) 99(5):763–9. doi: 10.1111/j.1651-2227.2009.01679.x

39. Zwaveling-Soonawala N, Witteveen ME, Marchal JP, Klouwer F, Ikelaar NA, Smets AMJB, et al. Early thyroxine treatment in Down syndrome and thyroid function later in life. *Eur J Endocrinol* (2017) 176(5):505–13. doi: 10.1530/EJE-16-0858

40. Kowalczyk K, Pukajło K, Malczewska A, Król-Chwastek A, Barg E. L-thyroxine therapy and growth processes in children with down syndrome. *Adv Clin Exp Med Off Organ Wroclaw Med Univ* (2013) 22(1):85–92.

41. Pierce MJ, LaFranchi SH, Pinter JD. Characterization of thyroid abnormalities in a Large cohort of children with down Syndrome. *Hormone Res Paediatric* (2017) 87(3):170–8. doi: 10.1159/000457952

42. Moreau M, Benhaddou S, Dard R, Tolu S, Hamzé R, Vialard F, et al. Metabolic diseases and down syndrome: How are they linked together? *Biomedicines* (2021) 9(2):221. doi: 10.3390/biomedicines9020221

43. Newberger DS. Down syndrome: prenatal risk assessment and diagnosis. *Am Fam Phys* (2000) 62:825–38.

44. Martinez GM, Daniels K, Febo-Vazquez I. Fertility of men and women aged 15-44 in the united states: national survey of family growth, 2011-2015. *Natl Health Stat Rep* (2018) 113:1–17.

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Association of gestational hypertension and preeclampsia with offspring adiposity: A systematic review and meta-analysis

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Background: The association of gestational hypertension (GH) and preeclampsia (PE) with offspring adiposity outcomes had controversial results in different studies.

Objective: We conducted a systematic review and meta-analysis to evaluate the relationship between maternal GH/PE and offspring adiposity outcomes.

Search strategy: Studies were identified in PubMed, Embase, and Cochrane databases, with keywords including "gestational hypertension", "preeclampsia", "offspring", "weight", "cohort study", etc., without year restriction. This study was registered with PROSPERO, CRD42022292084.

Selection criteria: We set the selection criteria for six aspects: population, outcome, time frame, study design, and availability. For the studies included in the meta-analysis, we required the potential confounders in these studies have been adjusted.

Data collection and analysis: Two reviewers independently evaluated the data from the included studies. The meta-analyses included mean differences, regression coefficients, and corresponding 95% confidence intervals. Results were performed using RevMan software (version 5.4; Cochrane Collaboration). Heterogeneity among the included studies was assessed using the I^2 statistic.

Main results: A total of 16 studies were included in our review, 15 of which were evaluated as high quality. In all offspring, during the early life (28 days-36 months), GH/PE exposure was found to be not or inversely associated with offspring obesity, then become positively associated at larger ages (3-19 years old). In offspring with adverse birth outcomes, the maternal GH/PE-exposed group had a lower weight in the short term (28 days to 18 months), but there

was a trend of rapid weight gain as they grew older, compared with the non-exposed group. The meta-analysis showed that the BMI of the female offspring in the maternal PE-exposed group was significantly higher than that of the non-exposed offspring (MD=1.04, 95% CI: 0.67~1.42, $P < 0.05$).

Conclusions: The systematic review suggested that maternal exposure to *de novo* hypertension disorders of pregnancy (HDP) was associated with obesity in offspring, extending from early childhood to adolescence. The meta-analysis showed that PE was associated with higher BMI in female offspring. More studies are needed to conduct stratified analyses by PE/GH, the severity of HDP, or gender.

Systematic review registration: PROSPERO, identifier CRD42022292084.

KEYWORDS

gestational hypertension, preeclampsia, maternal, offspring, adiposity

Introduction

Obesity in children and adolescents has become one of the most vital public health problems due to its high prevalence and its adverse outcomes. According to the data from World Health Organization, there were only less than 1% of children aged 5-19 with obesity in 1975, but 38.2 million children under 5 and over 340 million children and adolescents aged 5-19 were with overweight or obesity in 2019 (1). Childhood obesity would have both short- and long-term health effects, such as increased asthma risk, musculoskeletal problems, and cardiovascular disease in adulthood (2–4). Meanwhile, studies have shown that childhood obesity could persist into adulthood, which indicated the importance of preventing obesity at an early stage (5).

The influencing factors of childhood obesity, like nutritional factors, physical activity, sleep status, and family factors, mainly focused on the different stages of life after childbirth. However, more evidence showed that the origins of childhood obesity can be as early as maternal pregnancy. According to "Developmental Origins of Health and Disease Hypothesis" (DOHaD), a lack of nutrients in the uterus would cause epigenetic changes and evolutionary adaption of the offspring. After childbirth, the catch-up growth of offspring would have short-term survival benefits but permanent alterations in the body's metabolism (6–9). Both animal experiments and epidemiological studies have shown a significantly increased prevalence of obesity in the group exposed to adverse pregnancy factors than in the controls (10–12).

Hypertension disorders of pregnancy (HDP) may be one of the adverse pregnancy factors mentioned above. HDP complicates about 5–10% of pregnancies, contributing to severe maternal and fetal health outcomes (13). It can be divided into four categories: gestational hypertension (GH), preeclampsia (PE)/eclampsia, chronic hypertension (CH), and CH with superimposed PE (14). Different from pre-existing hypertension with or without PE, both GH and PE are characterized by *de novo* HDP. GH occurs at any time after 20 weeks of pregnancy, and if proteinuria (excretion of protein over 300 mg per day) occurs the disease was thought to develop into PE. In some cases, PE could occur without a prior diagnosis of GH. In 2017, a review of epidemiological studies reported the global prevalence of GH and PE were 1.8–4.4% and 0.2–9.2%, respectively (15).

Some studies have investigated the relationship between maternal HDP and offspring adiposity outcomes (16–18). However, since CH and *de novo* HDP may have different mechanisms, combining all types of HDP would cause bias (19, 20). Furthermore, most previous studies focused on adult offspring (21), and there were a few studies focused on obesity of offspring in children and adolescents, in which the findings were inconsistent (21, 22). Only two systematic reviews in the past have discussed the relationship between HDP and offspring adiposity outcomes, but there were limitations such as unclear classification of HDP, and less specific inclusion criteria (23, 24). In light of these inconsistent results and the limitations of previous systematic reviews, it is necessary to conduct a systematic review focusing on *de novo* HDP and offspring adiposity outcomes.

We conducted the systematic review aiming to systematically review and quantitatively analyze the current evidence on the association between *de novo* HDP and offspring adiposity outcomes.

Methods

We carried out this systematic review based on the recommendations of the Preferred Reporting Items for Systematic Review and Meta-Analysis (PRISMA) guidelines. The protocol of this review was registered in the PROSPERO database (registration number: CRD42022292084).

Data sources and search strategy

We performed a literature search within the PubMed, Embase, and Cochrane databases without year restriction. The search strategy was defined according to the "PICOS" principle: "P"-offspring, "I"-exposed to GH/PE, "C"-non-exposed to GH/PE, "O"-adiposity outcomes,"S"-cohort study. Thus, the searches were conducted using Medical Subject Heading terms and the keywords including ("Pregnancy" OR "Maternal") AND ("Pre-Eclampsia" OR "Preeclampsia") AND ("Hypertension, Pregnancy-Induced" OR "Gestational Hypertension" OR "Pregnancy Induced Hypertension") AND ("Infant, Newborn" OR "Adolescent" OR "Pediatrics" OR "offspring") AND ("Body Weight" OR "adiposity" OR "adipose" OR "body mass index" OR "obesity" OR "waist circumference") AND ("cohort" OR "retrospective" OR "prospective" OR "follow up"). The detailed search strategy is found in the Supplementary Table 1. Then we used the snowball method to manually scrutinize the reference lists of the identified articles, looking for the relevant studies.

Selection criteria

Two reviewers (SY and JL) independently evaluated the studies according to the following criteria (Table 1).

Studies were included in the meta-analysis if they met the following criteria:

(1) There were available data: mean differences (MD), regression coefficient (β), and their corresponding 95% confidence intervals (CI).

(2) Potential confounders were adjusted.

Data extraction and quality assessment

Two reviewers (SY and JL) independently screened the titles and abstracts according to the inclusion and exclusion criteria. For studies potentially eligible for the requirements, full texts were obtained to screen. Then the data from all potentially eligible studies were extracted with a predefined data extraction form (an Excel sheet). The following information was extracted: name of the first author, year of publication, study location, ethnicity, the sample size of the control group (without PE/GH), the sample size of the PE/GH group, the definition of PE/GH, follow-up time, evaluation of the offspring adiposity outcomes, and adjusted potential confounders. Any disagreements would be resolved by discussion. And when consensus could not be reached, disagreements were settled by a third reviewer (ZL). Quality evaluation was assessed using the Newcastle–Ottawa Quality Assessment Scale (NOS). A maximum of nine stars can be given to one study. Two reviewers independently carried out the assessment. The study with a NOS score ≥7 was considered high quality. When 5 ≤ NOS score ≤ 7, the study was considered as moderate quality. And when a NOS score was below 5, the study quality was considered as low.

Statistical analysis

In the meta-analysis, the study included the results of obesity-related indicators with adjustment for confounding factors, including MD, β and corresponding 95% CI. The analysis was performed using RevMan software (version 5.4; Cochrane Collaboration). Heterogeneity among the included studies was assessed using the I^2 statistic. When I^2 >50%, there was heterogeneity among studies and a random-effect model was used; When I^2 ≤50%, the heterogeneity between studies was low and a fixed-effect model was used.

Results

Search results

The flow chart of the literature search and selection of studies was shown in Figure 1. A total of 6,212 titles were retrieved from the search, only 14 met all selection criteria and were included in this systematic review. Two additional articles were incorporated after using the snowball method to search through the reference lists of identified articles. Finally, a total of 16 articles were included in our review. Important characteristics (author/year, sample size and grouping, observes age, a summary of results, control of confounding factors, and NOS) of these studies were shown in Table 2.

Qualitative description

Among the 16 included studies, 15 were evaluated as high-quality using NOS criteria, except for one study with moderate

TABLE 1 Selection criteria for studies included in the review.

	Inclusion Criteria	Exclusion Criteria
Population	●Mothers with antenatal exposure to PE or GH	●Including other pregnancy-induced hypertension without differentiation (e.g. eclampsia)
Outcome	●Mainly investigating the association between PE/GH and indicators relevant to offspring adiposity outcomes (including body weight, BMI, BMI Z score, etc.) ●Offspring aged 0-18 (We also included a study in which the offspring was aged 13-19 because most populations meet the requirement.)	●Focusing on other indicators of offspring adiposity outcomes not related to adiposity. ●Offspring over 18 years of age
Time frame	●Any time frame	
Study design	●Human studies ●Cohort studies (prospective cohort study, retrospective cohort study, birth cohort, follow-up of the nested case-control study) ●Having a control group (women with normotensive pregnancy)	●Randomized Controlled Trial ●Review ●Editorial ●Case report ●Commentary article ●Animal studies
Availability	●Able to find the full-text article ●Published in English	●Non-English language

Abbreviations: PE, preeclampsia; GH, gestational hypertension; BMI, body weight index.

quality published in 1989 (25). The detailed NOS scores were found in the Supplementary Table 2. Among 15 studies with high quality, 10 studies merely included the population exposed to PE, and 5 studies included people exposed to GH or PE. According to the anthropometry age of offspring, there were 4 studies in the age group of 0-5 years, one study in the age group of 6-9 years, and 8 studies in the age group of 10-19 years. In addition, one study conducted measurements in children aged 3-

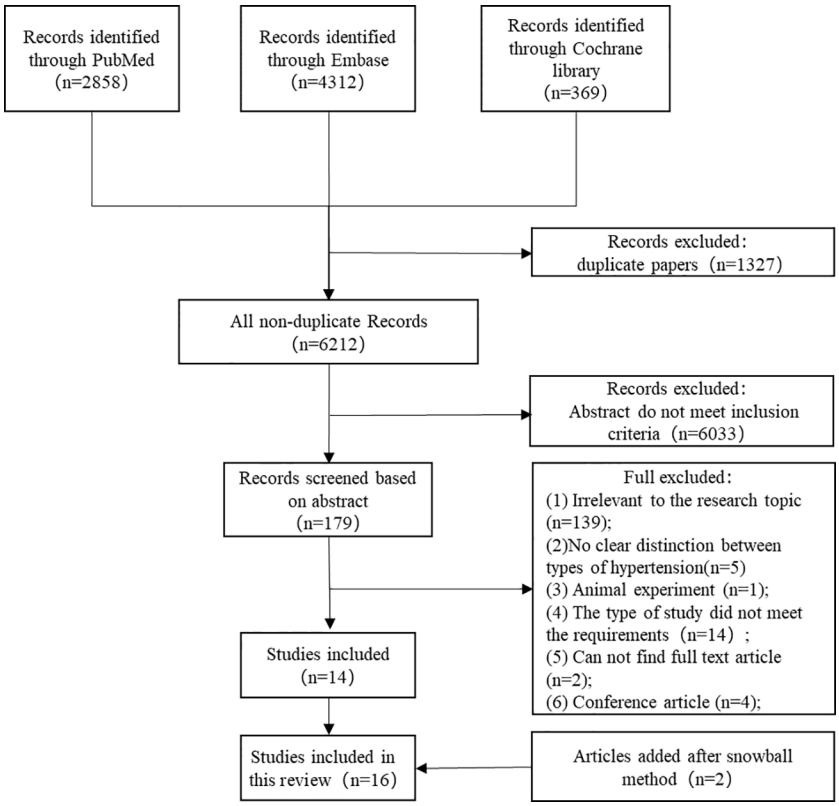

FIGURE 1
Flow chart of the literature search and selection of studies.

TABLE 2 Important characteristics of the included studies.

No	Author/ year	Sample size and grouping	Observed age	Summary of results	Control of confounding factors	NOS
1	Baulon E et al., 2005	Control 10069; GH 782; PE 365; Severe PE/eclampsia 105;	28 and 42 days after delivery	28 days Infant Growth Percentage, β (SE) [[(infant weight - birth weight)/birth weight] × 100%] GH/non-IUGR -4.4 (18.6); PE/non-IUGR 38.0 (25.8); 42 days Infant Growth Percentage, β (SE) GH/non-IUGR -12.8 (22.8); PE/non-IUGR 55.4 (33.5);	Maternal age, body mass index, diabetes, infant's sex, maternal anemia, gestational age	7
2	Megan L Gow et al. 2021	Control 298; PE 84;	0-6 months	6month Weight (kg), mean (SD) Non-PE 7.93 (0.96); PE7.61 (0.99)* ; BMI (kg/m^2), mean (SD) Non-PE 16.89 (1.50); PE 16.64 (1.46); Weight Z score, mean (SD) Non-PE 0.05 (0.94);PE -0.21 (0.95)*; Weight gain(kg), mean (SD) Non-PE 4.56 (0.94);PE 4.86 (0.99)*;	Crude results	9
3	Silveira RC et al., 2007	Control 40; PE 46;	12 months and 18 months corrected ages	At 12 months corrected age Weight (g), mean ± SD Non-PE 9223 ± 231; PE 8610 ± 120*; Weight/age Z, mean ± SD Non-PE −0.71 ± 1.36; PE−1.36 ± 0.88*; Weight/length Z, mean (SD) non-PE −0.37 ± 1.34; PE −1.08 ± 1.22*; At 18 months corrected age Weight (g), mean ± SD Non-PE 12510 ± 139; PE 9785 ± 142*; Weight/age Z, mean ± SD Non-PE −0.88 ± 1.17; PE −1.48 ± 0.97; Weight/length Z, mean (SD) non-PE −0.66 ± 1.24; PE−1.37 ± 1.09**;	Crude results	9
4	Jiang W et al., 2021	Normal 31171; GH408;	1, 3, 6, 8, 12, 18, 24, 30, 36 months	Weight, β(SE) GH - 0.15 (0.05)**; Weight-for-age Z score, β(SE) GH -0.05 (0.02)*;	Offspring sex, maternal age at delivery, maternal pre-pregnancy body mass index, parity, educational level, birthweight	9
5	Randhir K et al., 2020	Control 470; PE 681;	3-7 years old	Weight Z score, β (95%CI) PE 0.21 (0.059, 0.48)*; BMI Z score, β (95%CI) PE: 0.13 (-0.09, 0.36);	Birth weight, gestational age, maternal BMI, maternal height, and SLI score	9
6	Palti H et al. 1989	Control 94; PE94;	6 years old	Weight (kg) (male), mean(SD) Non-PE19.2 ± 3.6; PE 20.1 ± 3.4; Weight(kg) (female), mean(SD) Non-PE 18.8 ± 3.0; PE 19.1 ± 3.5;	Crude results	6
7	Huang Y et al., 2020	Control 332; PE 24;	18-72 months	Hierarchical Linear Modeling: BMI trajectory by preeclampsia increased over time (t ratio=3.153, β=0.65, 95% CI 0.11-1.18, p = 0.002)		8
8	Geelhoed JJ et al., 2017	Normal 5345; GH1118; PE205;	9 years old	BMI z score, β (95% CI) GH 0.05 (-0.02–0.12); PE -0.19 (-0.34–0.05) Waist circumference z score, β (95% CI) GH 0.04 (-0.03–0.10); PE -0.20 (-0.34–0.05); Fat mass z score, β (95% CI)	Offspring sex and age, maternal age at delivery, parental pre-pregnancy BMI, parity, social class, and maternal smoking during pregnancy, plus offspring weight, height, and height squared at the 9-year visit	9

(Continued)

TABLE 2 Continued

No	Author/ year	Sample size and grouping	Observed age	Summary of results	Control of confounding factors	NOS
				GH 0.04 (-0.03–0.10); PE -0.09 (-0.23–0.05); Lean mass z score, β (95% CI) GH 0.01 (-0.04–0.05); PE -0.19 (-0.28–0.09); Obese, OR (95% CI) GH 1.37 (0.99–1.89); PE 0.50 (0.21–1.20); Overweight or obese, OR (95% CI) GH 1.08 (0.90–1.30); PE 0.60 (0.39–0.93); Central obesity, OR (95% CI) GH 1.04 (0.89–1.21); PE 0.81 (0.57–1.15);		
9	Palma Dos Reis CR et al., 2021	Normal 5066; PE 67;	10 years old	BMI Z score, β (95%CI) PE -0.014 (−0.300, 0.272); Overweight/obesity status, OR (95% CI) PE 1.23 (0.74, 2.03);	BMI Z score: Pre-pregnancy BMI, primipara status, and tobacco smoke during pregnancy Overweight/obesity status: pre-pregnancy BMI, multipara status	9
10	Ogland B et al., 2009	Control: 194 pairs of mother and daughter and 166 pairs of mother and son; PE: 91 pairs of mother and daughter and 92 pairs of mother and son;	Girl: 10.8 years old; Boy: 11.8 years old;	BMI (female), mean PE 18.4; non-PE 17.5; Difference (95% CI) 0.96 (0.2, 1.7)*;	weight for gestational age (z score)	9
11	Aris IM et al., 2018	Normal 1194; GH 97; PE 45	From birth to 131.2 months	Predictors of age at BMI peak and rebound (in months) from stepwise regression analyses, β (95%CI) GH -0.1(-0.6, 0.5); PE 1.6(0.8, 2.4);		9

12	Byberg KK et al., 2017	Control 385;
		PE 229 (mild/ moderate: 164, 54)
			13 years old	weight SDS, β (95%CI)
				mild/moderate PE 0.08(-0.16,0.33);
				severe PE -0.19(-0.59, 0.21);
				BMI, β (95%CI)
				mild/moderate PE 0.21(-0.01,0.44);
				severe PE 0.14 (-0.24, 0.52);
				Waist-to-height ratio, β (95%CI)
				mild/moderate PE -0.16 (-0.06,0.38);
				severe PE 0.47 (0.15,0.79);
					Child's sex, Birth order, Maternal BMI, Maternal smoking in pregnancy, maternal age at delivery, maternal education at the time of deliver	9

13	Washburn L et al., 2013	Normal 121; PE 51	14 years old	BMI (kg/m^2), mean differences (95% CI) Male adolescent 1.0 (-0.7, 2.7); Female adolescent -0.4 (-2.1, 1.3); Abdominal circumference (cm), mean differences (95%CI) Male adolescent 2.6 (-2.4, 7.5); Female adolescent -1.9 (-6.1, 2.3); Triceps skinfold thickness (mm), mean differences (95%CI) Male adolescent -0.2 (-3.7,3.4); Female adolescent 1.5 (-1.3, 4.4); Subscapular skinfold thickness(mm), mean differences(95%CI) Male adolescent 0.4 (-3.8, 4.6); Female adolescent 1.4 (-2.5, 5.3); Percent body fat, mean differences (95%CI) Male adolescent -0.3 (-4.5, 3.9); Female adolescent -0.3 (-3.1, 2.6);	Antenatal steroid exposure and race, birth weight z score, weight z score at 1-y corrected age–birth weight z score, weight z score at 14 y–weight z score at 1-y corrected age.	9

(Continued)

TABLE 2 Continued

No	Author/ year	Sample size and grouping	Observed age	Summary of results	Control of confounding factors	NOS
14	Miettola S et al. 2013	Control 5045; GH 331; PE 197	16 years old	Cholesterol (mmol/L), percentage difference (95% CI) GH 2.3 (0.2,4.4); PE 0.1 (-2.7,2.9); LDL (mmol/L), percentage difference (95% CI) GH 2.1 (-0.8, 5.1); PE -0.1 (-3.9, 3.9); HDL (mmol/L), percentage difference (95% CI) GH 2.0 (-0.3, 4.4); PE -0.3 (-3.4, 2.8); Triglycerides (mmol/L), percentage difference (95% CI) GH -3.6(-8.0, 1.1); PE -1.2(-7.4, 5.3);	Sex, nulliparity, maternal pre-pregnancy BMI and socioeconomic position, offspring BMI at age 16, offspring birth weight	9
15	Davidesko S et al., 2020	Control 243701; PE 10107	18 years old	Overweight and obesity rate, % PE 0.4; Non- PE 0.2**;	Crude	8
16	Vatten LJ et al., 2003	Normal 3486; PE 220	13-19 years old	Weight(kg), mean (95%CI) PE 62.5 (61.3,63.7); Non-PE 59.1 (58.8, 59.4) **; BMI (kg/m^2), mean (95%CI) PE 22.6 (22.2, 23.0); Non-PE 21.5 (21.3, 21.6)**; Waist (cm), mean (95%CI) PE 72.6 (71.6, 73.6); Non-PE 70.5 (70.3, 70.8)**;	Birth weight, gestational age at birth, and age at survey attendance	9

GH, gestational hypertension; PE, preeclampsia; β, regression coefficient; SE, standard error; IUGR, intrauterine growth retardation; SD, standard deviation; BMI, body mass index; CI, confidence interval; SDS, standard deviation score; LDL, low-density lipoprotein cholesterol; HDL, high-density lipoprotein cholesterol.
*P<0.05; **P<0.01.

7 and one conducted at 1.5-6 years of age. For adverse birth outcomes, 4 out of 15 studies observed the association in offspring with the adverse birth outcome, including intrauterine growth retardation (IUGR), very low birth weight (VLBW), small for gestational age (SGA), and preterm birth. For the other 11 studies on all offspring, the outcomes were different, with 9 comparing the continuous adiposity variables, 3 comparing the obesity/overweight prevalence, and 2 exploring the growth trajectory. The adverse birth outcomes and adiposity outcome measurements were concluded in Table 3.

TABLE 3 The adverse birth outcomes and adiposity outcome measurements of the included studies.

Author	Adverse birth outcomes	Continuous variable		Categorical variable(rate)		Growth trajectory
		Univariate analysis	Multivariate analysis	Univariate analysis	Multivariate analysis	
Baulon E et al.	●	●	●			●
Gow ML et al.	●	●				●
Silveira RC et al.	●	●				
Jiang W et al.			●			
Randhir K et al.		●	●			
Huang Y et al.						●
Geelhoed JJ et al.		●	●		●	
Palma Dos Reis CR et al.		●	●		●	
Ogland B et al.		●	●			
Aris IM et al.						●
Byberg KK et al.			●			
Washburn L et al.	●	●	●	●		●
Miettola S et al.		●	●			
Davidesko S et al.				●		
Vatten LJ et al.			●			

The analysis of all offspring

Continuous variable

During early life (28 days-36 months), maternal exposure to GH/PE was found to be not or inversely associated with offspring obesity. One study found that there was no difference in weight at 28 and 42 days postpartum between the exposed group and the control group in both univariate and multivariate analysis (26). The study by Gow ML et al. found that at 6 months, the weight, and weight Z score of offspring exposed to PE remained significantly lower compared with the control group, but there was a significant absolute increase in BMI and weight from birth to 6 months (27). In a study by Jiang W et al., both GH and PE were inversely associated with offspring's weight and weight-for-age Z score from birth to the age of 36 months, but the associations became nonsignificant after adjusting for birth weight in PE-exposed infants (28).

However, GH/PE tended to be positively associated with offspring adiposity indicators at larger ages (3-19 years old). Four studies adjusted for potential confounders including birth weight, gestational age at birth, maternal BMI, maternal height, maternal smoking status, maternal age at delivery, maternal education, and other factors, and all found a positive association between *de novo* HDP and offspring adiposity indicators (weight, BMI, waist-to-height ratio SDS, total cholesterol, waist circumference, and hip circumference) at the age of 3-19 (22, 29–31).

There were 2 studies with inconsistent findings. In a study by Geelhoed JJ et al., for offspring aged 9 years, the previous positive association between GH and offspring adiposity outcomes became insignificant after adjusting for gestational weeks and birth weight, but the insignificant association between PE and adiposity outcomes (BMI, waist circumference, fat mass) became inverse once the maternal BMI was controlled for. The authors suggested GH/PE may be distinct conditions and that IUGR may be a mediator between PE and offspring obesity (32). Palma Dos Reis CR et al. also found no significant difference in BMI, or BMI *Z* score between PE-exposed 10-year-old offspring and the controls after adjusting for pre-pregnancy BMI and tobacco use. The authors suggested that maternal pre-pregnancy BMI, primiparity, and tobacco use were important confounders of PE and offspring obesity. However, as the authors pointed out, one of the reasons for the inconsistent results may be the limited number of PE cases (n=67) (33).

Categorical variable (overweight/obesity)

Three studies compared the overweight/obesity prevalence of offspring between the GH/PE-exposed and normotensive groups. In a study by Geelhoed JJ et al., a positive association of offspring overweight/obesity with GH was identified, but offspring exposed to PE were less likely to be obese at 9 years old compared with the normotensive group (32). Davidesko S et al. observed a higher prevalence of overweight and obesity in 18-year-old offspring born to mothers with PE (34). However, Palma Dos Reis CR et al. found that there was no association between PE and risk of obesity at 10 years old (33). The inconsistent results may be due to different sample sizes and study populations, as well as pathological differences between GH and PE, which needed to be further confirmed.

Growth trajectory

There were 2 studies evaluating the BMI trajectories of offspring and both found the growth trend related to obesity. Using mixed-effects models to fit BMI curves, Aris IM et al. indicated that PE-exposed children had a later age (1.8 months) at the peak of the BMI trajectory than the control group, which was associated with obesity later in childhood (35). For the offspring at larger age, the study by Huang Y et al. used hierarchical linear modeling and explored that the BMI trajectory of the PE group increased over time from 18 months to 6 years compared to the non-exposed group (36).

Analysis of offspring with adverse birth outcomes

Baulon E et al. found that GH/PE-exposed offspring with IUGR had a significantly lower birth weight but higher infant growth percentage (defined as weight gain from birth to infancy divided by birth weight) at 28 and 42 days postpartum compared to the group without IUGR (26). The other study found that PE-exposed infants with SGA had significantly greater weight Z score gain compared with PE-exposed infants without SGA from birth to 6 months (27). And among PE-exposed and non-exposed infants with VLBW, Silveira RC et al. observed lower weight, weight/age Z score, and weight/length Z score at the 12- and 18-months corrected age in the PE-exposed group (37). Washburn L et al. focused on 14-year-old offspring born prematurely and found PE-exposed offspring had higher BMI, waist circumference, subscapular skinfold thickness, and triceps skinfold thickness compared to offspring born prematurely after normotensive pregnancies, and the association still existed after adjusting for birth weight Z score. In addition, males with PE exposure showed more weight gain during infancy, and females with PE exposure showed more weight gain from 1-year corrected age to 14 years compared to the control group (38). The studies above suggested that GH/PE-exposed offspring with adverse birth outcomes had a lower weight in early life (28 days to 18 months), but there was a rapid weight gain as they grew older.

Subgroup results

Three studies further discussed the influence of different types or severity of *de novo* HDP. In the study by Geelhoed JJ

et al., GH and PE showed opposite effects on obesity among 9-year-old children (32). Another study indicated that offspring exposed to GH tended to have higher lipid levels, but no association was observed between PE and offspring lipid values, suggesting these two types of HDP may have different mechanisms underlying their effects on offspring growth (22). Byberg KK et al. classified PE into mild/moderate and severe groups and found that mild/moderate PE was positively associated with body weight and BMI at 13 years old, but severe PE was negatively associated with them (30). Overall, given the small amount of available evidence and the lack of consistency across studies, it is unclear whether there were differences in the effects of different types (PE/GH) or severity on offspring obesity.

Four studies discussed the different effects of *de novo* HDP between girls and boys. One study showed that, for adolescents aged 14 years who were born prematurely with VLBW, male offspring exposed to PE had higher levels of adiposity, but no significant association was found among females (38). Ogland B et al. found significantly higher obesity indicators in female offspring aged 10.8 years of mothers with both PE and obesity (BMI≥30) but found no significant results in male offspring aged 11.8 years. The association of PE with adiposity measurements was significantly different between girls and boys when testing for interaction by gender (39). The other 2 studies also tested and found the interaction effects of gender with GH/PE (30, 33). Although the results for each gender were inconsistent, significant interaction terms of GH/PE and gender suggested that *de novo* HDP may have gender-specific effects.

Meta-analysis

There were 3 studies with available BMI Z score data, two of which focused on the PE group only, and one included both GH and PE. As shown in Figure 2, the heterogeneity of the results for PE offspring was moderate(I^2 = 65%). The mean difference in BMI Z score between exposed and control groups was insignificant (MD=-0.04, 95%CI: -0.25, 0.17). For offspring exposed to PE or GH, the heterogeneity was moderate (I^2 = 70%), and the mean difference in BMI Z score between groups was also insignificant (MD=0.00, 95%CI: -0.15, 0.15) (Figure 3).

Only 2 studies with BMI data met the criteria for meta-analysis, and the offspring of both studies were female and over 10 years old. The heterogeneity was low (I^2 =0%). The results showed that the PE-exposed group had significantly higher BMI compared to the normotensive group (MD=1.04, 95% CI: 0.67~1.42, P < 0.05) (Figure 4). One study adjusted for birth weight, gestational age at birth, and age at survey attendance, and the other study adjusted for the Z score of weight for gestational age.

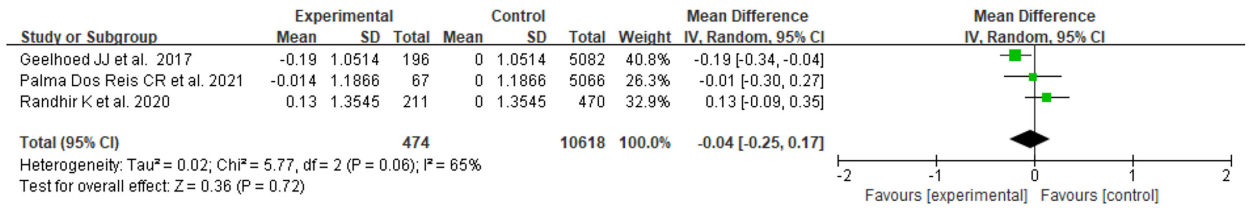

FIGURE 2
Forest plot for the mean difference in BMI Z score between PE-exposed and unexposed offspring.

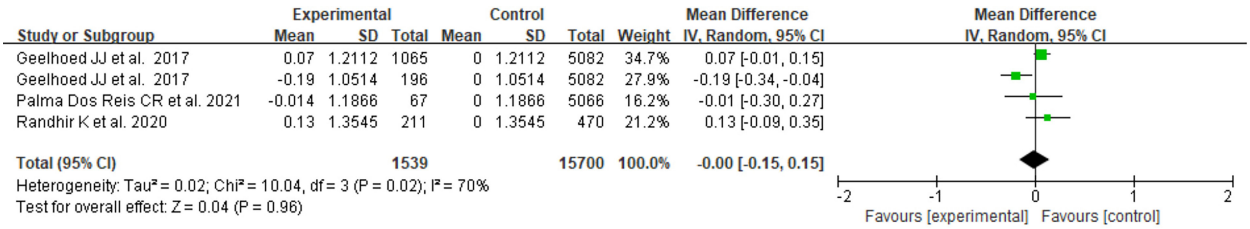

FIGURE 3
Forest plot for the mean difference in BMI Z score between GH- or PE-exposed and unexposed offspring.

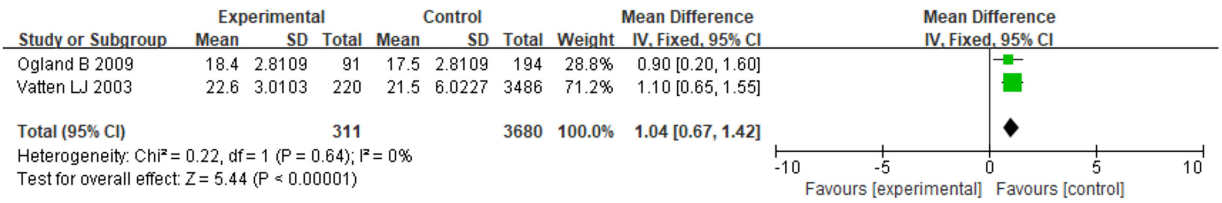

FIGURE 4
Forest plot for the mean difference in BMI between PE-exposed and unexposed female offspring.

Discussion

A total of 16 studies were included in our study, 15 of which were evaluated as high quality. In all offspring, GH/PE was found to be not or inversely associated with offspring adiposity outcomes in the early life (28 days-36 months) and became positively associated at larger ages (3-19 years old). In offspring with adverse birth outcomes, GH/PE-exposed group had a lower weight in the short term (28 days to 18 months), but there was a trend of rapid weight gain as they grew older, compared with the non-exposed group. Although the results of obesity prevalence were inconsistent, studies on growth trajectories also found a trend toward obesity in GH/PE offspring. Subgroup analyses indicated that the association might differ in different types of HDP or different genders. The meta-analysis supported that maternal PE exposure was associated with obesity in female offspring. There were no significant results for BMI Z score, which may be related to the large heterogeneity (I^2 =70%). And although the study of Palma Dos Reis CR et al. adjusted for confounding factors, it did not analyze important factors such as birth weight and gestational age, which may also be the reason for the biased results. Although the BMI indicator was inferior to the BMI Z score, after adjusting for covariates when analyzing BMI, we considered the conclusions to be suggestive.

Although adverse birth outcomes might partly mediate the association of GH/PE with rapid weight gain, GH/PE itself could have other mechanisms underlying the incidence of offspring adiposity. Offspring exposed to PE mothers are associated with adverse birth outcomes (e.g. SGA, IUGR) (37), and catch-up growth is often observed in these offspring (40). For example, in the study by Gow ML et al., the PE group had a higher proportion of SGA and higher weight gain than the control group. However, in the study of Washburn L et al., the association between *de novo* HDP and greater adiposity was robust after adjustment for birth weight Z score (38). HDP was related to dysregulated maternal inflammatory responses to pregnancy and could reduce the transportation of nutrients and oxygen to the fetus. According to the DOHaD, suboptimal fetal nutrition may have an epigenetic change, and the fetus would undergo adaptive physiological changes (41, 42). And by microarray analysis, PE was found to be associated with the up-regulated obese gene (43). Although in the early stage, the difference in gene expression was not significant, it would start to exert a long-term impact on adiposity outcomes.

Current evidence suggested that GH and PE might influence offspring adiposity outcomes through different mechanisms. They may have different pathophysiological effects on endothelial function, placental function, and subsequent programming of fetal growth and development (44). Different adverse birth outcomes of GH or PE also supported their different pathologic mechanisms. Researchers pointed out that PE significantly increased the risk of adverse outcomes such as placental abruption, SGA, 5 min Apgar score < 7 but GH increased the risk of preterm birth (45). In addition, a cohort study identified different serum cytokine profiles for GH and PE (46). Further research is needed to confirm the different impacts of these two diseases on offspring adiposity. For PE of varying severity, evidence showed that the inconsistent results may be related to the levels of IGF-1, which would influence prematurity and inflammation (47). In addition, early PE (before 34 + 0 weeks) and late PE (after 34 + 0 weeks) were considered distinct diseases as they were caused by different hemodynamic states (48).

In different genders, GH/PE may have different associations with offspring adiposity outcomes. Some studies have found the interaction effects of gender with GH/PE on childhood obesity. For explanation, first, different genders were considered to hold different sensitivity to tolerance to the environment and pregnancy outcomes. The male gender has been identified in many studies as an independent risk factor for adverse childbirth outcomes (49–51). Therefore, male infants exposed to GH/PE may have a more significant response. Second, altered placental function leads to related hormonal changes, including decreased estrogens, and increased progesterone and androgens (52). These sex hormones play different roles in different genders, leading to differences in growth and development. Furthermore, fetal exposure to glucocorticoids plays an important role in postpartum reproductive development (53). The adverse fetal environment, including HDP, can cause glucocorticoid disorders by regulating the expression of 11β-hydroxysteroid

dehydrogenase (11β-HSD) in the placenta (54). In animal experiments, it was found that females had a greater response to glucocorticoids than males (55). In addition, leptin levels in fetuses and children also exhibit sexual dimorphism (56). The combination of gender-different susceptibility, complex hormonal changes, or other underlying factors can influence the growth trends. More research is needed to clarify the specific mechanisms and provide important implications for gender-specific interventions.

The control of confounding factors is important to the credibility of research conclusions. The studies included in our meta-analysis ensured that the results were adjusted for important factors. However, this criterion did not present in the previous systematic review (24). In the early life period, maternal factors and the state of birth can play an important role in the appearance of offspring adiposity outcomes. Maternal pre-pregnancy BMI, smoking status, parity, gestational diabetes, maternal tobacco usage during pregnancy, and maternal socioeconomic status are strong confounders of the association between maternal GH/PE and childhood obesity (30, 33). In the study by Gow ML et al., there was a lower rate for PE mothers to be breastfeeding at discharge and 6 months (27), indicating potential influence caused by breastfeeding. Especially in the poor area with lower socioeconomic strata, vulnerable infants are less likely to be breastfed than in other more developed areas (57). As children grow older, their weight can be highly influenced by lifestyle, which was also an important factor to be controlled for exploring the association between maternal GH/PE and childhood obesity (58). Differences in controlling for confounding factors among studies are also one of the reasons for inconsistent findings.

The findings of this study have important implications for the management of obesity in offspring. Maternal GH/PE-exposed offspring were observed to have more adverse birth outcomes and these offspring would present with significant catch-up growth. Although catch-up growth has short-term benefits, allowing newborns to erase the growth deficit and reduce hospital admission rates and mortality (40), it is related to long-term negative consequences such as obesity, hypertension, dyslipidemia, and insulin resistance in adulthood (59–61). Balancing the short-term benefits and adverse long-term outcomes of catch-up growth is important. Although the mechanism is unclear, prevention of GH/PE may be beneficial in alleviating childhood obesity. Thus, special attention should be paid to high-risk pregnant women, such as those with advanced age, obesity, or a history of HDP. Furthermore, for offspring exposed to GH/PE, it is important to start the management of weight growth at an early age.

Our research has three strengths. First, we used relatively strict inclusion and exclusion criteria to include high-quality studies. Studies that included women with pre-existing hypertension or those with unknown disease classification were excluded. And the type of study was strict to the cohort study, which guaranteed the quality of evidence to some extent. Second, previous systematic reviews only analyzed BMI, which could not comprehensively represent the obesity status of children (24). In this study, the outcome indicators were diverse. In addition to the main adiposity indicators (weight, BMI, etc.), we also summarized the results of other indicators (triceps skinfold thickness, waist-hip ratio, etc.). Third, we limited the observation age range. The outcome of the offspring observed in this study was 0-18 years old (only one article included a small number of 19-year-olds). This stage is the most important growth and development period in the entire life stage and is an important window for intervention.

However, this study has limitations. Few existing studies met the selection criteria, so it was difficult to have large amounts of data and perform subgroup analysis. The observation age also lacked continuity and a limited number of publications made it hard to confirm the influence caused by age heterogeneity. In addition, the control of confounding factors in different studies was inconsistent.

Future researchers could extend more in this field to provide more reliable evidence. First, the follow-up time can be extended. The study by Gow ML et al. did not observe a significant difference between the PE group and the controls, but there was a rapid growth trend, suggesting that the difference might turn out to be significant if follow-up continues (37). Second, in the future, the clinical system for pregnant women should be further improved. Many retrospective cohort studies were based on hospital case data. Although some studies had a large sample size, they lacked accurate diagnosis and treatment information. Third, observation indicators should be diversified. BMI Z score was thought to be more representative of childhood obesity than BMI. More research should be conducted using the BMI Z score for further analysis in the future. It was also recommended to supplement waist to height ratio, skinfold thickness, and other information or apply better measures, such as dual-X-ray absorptiometry in future studies to comprehensively evaluate the obesity status (29). Fourth, since different ages and races may have different results(for example, Indians tend to have lower lean body mass), studies in different populations are necessary to confirm the findings (62).

Conclusion

The systematic review suggested that maternal exposure to *de novo* HDP was associated with obesity in offspring, extending from early childhood to adolescence. The meta-analysis showed that PE was associated with higher BMI in female offspring. More studies are needed to conduct stratified analyses by PE/GH, the severity of HDP, or gender. The future exploratory study should expand to examine the effectiveness of the

prevention of *de novo* HDP in controlling the rapid growth of offspring adiposity.

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.

Author contributions

SY and JL contributed to the literature search, screening, writing, and proofreading of the manuscript. ZL helped to settle disagreements in screening the studies. SZ helped to revise and proofread the manuscript. YJ and HW designed the study, and reviewed and edited the manuscript. All authors contributed to the article and approved the submitted version.

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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.

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Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2022.906781/full#supplementary-material

References

1. World Health Organization. *Obesity and overweight* (2021). Available at: https://www.who.int/en/news-room/fact-sheets/detail/obesity-and-overweight.

2. Geng T, Smith CE, Li C, Huang T. Childhood BMI and adult type 2 diabetes, coronary artery diseases, chronic kidney disease, and cardiometabolic traits: A mendelian randomization analysis. *Diabetes Care* (2018) 41(5):1089–96. doi: 10.2337/dc17-2141

3. Kumar S, Kelly AS. Review of childhood obesity: From epidemiology, etiology, and comorbidities to clinical assessment and treatment. *Mayo Clinic Proc* (2017) 92(2):251–65. doi: 10.1016/j.mayocp.2016.09.017

4. Di Cesare M, Sorić M, Bovet P, Miranda JJ, Bhutta Z, Stevens GA, et al. The epidemiological burden of obesity in childhood: a worldwide epidemic requiring urgent action. *BMC Med* (2019) 17(1):212. doi: 10.1186/s12916-019-1449-8

5. Freedman DS, Khan LK, Dietz WH, Srinivasan SR, Berenson GS. Relationship of childhood obesity to coronary heart disease risk factors in adulthood: the bogalusa heart study. *Pediatrics* (2001) 108(3):712–8. doi: 10.1542/peds.108.3.712

6. Barker DJ. The fetal and infant origins of adult disease. *BMJ (Clinical Res ed)* (1990) 301(6761):1111. doi: 10.1136/bmj.301.6761.1111

7. Gluckman PD, Hanson MA, Cooper C, Thornburg KL. Effect of *in utero* and early-life conditions on adult health and disease. *N Engl J Med* (2008) 359(1):61–73. doi: 10.1056/NEJMra0708473

8. Charalambous M, da Rocha ST, Ferguson-Smith AC. Genomic imprinting, growth control and the allocation of nutritional resources: consequences for postnatal life. *Curr Opin Endocrinol Diabetes Obes* (2007) 14(1):3–12. doi: 10.1097/MED.0b013e328013daa2

9. Heindel JJ, Skalla LA, Joubert BR, Dilworth CH, Gray KA. Review of developmental origins of health and disease publications in environmental epidemiology. *Reprod Toxicol (Elmsford NY)* (2017) 68:34–48. doi: 10.1016/j.reprotox.2016.11.011

10. Vickers MH, Breier BH, Cutfield WS, Hofman PL, Gluckman PD. Fetal origins of hyperphagia, obesity, and hypertension and postnatal amplification by hypercaloric nutrition. *Am J Physiol Endocrinol Metab* (2000) 279(1):E83–7. doi: 10.1152/ajpendo.2000.279.1.E83

11. Luyckx VA. Preterm birth and its impact on renal health. *Semin Nephrol* (2017) 37(4):311–9. doi: 10.1016/j.semnephrol.2017.05.002

12. Jain V, Singhal A. Catch up growth in low birth weight infants: striking a healthy balance. *Rev Endocrine Metab Disord* (2012) 13(2):141–7. doi: 10.1007/s11154-012-9216-6

13. Regitz-Zagrosek V, Roos-Hesselink JW, Bauersachs J, Blomström-Lundqvist C, Cífková R, De Bonis M, et al. 2018 ESC Guidelines for the management of cardiovascular diseases during pregnancy. *Eur Heart J* (2018) 39(34):3165–241. doi: 10.1093/eurheartj/ehy340

14. Shah S, Gupta A. Hypertensive disorders of pregnancy. *Cardiol Clinics* (2019) 37(3):345–54. doi: 10.1016/j.ccl.2019.04.008

15. Umesawa M, Kobashi G. Epidemiology of hypertensive disorders in pregnancy: prevalence, risk factors, predictors and prognosis. *Hypertension Res* (2017) 40(3):213–20. doi: 10.1038/hr.2016.126

16. Thoulass JC, Robertson L, Denadai L, Black C, Crilly M, Iversen L, et al. Hypertensive disorders of pregnancy and adult offspring cardiometabolic outcomes: a systematic review of the literature and meta-analysis. *J Epidemiol Community Health* (2016) 70(4):414–22. doi: 10.1136/jech-2015-205483

17. Mogren I, Högberg U, Stegmayr B, Lindahl B, Stenlund H. Fetal exposure, heredity and risk indicators for cardiovascular disease in a Swedish welfare cohort. *Int J Epidemiol* (2001) 30(4):853–62. doi: 10.1093/ije/30.4.853

18. Mamun AA, Kinarivala MK, O'Callaghan M, Williams G, Najman J, Callaway L. Does hypertensive disorder of pregnancy predict offspring blood pressure at 21 years? evidence from a birth cohort study. *J Hum Hypertension* (2012) 26(5):288–94. doi: 10.1038/jhh.2011.35

19. Ananth CV, Peedicayil A, Savitz DA. Effect of hypertensive diseases in pregnancy on birthweight, gestational duration, and small-for-gestational-age births. *Epidemiol (Cambridge Mass)* (1995) 6(4):391–5. doi: 10.1097/00001648-199507000-00011

20. Ahmad AS, Samuelsen SO. Hypertensive disorders in pregnancy and fetal death at different gestational lengths: a population study of 2 121 371 pregnancies. *BJOG* (2012) 119(12):1521–8. doi: 10.1111/j.1471-0528.2012.03460.x

21. Kuciene R, Dulskiene V. Associations of maternal gestational hypertension with high blood pressure and overweight/obesity in their adolescent offspring: a retrospective cohort study. *Sci Rep* (2022) 12(1):3800. doi: 10.1038/s41598-022-07903-z

22. Miettola S, Hartikainen AL, Vääräsmäki M, Bloigu A, Ruokonen A, Järvelin MR, et al. Offspring's blood pressure and metabolic phenotype after exposure to gestational hypertension in utero. *Eur J Epidemiol* (2013) 28(1):87–98. doi: 10.1007/s10654-013-9763-5

23. Pinheiro TV, Brunetto S, Ramos JG, Bernardi JR, Goldani MZ. Hypertensive disorders during pregnancy and health outcomes in the offspring: a systematic review. *J Dev origins Health Dis* (2016) 7(4):391–407. doi: 10.1017/S2040174416000209

24. Wang LB, Qu B, Xu P, Wu LL, Gu JS, Shah NK, et al. Preeclampsia exposed offspring have greater body mass index than non-exposed offspring during peripubertal life: A meta-analysis. *Pregnancy Hypertension* (2020) 19:247–52. doi: 10.1016/j.preghy.2019.09.010

25. Palti H, Rothschild E. Blood pressure and growth at 6 years of age among offsprings of mothers with hypertension of pregnancy. *Early Hum Dev* (1989) 19 (4):263–9. doi: 10.1016/0378-3782(89)90061-3

26. Baulon E, Fraser WD, Piedboeuf B, Buekens P, Xiong X. Pregnancy-induced hypertension and infant growth at 28 and 42 days postpartum. *BMC pregnancy childbirth* (2005) 5:10. doi: 10.1186/1471-2393-5-10

27. Gow ML, Roberts L, Henry A, Davis G, Mangos G, Pettit F, et al. Growth from birth to 6 months of infants with and without intrauterine preeclampsia exposure. *J Dev Origins Health Dis* (2021) 13(2):151–5. doi: 10.1017/S2040174421000167

28. Jiang W, Mo M, Si S, Wu J, Pu L, Huang M, et al. Association of hypertensive disorders of pregnancy with infant growth in the first 36 months of life. *Eur J Pediatr* (2021) 181(1):133–41. doi: 10.1007/s00431-021-04173-1

29. Randhir K, Pisal H, Kadam V, Khaire-Ghadge A, Malshe N, Deshpande R, et al. Association of preeclampsia with anthropometric measures and blood pressure in Indian children. *PloS One* (2020) 15(5):e0231989. doi: 10.1371/journal.pone.0231989

30. Byberg KK, Øymar K, Eide GE, Forman MR, Júlíusson PB. Exposure to preeclampsia *in utero* affects growth from birth to late childhood dependent on child's sex and severity of exposure: Follow-up of a nested case-control study. *PloS One* (2017) 12(5):e0176627. doi: 10.1371/journal.pone.0176627

31. Vatten LJ, Romundstad PR, Holmen TL, Hsieh CC, Trichopoulos D, Stuver SO. Intrauterine exposure to preeclampsia and adolescent blood pressure, body size, and age at menarche in female offspring. *Obstetrics Gynecol* (2003) 101 (3):529–33. doi: 10.1016/s0029-7844(02)02718-7

32. Geelhoed JJ, Fraser A, Tilling K, Benfield L, Davey Smith G, Sattar N, et al. Preeclampsia and gestational hypertension are associated with childhood blood pressure independently of family adiposity measures: the Avon longitudinal study of parents and children. *Circulation* (2010) 122(12):1192–9. doi: 10.1161/CIRCULATIONAHA.110.936674

33. Palma Dos Reis CR, Serrano F, Fonseca MJ, Martins AT, Campos A, McMahon GM, et al. The fetal origins of disease: a prospective cohort study on the association of preeclampsia and childhood obesity. *J Dev Origins Health Dis* (2021) 13(1):68–74. doi: 10.1017/S2040174421000027

34. Davidesko S, Nahum Sacks K, Friger M, Haim A, Sheiner E. Prenatal exposure to preeclampsia as a risk factor for long-term endocrine morbidity of the offspring. *Hypertension Pregnancy* (2021) 40(1):21–8. doi: 10.1080/10641955.2020.1854300

35. Aris IM, Rifas-Shiman SL, Li LJ, Kleinman K, Coull BA, Gold DR, et al. Pre-, perinatal, and parental predictors of body mass index trajectory milestones. *J Pediatr* (2018) 201:69–77.e8. doi: 10.1016/j.jpeds.2018.05.041

36. Huang Y, Zhang W, Go K, Tsuchiya KJ, Hu J, Skupski DW, et al. Altered growth trajectory in children born to mothers with gestational diabetes mellitus and preeclampsia. *Arch Gynecol Obstetrics* (2020) 301(1):151–9. doi: 10.1007/s00404-020-05436-2

37. Silveira RC, Procianoy RS, Koch MS, Benjamin AC, Schlindwein CF. Growth and neurodevelopment outcome of very low birth weight infants delivered by preeclamptic mothers. *Acta Paediatrica (Oslo Norway 1992)* (2007) 96(12):1738–42. doi: 10.1111/j.1651-2227.2007.00552.x

38. Washburn L, Nixon P, Russell G, Snively BM, O'Shea TM. Adiposity in adolescent offspring born prematurely to mothers with preeclampsia. *J Pediatr* (2013) 162(5):912–7.e1. doi: 10.1016/j.jpeds.2012.10.044

39. Ogland B, Vatten LJ, Romundstad PR, Nilsen ST, Forman MR. Pubertal anthropometry in sons and daughters of women with preeclamptic or normotensive pregnancies. *Arch Dis Childhood* (2009) 94(11):855–9. doi: 10.1136/adc.2008.150870

40. Victora CG, Barros FC, Horta BL, Martorell R. Short-term benefits of catch-up growth for small-for-gestational-age infants. *Int J Epidemiol* (2001) 30(6):1325–30. doi: 10.1093/ije/30.6.1325

41. Barker DJ, Gluckman PD, Godfrey KM, Harding JE, Owens JA, Robinson JS. Fetal nutrition and cardiovascular disease in adult life. *Lancet (London England)* (1993) 341(8850):938–41. doi: 10.1016/0140-6736(93)91224-A

42. Heijmans BT, Tobi EW, Stein AD, Putter H, Blauw GJ, Susser ES, et al. Persistent epigenetic differences associated with prenatal exposure to famine in humans. *Proc Natl Acad Sci USA* (2008) 105(44):17046–9. doi: 10.1073/pnas.0806560105

43. Reimer T, Koczan D, Gerber B, Richter D, Thiesen HJ, Friese K. Microarray analysis of differentially expressed genes in placental tissue of pre-eclampsia: up-regulation of obesity-related genes. *Mol Hum Reprod* (2002) 8(7):674–80. doi: 10.1093/molehr/8.7.674

44. de Beer M, Vrijkotte TG, van der Wal MF, van Weissenbruch MM, Gemke RJ. Relation of maternal hypertension with infant growth in a prospective birth cohort: the ABCD study. *J Dev Origins Health Dis* (2010) 1(5):347–55. doi: 10.1017/S2040174410000462

45. Shen M, Smith GN, Rodger M, White RR, Walker MC, Wen SW. Comparison of risk factors and outcomes of gestational hypertension and pre-eclampsia. *PLoS One* (2017) 12(4):e0175914. doi: 10.1371/journal.pone.0175914

46. Tangerås LH, Austdal M, Skråstad RB, Salvesen K, Austgulen R, Bathen TF, et al. Distinct first trimester cytokine profiles for gestational hypertension and preeclampsia. *Arteriosclerosis Thrombosis Vasc Biol* (2015) 35(11):2478–85. doi: 10.1161/ATVBAHA.115.305817

47. Hellström A, Ley D, Hansen-Pupp I, Hallberg B, Löfqvist C, van Marter L, et al. Insulin-like growth factor 1 has multisystem effects on foetal and preterm infant development. *Acta Paediatrica (Oslo Norway 1992)* (2016) 105(6):576–86. doi: 10.1111/apa.13350

48. Valensise H, Vasapollo B, Gagliardi G, Novelli GP. Early and late preeclampsia: two different maternal hemodynamic states in the latent phase of the disease. *Hypertension (Dallas Tex 1979)* (2008) 52(5):873–80. doi: 10.1161/HYPERTENSIONAHA.108.117358

49. Aibar L, Puertas A, Valverde M, Carrillo MP, Montoya F. Fetal sex and perinatal outcomes. *J Perinatal Med* (2012) 40(3):271–6. doi: 10.1515/jpm-2011-0137

50. Simchen MJ, Weisz B, Zilberberg E, Morag I, Weissmann-Brenner A, Sivan E, et al. Male Disadvantage for neonatal complications of term infants, especially in small-for-gestational age neonates. *J Maternal-fetal Neonatal Med* (2014) 27 (8):839–43. doi: 10.3109/14767058.2013.845658

51. Månsson J, Fellman V, Stjernqvist K. Extremely preterm birth affects boys more and socio-economic and neonatal variables pose sex-specific risks. *Acta paediatrica (Oslo Norway 1992)* (2015) 104(5):514–21. doi: 10.1111/apa.12937

52. Ros HS, Lichtenstein P, Ekbom A, Cnattingius S. Tall or short? twenty years after preeclampsia exposure *in utero*: comparisons of final height, body mass index, waist-to-hip ratio, and age at menarche among women, exposed and unexposed to preeclampsia during fetal life. *Pediatr Res* (2001) 49(6):763–9. doi: 10.1203/00006450-200106000-00008

53. Baxter JD, Rousseau GG. Glucocorticoid hormone action: an overview. *Monogr Endocrinol* (1979) 12:1–24. doi: 10.1007/978-3-642-81265-1_1

54. Burton PJ, Waddell BJ. Dual function of 11beta-hydroxysteroid dehydrogenase in placenta: modulating placental glucocorticoid passage and local steroid action. *Biol Reprod* (1999) 60(2):234–40. doi: 10.1095/biolreprod60.2.234

55. Smith JT, Waddell BJ. Increased fetal glucocorticoid exposure delays puberty onset in postnatal life. *Endocrinology* (2000) 141(7):2422–8. doi: 10.1210/endo.141.7.7541

56. Hassink SG, Sheslow DV, de Lancey E, Opentanova I, Considine RV, Caro JF. Serum leptin in children with obesity: relationship to gender and development. *Pediatrics* (1996) 98(2 Pt 1):201–3. doi: 10.1542/peds.98.2.201

57. Gibbs BG, Forste R. Socioeconomic status, infant feeding practices and early childhood obesity. *Pediatr Obes* (2014) 9(2):135–46. doi: 10.1111/j.2047-6310.2013.00155.x

58. Kvehaugen AS, Andersen LF, Staff AC. Dietary intake and physical activity in women and offspring after pregnancies complicated by preeclampsia or diabetes mellitus. *Acta Obstetricia Gynecologica Scandinavica* (2010) 89(11):1486–90. doi: 10.3109/00016349.2010.519378

59. Kerkhof GF, Willemsen RH, Leunissen RW, Breukhoven PE, Hokken-Koelega AC. Health profile of young adults born preterm: negative effects of

rapid weight gain in early life. *J Clin Endocrinol Metab* (2012) 97(12):4498–506. doi: 10.1210/jc.2012-1716

60. Leunissen RW, Kerkhof GF, Stijnen T, Hokken-Koelega A. Timing and tempo of first-year rapid growth in relation to cardiovascular and metabolic risk profile in early adulthood. *Jama* (2009) 301(21):2234–42. doi: 10.1001/jama.2009.761

61. Singhal A, Lucas A. Early origins of cardiovascular disease: is there a unifying hypothesis? *Lancet (London England)* (2004) 363(9421):1642–5. doi: 10.1016/S0140-6736(04)16210-7

62. Yajnik CS. Confessions of a thin-fat Indian. *Eur J Clin Nutr* (2018) 72 (4):469–. doi: 10.1038/s41430-017-0036-3

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#childhoodobesity – A brief literature review of the role of social media in body image shaping and eating patterns among children and adolescents

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Children's food preferences are closely related to their parents' food preferences and knowledge of food is linked to what their parents share with them. Parents, however, are not the only people who model such behavior. Paradoxically, the ubiquitous technological development can also pose a huge threat. In developed countries, 94% of teenagers use social media platforms such as: Instagram, Snapchat, Facebook, or TikTok, and this also applies to children. It can therefore be argued that parents' nutritional preferences and behavior are related to the same behaviors of children and there is an extensive literature on this subject. It is therefore important to check how other factors – new technology (and social media in particular) – can influence changes in this area. A literature search was conducted in the following databases: Google Scholar, PubMed, EBSCO in December 2021. After applying all the filters and verification of relevance in terms of the research on the topic of interest to us, 4 articles related to research on body image and social media and 4 articles related to research on food choices and social media among children and adolescents were obtained. The conducted analysis showed that various studies so far indicate that social media can have a very strong influence on the development of eating patterns and body image in children and adolescents, which in turn may be one of the risk factors for developing obesity when promoted behaviors are not associated with a healthy lifestyle. It is also worth pointing out that social media can be used as a resource in the prevention and treatment of obesity. A closer look at this topic seems to be particularly important due to the fact that, among adults, social media is not only a very important source of information about lifestyle, but also a source of social support when people attempting to lose weight.

Therefore, by increasing preventive activity in social media and using modern solutions related to social media (including the use of hashtag signs), we can have a greater impact on the health awareness of children and adolescents around the world.

KEYWORDS

childhood obesity, social media, body image, eating patterns, children, adolescents

Introduction

Childhood is the most important period in which eating habits are formed, which undoubtedly influences later health condition (1). The result of the transmission of incorrect eating patterns is obesity, other metabolic diseases and eating disorders (2–4). As is commonly known, these disorders are more and more often diagnosed in children and adolescents, and the recent situation related to COVID (e.g., due to the frequent change of lifestyle to a less healthy one) was conducive to the development of excessive body weight in this age group (e.g., (5–11).

The first educator in the field of eating behavior is the family (12). Research shows that children's food preferences are closely related to their parents' food preferences. Also, children's knowledge of food is linked to what their parents share with them (13). It is well known that children learn to eat through their own experience as well as through the observation of others (14). It can therefore be argued that parents' nutritional preferences and behavior are related to the same behaviors of children and there is an extensive literature on this subject (15–17). As the knowledge about eating behaviors is quite extensive, so it is important to check how new technology (and social media in particular) can influence changes in this area.

Parents, however, are not the only people who model such behavior (18). Paradoxically, the ubiquitous technological development, which is supposed to be a convenience, can also pose a huge threat (19). In developed countries, 94% of teenagers use social media platforms such as: Instagram, Snapchat, Facebook, or TikTok (20), and this also applies to children (21). Many studies around the world indicate that children and adolescents use social media for up to several hours a day, an example of such research is an Iranian study in which almost 80% of children and adolescents use social media 3-4 h or more a day (19). Using these media, we often come across information on the subject of obesity, which can very quickly spread around the world, including by using a hashtag (#) for them appropriately (22). For example, when analyzing the titular #childhoodobesity, on the one hand, we come across (good quality) educational materials that raise people's awareness of the prevention and treatment of obesity. On the other hand, there are also numerous posts that can be strongly stigmatizing. The latter very often relate to two areas of functioning – body image and eating patterns (19). As shown by the research carried out so far, eating patterns and body image can play a very important role in developing obesity (e.g., (10, 23, 24), and one of the theoretical models explaining this is the Homeostatic Theory of Obesity (25). This model has also been empirically verified in studies conducted in a group of Polish children and adolescents (26). However, it is worth continuing research in this area to check how important social media in childhood are in shaping body image and eating patterns, especially given that (in general) children's and adolescents' use of social media has a significant impact on their body mass index (19).

Most children and adolescents publish their photos on the above-mentioned portals, so-called "selfies" (27). Considering the sociocultural model (28), teenagers internalize the ideals of appearance that are conveyed by the media and make comparisons with them (29, 30). According to the National Eating Disorders Association (31), body image is how an individual believes what they look like in the mirror, how they feel about their body, and how they feel in control about the body. This image can be positive or negative (31). The negative ones are an early indicator of an eating disorder. Regarding the shaping of one's body image and exposure in social media, it is worth mentioning that social media promotes an unnaturally slim figure as desirable, often building a negative body image, and children and adolescents seem to be the most vulnerable group (31). This pattern often promotes stigma and strengthens the tendency of obese children to lose weight by means of maladaptive measures (e.g., very restrictive diets, use of laxatives, vomiting), which often lead to further weight gain in the long term (19, 32–34). Thus, as research shows, social media and peer groups functioning in it can, on the one hand, be an important source of knowledge and support in preventing obesity and promoting healthy growth (19, 35, 36). On the other hand, they can be a source of great discomfort, spreading myths about obesity and its treatment (19, 33). That is why it is so important to look carefully at how the content available on social media can influence the shaping of the body image and eating patterns of children and adolescents.

There is a strong international interest in research into eating behavior, the transmission of eating patterns through the family environment, and body image formation, but few

studies in this context have analyzed the impact of social media (15–17). Importantly, not only selfies but also sharing pictures of meals with friends is also a popular phenomenon on social media (37). The reaction to such a photo is reacted on social media can also perpetuate and modify various eating and lifestyle behaviors. Therefore, this review of research aims to: (I) summarize the current research on the role of social media in the group of children and adolescents in (Ia) body image shaping, (Ib) shaping eating patterns, (II) indicating the essence of the problem and the direction of obesity preventive measures to public health institutions and other entities with a significant influence on the promotion of healthy eating behavior, mental health, and the proper use of social media, and (III) indicating further studies directions among obese children and adolescents in the context described above.

Materials and methods

A literature search was conducted in the following databases: Google Scholar, PubMed, EBSCO from December 2021 to January 2022. The following keyword alone combinations were used: body image, body image and social media, food choices and social media, food choices, food choices and Facebook/TikTok/Instagram/Snapchat, body image and Facebook/TikTok/Instagram/

Snapchat. Filters in search engines were also used, such as: "Free full text," ten-year articles, English and Polish, and the target research group "children and adolescents."

After applying all the filters, 26 results related to the subject of body image in social media and 8 results related to food choices in social media were obtained. Then, all the results were verified in terms of the relevance of the research on the topic of interest to us. As a result, 4 articles related to research on body image and social media among children and adolescents and 4 articles related to research on food choices and social media among children and adolescents were obtained. A block diagram of this process is shown in **Figure 1**. The "Supplementary Material" section provides more details about these studies (i.e., group description, variables and measures, results and detailed statistics for the measured variables; (Appendix **Table 1**)).

Results

Body image and social media

Self-body perceptions, especially among girls in recent decades, have become a cause of global adolescent self-esteem (38). The basis of adolescent self-presentation is increasingly based on photos and videos on social media (39, 40). Social media can create appearance standards that are difficult to achieve (39), especially by adolescents and children. This situation can lead to lower self-esteem and emotional disturbances (41).

Previous research, for example, conducted in Norway on a group of 1998 respondents aged 10-14 (boys and girls), looked at which social media platforms they use, how often they post something on their account per month and how often they post photos of themselves and how often they comment on other people's statuses and photos (41). Results indicate, among other things, that other-oriented social media use lowered self-reported appearance among respondents aged 10-12 and 12-14, while self-oriented use had no effect on this (41).

Another Dutch study, involved 440 teenagers of both genders aged 12 to 19 (29). The study was aimed at, *inter alia*, indicating whether the use of social media is a significant predictor of body dissatisfaction. The results showed that teens who reported more use of social media also reported higher levels of body dissatisfaction (29).

Interesting research in this area was also carried out in Singapore, where 100 female teenagers aged 13 to 18 were recruited from various local communities, such as the Chinese, Malayas, and Indians (42). Total smartphones use time, social media use, cognitive internalization, anxiety about social appearance, respect for the body, and position of weight control were assessed. The results suggest that only excessive use of social media, according to the authors, more than 3 h a day results in lower body evaluation results. Interestingly, the authors also explored the issue of engaging in online and offline appearance comparisons. They found that while social media escalates unhealthy cognitive patterns, it also does so outside of the time spent in these media, harming teenagers' own body assessment, including a girl study (42).

A very different report is indicated by researchers from Denmark, who focused on the effects of social networking on body image among 604 adolescents (male and female) aged 11 to 18 (43). The study was related to the frequency of use of social media and information about the appearance that teenagers obtained from their peers and its impact on their body image. The results show that the more teens used social networking sites, the more often they received feedback about their appearance. Interestingly, the feedback received did not predict body dissatisfaction (43). This finding contradicts most studies about the association of social media with negative body image.

Eating patterns and social media

Visual representations of food and beverage products in traditional communications and digital marketing primarily involve products high in fat, sugar and salt (44). YouTube is also very popular with children aged 5 to 15 (45), and the information contained in the content viewed influences their eating behavior (44). Research shows that exposure to food-related information contained in social media content, known

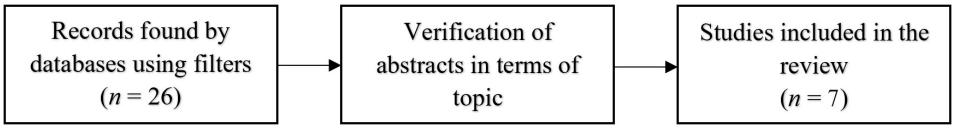

FIGURE 1
Flow chart of article selection process.

as influencers, directly, immediately influences the choice and consumption of promoted foods by 9–11-year-old children (46, 47).

An equally interesting study on the marketing impact of a product promoted via the YouTube platform conducted in qualitative terms is a British study on a group of children (boys and girls) aged 10-11 years (48). The children watched a marketing video promoting the sweet product and were informed that the purpose of the study was to gather their views on YouTubers advertising food and drinks. The results of this study indicated that youtubers are a source of entertainment, information, social acceptance, and experiences for children, moreover, the products promoted by youtubers were desired by children (48).

The same authors also analyzed the channels of Youtubers popular among children to determine the scope and nature of the recommendations of food products and drinks (44). They also examined the proportion of "healthy" and "unhealthy" referrals. As it turned out, each of the commands had at least one food or drink tip, more often they were unhealthy than healthy. As many as 92.6% of the analyzed videos contained food and drink tips, which corresponds to 29.9 tips per hour (44).

A study by American researchers on a sample of 884 male and female adolescents aged 13-17 indicated that food ads posted on Instagram were very attractive to the respondents compared to traditional food ads (49). Interestingly, the Instagram symbol itself caused much more interest in the promoted product (49).

Discussion

Various studies cited so far indicate that social media can have a very strong influence on the development of eating patterns and body image in children and adolescents, which in turn may be one of the risk factors for developing obesity when promoted behaviors are not associated with a healthy lifestyle.

Originally, the sociocultural model proposed by Thompson et al. (28) focused on traditional media, e.g., television, magazines, and the traditional "face-to-face" perception of the other person. Today, teenagers derive their ideal-looking messages from social media. According to the above-mentioned sociocultural model of comparison, the internalization of ideal appearance communicated through social media results in body dissatisfaction (50). This is supported by the analysis of the research presented in this article. In most studies on the impact of social media on body image, the target respondents are teenagers and adults and their results show a negative relationship between social media and body dissatisfaction (e.g., 43, 50–52). Therefore, attention should be paid to the importance of the problem of social media in the context of incorrect body image. If the problem is large among adult users of social media platforms, the group of children and adolescents may be even more at risk (53). Unfortunately, there are few studies that can approximate to the magnitude of this problem among children, and thus allow for the design of prevention activities aimed at child caregivers that would help monitor online behavior and allow for the protection of children from negative self-perception.

Social media also contributes to the promotion of food products to users (54). Influencers, youtubers show specific food products, recommend their purchase, and they are not always healthy (55). These are sponsored advertisements paid for by large food concerns. Research to date provides sufficient evidence of the effectiveness of influencer marketing on consumption primarily among adolescents (56). What is important for such marketing activities is that about 98% of people from the "Z" generation, i.e., people born after 1995, have a smartphone, and moreover, half of teenagers spend 10 or more hours a day using the telephone (57). It is therefore a powerful tool to influence choices, including food preferences. The Norwegian Consumer Council in 2019 showed that about 20% of all influencer-related marketing activities were for food and drinks (58). So far, research on this subject is not sufficient, and as far as we are aware, such research has not been conducted on a Polish sample. The exposure of children and adolescents to the influence of people for whom the most important thing is to sell a not necessarily healthy product is underestimated. Influencers often choose the way they present themselves on the Internet (59), using products and brands for self-presentation rather than actual consumption (60). The cognitive development of younger children (12 years of age and younger) is still developing, and thus a critical understanding of the commercial world will not be the same as the critical thinking skills of adults (61), therefore the popularity of influencers and content that they post on their profiles is particularly attractive to young audiences. Given

TABLE 1 Characteristic of the analyzed studies.

Authors (year)	Sample	Variables and measures	Descriptive statistics for variables and type of study
Steinsbekk et al. (41)	10 years old: N = 702 M_{age} = 10.51; SD = 0.17 12 years old: N = 668 M_{age} = 12.49; SD = 0.15 14 years old: N = 628 M_{age} = 14.33; SD = 0.59 Girls: 10 years old: 52.3% 12 years old: 51.9% 14 years old: 53.0% Boys: 10 years old: 47.7% 12 years old: 48.1% 14 years old: 47.0%	Social media use – report which social media platform use (e.g., Facebook, Instagram, Snapchat, Twitter) + characteristics of their use – self-oriented social media use: (a) the number of times per month they post something of their own social media sites, (b) how often they post photographs (never/rarely/weekly/daily) – other-oriented social media use: how often they commented on others" status updates and photographs, how often they "like"" others" statuses (6-point Likert scale) Appearance self-esteem 10-years old: *Self-Description Questionnaire* (SDQ-I; Marsh, (68) 12- and 14-years old: *Revised Self-Perception Profile for Adolescents* (SPPA-R; Harter (69); Wichstrøm (70)	Cross-secional study 10 years old: – *self-oriented social media use* (M = 10.33; SD = 26.15) – *other-oriented social media use* (M = 4.37; SD = 3.06) – *appearance self-esteem* – (M = 4.11; SD = 0.10) 12 years old: – *self-oriented social media use* (M = 17.95; SD = 30.60) – *other-oriented social media use* (M = 6.38; SD = 2.54) – *appearance self-esteem* – (M = 3.39; SD = 0.53) 14 years old: – *self-oriented social media use* (M = 15.02; SD = 30.19) – *other-oriented social media use* (M = 8.20; SD = 3.10) – *appearance self-esteem* – (M = 3.06; SD = 0.69)
de Vires et al. (29)	N = 440 M_{age} = 14.86; SD = 1.79 Girls: N = 205 Boys: N = 232 12 years: 12% 13 years: 18.2% 14 years: 11.8% 15 years: 13.4% 16 years: 22.6% 17 years: 18.7% 18 years: 3.2% 19 years: 0.2%	Body dissatisfaction *The Body Attitude Test* (BAT), (Probst et al. (71) Social media use *Multidimensional Scale of Facebook Use* (MSFU), (Frison and Eggermont (72) Relationship qualities *The Network of Relationship Questionnaire- Relationship Qualities Version* (NRI-RQV), (Buhrmester and Furman (73)	Cross-secional study *mean score of body dissatisfaction* (M = 2.81; SD = 1.43) – *mean score of social media use* (M = 3.92; SD = 1.41) – *a positive father-adolescent relationship measure* (M = 3.40; SD = 0.64) – *a positive mother-adolescent relationship measure* (M = 3.72; SD = 0.63)
Yang et al. (42)	N = 100 (female adolescents) M_{age} = 15.07; SD = 1.33 M_{BMI} = 19.05; SD = 3.45	Overall smartphone screen time *Smartphone Use* (questions: time on smartphone per day: sending and receiving e-mails, sending and receiving text messages, browsing websites, watching TV shows, taking photos, online shopping, listening to music) Daily frequency engaged in social networking sites *Media and Technology Usage and Attitudes Scale* (Rosen et al. (74) Cognitive internalization of thin ideals *the Sociocultural Attitudes Toward Comparison Scale-3* (Thompson et al. (75) Overall social comparison (online/offline) *The Physical Appearance Comparison Scale-Revised* (Schaefer and Thompson (76) Social appearance anxiety *Social Appearance Anxiety Scale* (Hart et al. (77) Body esteem *Body Esteem Scale for Adolescents and Adults* (Mendelson et al. (78) Internal control beliefs *Dieting Beliefs Scale* (Stotland and Zuroff (79)	Cross-secional study *smartphone screen time* (M = 4.10; SD = 1.41) – *website browsing* (M = 3.44; SD = 1.67) – *emailing* (M = 1.14; SD = 1.48) – *texting* (M = 3.34; SD = 1.60) – *listening to music* (M = 3.17; SD = 1.52) – *taking photos* (M = 1.70; SD = 1.34) – *taking videos* (M = 0.91; SD = 1.27) – *watching TV shows* (M = 3.21; SD = 1.58) – *online shopping* (M = 1.17; SD = 1.41) – *social media screen time* (M = 3.70; SD = 1.44) – *cognitive internalization* (M = 3.10; SD = 1.08) – *appearance comparison* (M = 5.74; SD = 2.20) – *appearance anxiety* (M = 3.37; SD = 0.98) – *body esteem* (M = 2.95; SD = 0.64) – *internal locus of control* (M = 3.33; SD = 0.34)
de Vires et al. (43)	N = 604 M_{age} = 14.7; SD = 1.7 M_{BMI} = 20.04; SD = 3.54 BMI: BMI under 30: 98.9% BMI under 25: 91.4% BMI under 20: 52.3% BMI under 18: 30%	Frequency of social network site use *Social Network Site Use* - questions about how often did you visit. . . in the past 6 months? Peer appearance-related feedback *Peer Appearance-Related Feedback* - questions about how often their friends (1) give them tips how to get a more beautiful body (2) criticize their appearance or clothes (3) give them tips how to look sexy (4) tell them it is important to look good Body dissatisfaction *The Body Areas Satisfaction Scale* (Cash (80) *The Multidimensional Body-Self Relations Questionnaire* (Cash (80)	Cross-secional study – *frequency of social network site use* (M = 2.4; SD = 1.5 – at time 1; M = 2.6; SD = 1.4 – at time 2) – *peer appearance-related feedback* (M = 0.53; SD = 0.57 -at time 1; M = 0.59; SD = 0.60 - at time 2) – *body dissatisfaction* (M = 1.46; SD = 0.65 - at time 1; M = 1.54; SD = 0.65 - at time 2)

(Continued)

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TABLE 1 (Continued)

Authors (year)	Sample	Variables and measures	Descriptive statistics for variables and type of study
Coates et al. (44)	*N* = 24 (six focus groups with children aged 10-11 years) One focus group: (*N* = 4)	Children"s understanding and attitudes about marketing *YouTube Video Featuring Influencer Marketing* (video "Nutella Breakfast Party"") *Photographs of Influencer Marketing Techniques* *Interview Guide*	Qualiative research
Coates et al. (46)	YouTube videos (the authors two influencers - female aged 29, male – aged 24; both with a healthy weight).	*YouTube videos uploaded by two influencers*	Qualiative research Analysis of YouTube video blogs of influencers popular with children and determination of the extent and nature of food and beverage cues featured.
Bragg et al. (49)	*N* = 832 M_{age} = 14.73; *SD* = 1.67 Male: *N* = 426 Female: *N* = 406	Instagram vs non-Instagram ads *Photo presentation and question:* How much do you like this image? *Photo presentation and question:* How artistic is this image? *Photo presentation and question:* How trendy is this image? *Photo presentation and question:* How delicious do you think this product is? *Photo presentation and question:* How likely are you to purchase this product in the next 4 weeks?	Cross-secional study Instagram advertisement Unlabeled advertisement condition/labeled advertisement condition - *How much do you like this image?* (*M* = 68.56; *SD* = 0.93/*M* = 67.43; *SD* = 1.04) - *How artistic is this image* (*M* = 68.58; *SD* = 0.93/*M* = 66.80; *SD* = 1.07) - *How trendy is this image* (*M* = 69.60; *SD* = 0.90/*M* = 68.16; *SD* = 1.02) - *How delicious do you think this product is?* (*M* = 66.80; *SD* = 0.94/*M* = 66.62; *SD* = 1.02) - *How likely are you to purchase this product in the next 4 weeks?* (*M* = 56.25; *SD* = 1.04/*M* = 56.03; *SD* = 1.38) Traditional advertisement Unlabeled advertisement condition/labeled advertisement condition - *How much do you like this image?*(*M* = 65.72; *SD* = 0.93/*M* = 67.11; *SD* = 1.04) - *How artistic is this image* (*M* = 66.86; *SD* = 0.93/*M* = 66.84; *SD* = 1.07) - *How trendy is this image* (*M* = 66.28; *SD* = 0.90/*M* = 66.92; *SD* = 1.02) - *How delicious do you think this product is?* (*M* = 65.66; *SD* = 0.94/*M* = 67.51; *SD* = 1.02) - *How likely are you to purchase this product in the next 4 weeks?* (*M* = 55.27; *SD* = 1.23/*M* = 56.30; *SD* = 1.38)

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this information, it is important to ensure the protection and control of young people in the digital space, and it should also be crucial for preventive health. To effectively protect children and adolescents, an intervention in the use of social media must be developed, and to implement it, a better understanding of how the use of social networking sites affects body image and food choices should be developed. Taking these actions is also important due to such phenomena as "echo chamber" (this can be defined as personalizing the content used on the Internet and matching it to the profile of a specific user, which means that we only receive information on social media that has been determined by appropriate algorithms as consistent with our interests and views; (62) and "mukbang" (this can be defined as an online audiovisual broadcast through a video-streaming platforms such as TikTok or YouTube in which a host consumes different amounts and types of food and interacts with the audience using a multimodal communication; (63), which may have a significant impact on shaping the awareness, body image and eating patterns of children and adolescents.

Summarizing the current knowledge, in future studies related to childhood obesity we should focus on: (I) analysis of the impact on eating patterns and body image of content from TikTok/Instagram/Snapchat, (II) taking into account the interaction of parents with social media in shaping un(healthy) eating patterns and (positive and negative) body image in their children and adolescents, (III) taking into account the assessment of children's mental health (e.g., depression, eating disorders), (IV) taking into account children under the age of 10, (V) research among Polish children and adolescents including the division into genders, (VI) doing more experimental research in this topic.

Finally, it is also worth pointing out that social media can be used as a resource in the prevention and treatment of obesity. A closer look at this topic seems to be particularly important due to the fact that, among adults, social media is not only a very important source of information about lifestyle, but also a source of social support when people attempting to lose weight (e.g., 64, 65). Interestingly, this research shows that this online support is even greater than that they receive from their family and friends in the non-virtual world (64). Therefore, it would be interesting to check whether we recognize a similar effect in children and adolescents. Moreover, as is commonly known, many materials available on social media are not prepared on the basis of reliable and credible sources of information (e.g., 22, 66, 67). However, by increasing preventive activity in social media and using modern solutions related to social media (including the use of hashtag signs), we can have a greater impact on the health awareness of children and adolescents around the world, including fighting myths about obesity and patients who have been subject to stigmatization. Moreover, it seems clear that the topic of social media and their relationship with body image and eating patterns should be obligatorily addressed by psychologists and nutritionists during obesity therapy, thanks to which we can correct patients' attitudes in this regard and increase knowledge and raise awareness among their caregivers.

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AM, KC-B, and JM conceived the study and performed literature search. All authors were involved in writing the manuscript and had final approval of the submitted and published versions.

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References

1. Birch L, Savage JS, Ventura A. Influences on the development of children's eating behaviours: from infancy to adolescence. *Can J Diet Pract Res.* (2007) 68:s1–56.

2. World Health Organization. *Obesity and Overweight.* (2021). Available online at: https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight (accessed July 13, 2022)

3. 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) 19:1–336. doi: 10.3310/hta19430

4. Llewellyn A, Simmonds M, Owen CG, Woolacott N. Childhood obesity as a predictor of morbidity in adulthood: a systematic review and meta-analysis. *Obes Rev.* (2016) 17:56–67. doi: 10.1111/obr.12316

5. Dattani S, Ritchie H, Roser M. *Mental Health. Eating Disorders*. (2021). Available online at:https://ourworldindata.org/mental-health (accessed July 13, 2022).

6. Ritchie H, Roser M. *Obesity*. (2017). Available online at: https://ourworldindata.org/obesity (accessed July 13, 2022).

7. Scapaticci S, Neri CR, Marseglia GL, Staiano A, Chiarelli F, Verduci E. The impact of the COVID-19 pandemic on lifestyle behaviors in children and adolescents: an international overview. *Ital J Pediatr.* (2022) 48:22. doi: 10.1186/s13052-022-01211-y

8. Taquet M, Geddes JR, Luciano S, Harrison PJ. Incidence and outcomes of eating disorders during the COVID-19 pandemic. *Br J Psychiatry.* (2021) 220:1–3. doi: 10.1192/bjp.2021.105

9. World Health Organization. *World Obesity Day 2022 – Accelerating Action to Stop Obesity*. (2022). Available online at: https://www.who.int/news/item/04-03-2022-world-obesity-day-2022-accelerating-action-to-stop-obesity#:~{}:text=More%20than%201%20billion%20people,This%20number%20is%20still%20increasing (accessed July 13, 2022)

10. Valenzise M, D'Amico F, Cucinotta U, Lugarà C, Zirilli G, Zema A, et al. The lockdown effects on a pediatric obese population in the COVID-19 era. *Ital J Pediatr.* (2021) 47:209. doi: 10.1186/s13052-021-01142-0

11. Zipfel S, Schmidt U, Giel KE. The hidden burden of eating disorders during the COVID-19 pandemic. *Lancet Psychiatry.* (2022) 9:9–11. doi: 10.1016/S2215-0366(21)00435-1

12. Savage JS, Fisher JO, Birch LL. Parental influence on eating behavior: conception to adolescence. *J Law Med Ethics.* (2007) 35:22–34. doi: 10.1111/j.1748-720X.2007.00111.x

13. Patrick H, Nicklas TA. A review of family and social determinants of children's eating patterns and diet quality. *J Am Coll Nutr.* (2005) 24:83–92. doi: 10.1080/07315724.2005.10719448

14. Hayman LL. The Dietary Intervention Study in Children (DISC): progress and prospects for primary prevention. *Prog Cardiovasc Nurs.* (2003) 18:4–5. doi: 10.1111/j.0889-7204.2003.01781.x

15. Trevino SD, Kelly NR, Budd EL, Giuliani NR. Parent gender affects the influence of parent emotional eating and feeding practices on child emotional eating. *Front Psychol.* (2021) 12:654237. doi: 10.3389/fpsyg.2021.654237

16. Scaglioni S, De Cosmi V, Ciappolino V, Parazzini F, Brambilla P, Agostoni C. Factors influencing children's eating behaviours. *Nutrients.* (2018) 10:706. doi: 10.3390/nu10060706

17. Yee AZ, Lwin MO, Ho SS. The influence of parental practices on child promotive and preventive food consumption behaviors: a systematic review and meta-analysis. *Int J Behav Nutr Phys Act.* (2017) 14:47. doi: 10.1186/s12966-017-0501-3

18. Kearney MS, Levine PB. Role models, mentors, and media influences. *Fut Child.* (2020) 30:83–106.

19. Khajeheian D, Colabi AM, Ahmad Kharman Shah NB, Bt Wan Mohamed Radzi CWJ, Jenatabadi HS. Effect of social media on child obesity: application of structural equation modeling with the taguchi method. *Int J Environ Res Public Health.* (2018) 15:1343. doi: 10.3390/ijerph15071343

20. Dilon C. Tiktok influences on teenagers and young adults students: the common usages of the application Tiktok. *ASRJETS.* (2020) 68:132–42.

21. Norc Center for Public Affairs Research. *Instagram and Snapchat are Most Popular Social Networks for Teens; Black Teens are Most Active on Social Media, Messaging Apps*. (2017). Available online at: https://policycommons.net/artifacts/2141928/instagram-and-snapchat-are-most-popular-social-networks-for-teens-black-teens-are-most-active-on-social-media/2897226/ on 13 Jul 2022 (accessed July 13, 2022).

22. Li C, Ademiluyi A, Ge Y, Park A. Using social media to understand web-based social factors concerning obesity: systematic review. *JMIR Public Health Surveill.* (2022) 8:e25552. doi: 10.2196/25552

23. Leppänen MH, Lehtimäki AV, Roos E, Viljakainen H. Body mass index, physical activity, and body image in adolescents. *Children.* (2022) 9:202. doi: 10.3390/children9020202

24. Zandian M, Bergh C, Ioakimidis I, Esfandiari M, Shield J, Lightman S, et al. Control of body weight by eating behavior in children. *Front Pediatr.* (2015) 3:89. doi: 10.3389/fped.2015.00089

25. Marks DF. Homeostatic theory of obesity. *Health Psychol Open.* (2015) 2:2055102915590692. doi: 10.1177/2055102915590692

26. Czepczor-Bernat K, Brytek-Matera A, Matusik P. The homeostatic theory of obesity: an empirical verification of the circle of discontent with an assessment of its relationship to restrained and uncontrolled eating among children and adolescents. *Int J Environ Res Public Health.* (2020) 17:6028. doi: 10.3390/ijerph17176028

27. McLean SA, Jarman HK, Rodgers RF. How do "selfies" impact adolescents' well-being and body confidence? A narrative review. *Psychol Res Behav Manag.* (2019) 12:513–21. doi: 10.2147/PRBM.S177834

28. Thompson JK, Heinberg LJ, Altabe M, Tantleff-Dunn S. *Exacting Beauty: Theory Assessment and Treatment of Body Image Disturbance*. Washington, DC: American Psychological Association (1999). doi: 10.1037/10312-000

29. de Vries DA, Vossen HGM, van der Kolk-van der Boom P. Social media and body dissatisfaction: investigating the attenuating role of positive parent-adolescent relationships. *J Youth Adolesc.* (2019) 48:527–36. doi: 10.1007/s10964-018-0956-9

30. Rodgers RF, Wertheim EH, Paxton SJ, Tylka TL, Harriger JA. #Bopo: enhancing body image through body positive social media- evidence to date and research directions. *Body Image.* (2022) 41:367–74. doi: 10.1016/j.bodyim.2022.03.008

31. National Eating Disorders Association. *What is Body Image?*. (2016). Available online at: https://www.nationaleatingdisorders.org/body-image-0 (accessed July 13, 2022)

32. Contreras RE, Schriever SC, Pfluger PT. Physiological and epigenetic features of yoyo dieting and weight control. *Front Genet.* (2019) 10:1015. doi: 10.3389/fgene.2019.01015

33. Clark O, Lee MM, Jingree ML, O'Dwyer E, Yue Y, Marrero A, et al. Weight stigma and social media: evidence and public health solutions. *Front Nutr.* (2021) 8:739056. doi: 10.3389/fnut.2021.739056

34. Di Germanio C, Di Francesco A, Bernier M, de Cabo R. Yo-Yo dieting is better than none. *Obesity.* (2018) 26:1673. doi: 10.1002/oby.22335

35. Gruver RS, Bishop-Gilyard CT, Lieberman A, Gerdes M, Virudachalam S, Suh AW, et al. A social media peer group intervention for mothers to prevent obesity and promote healthy growth from infancy: development and pilot trial. *JMIR Res Protoc.* (2016) 5:e159. doi: 10.2196/resprot.5276

36. Swindle TM, Ward WL, Whiteside-Mansell L. Facebook: the use of social media to engage parents in a preschool obesity prevention curriculum. *J Nutr Educ Behav.* (2018) 50:4–10. doi: 10.1016/j.jneb.2017.05.344

37. Zhu J, Jiang L, Dou W, Liang L. Post, eat, change: the effects of posting food photos on consumers' dining experiences and brand evaluation. *J Interac Mark.* (2019) 46:101–12. doi: 10.1016/j.intmar.2018.10.002

38. Harter S. *The Construction of the Self. In Developmental and Sociocultural Foundations*. 2nd ed. New York, NY: Guilford Publications Inc (2012).

39. Chua THH, Chang L. (2016). Follow me and like my beautiful selfies: Singapore teenage girls' engagement in self-presentation and peer comparison on social media. *Comput Hum Behav.* (2016) 55:190–7. doi: 10.1016/j.chb.2015.09.011

40. Mascheroni G, Vincent J, Jimenez E. Girls are addicted to likes so they postsemi-naked selfies": Peer mediation, normativity and the construction of identity online. *Cyberpsychol J Psychos Res Cyberspace.* (2015) 9:5. doi: 10.5817/CP2015-1-5

41. Steinsbekk S, Wichstrøm L, Stenseng F, Nesi J, Hygen BW, Skalická V. (2021). The impact of social media use on appearance self-esteem from childhood to adolescence – A 3-wave community study. *Comput Hum Behav.* (2021) 114:106528. doi: 10.1016/j.chb.2020.106528

42. Yang H, Wang JJ, Tng GYQ, Yang S. Effects of social media and smartphone use on body esteem in female adolescents: testing a cognitive and affective model. *Children.* (2020) 7:148. doi: 10.3390/children7090148

43. de Vries DA, Peter J, de Graaf H, Nikken P. Adolescents' social network site use, peer appearance-related feedback, and body dissatisfaction: testing a mediation model. *J Youth Adolesc.* (2016) 45:211–24. doi: 10.1007/s10964-015-0266-4de

44. Coates AE, Hardman CA, Halford JCG, Christiansen P, Boyland EJ. Food and beverage cues featured in youtube videos of social media influencers popular with children: an exploratory study. *Front Psychol.* (2019) 10:2142. doi: 10.3389/fpsyg.2019.02142

45. OFCOM. *Children and Parents: Media Use and Attitudes Report*. London: Ofcom (2018).

46. Coates AE, Hardman CA, Halford JCG, Christiansen P, Boyland EJ. Social media influencer marketing and children's food intake: a randomized trial. *Pediatrics.* (2019) 143:e20182554. doi: 10.1542/peds.2018-2554

47. Coates AE, Hardman CA, Halford JCG, Christiansen P, Boyland EJ. The effect of influencer marketing of food and a "protective" advertising disclosure on children's food intake. *Pediatr Obes.* (2019) 14:e12540. doi: 10.1111/ijpo.12540

48. Coates AE, Hardman CA, Halford JCG, Christiansen P, Boyland EJ. "It's just addictive people that make addictive videos": children's understanding of and attitudes towards influencer marketing of food and beverages by youtube video bloggers. *Int J Environ Res Public Health.* (2020) 17:449. doi: 10.3390/ijerph17020449

49. Bragg M, Lutfeali S, Greene T, Osterman J, Dalton M. How food marketing on instagram shapes adolescents' food preferences: online randomized trial. *J Med Internet Res.* (2021) 23:e28689. doi: 10.2196/28689

50. Tiggemann M, Miller J. The internet and adolescent girls' weight satisfaction and drive for thinness. *Sex Roles.* (2010) 63:79–90. doi: 10.1007/s11199-010-9789-z

51. Rousseau A, Eggermont S, Frison E. The reciprocal and indirect relationships between passive Facebook use, comparison on Facebook, and adolescents' body dissatisfaction. *Comput Hum Behav.* (2017) 73:336–44. doi: 10.1016/j.chb.2017.03.056

52. Kleemans M, Daalmans S, Carbaat I, Anschütz D. Picture perfect: the direct effect of manipulated Instagram photos on body image in adolescent girls. *Media Psychol.* (2018) 21:93–110. doi: 10.1080/15213269.2016.1257392

53. Núñez-Gómez P, Larrañaga KP, Rangel C, Ortega-Mohedano F. Critical analysis of the risks in the use of the internet and social networks in childhood and adolescence. *Front Psychol.* (2021) 12:683384. doi: 10.3389/fpsyg.2021.683384

54. Simeone M, Scarpato D. Sustainable consumption: how does social media affect food choices? *J Clean Prod.* (2020) 277:124036. doi: 10.1016/j.jclepro.2020.124036

55. Packer J, Russell SJ, Siovolgyi G, McLaren K, Stansfield C, Viner RM, et al. The impact on dietary outcomes of celebrities and influencers in marketing unhealthy foods to children: a systematic review and meta-analysis. *Nutrients.* (2022) 14:434. doi: 10.3390/nu14030434

56. Lou C, Kim HK. Fancying the new rich and famous? Explicating the roles of influencer content, credibility, and parental mediation in adolescents' parasocial relationship, materialism, and purchase intentions. *Front Psychol.* (2019) 10:2567. doi: 10.3389/fpsyg.2019.02567

57. Shout Out UK [SOUK]. *Generation Z And Their Phones: Separating The Myths From The Facts*. (2020). Available online at: https://www.shoutoutuk.org/2020/09/22/generation-z-andtheir-phones-separating-the-myths-from-the-facts/ (accessed august 18, 2022).

58. Norwegian Consumer Council. *Young and Exposed to Unhealthy Food Marketing: Digital Food Marketing Using Influencers*. (2019). Available online at: https://fil.forbrukerradet.no/wp-content/uploads/2019/02/young-and-exposed-to-unhealthy-marketing-digitalfood-marketing-using-influencers-report-february-2019.pdf (accessed July 13, 2022).

59. Blinka L, Smahel D. Fourteen is fourteen and a girl is a girl: validating the identity of adolescent bloggers. *Cyberpsychol Behav.* (2009) 12:735–9. doi: 10.1089/cpb.2009.0044

60. Boyd DM, Ellison NB. Social network sites: definition, history, and scholarship. *J Comput Commun.* (2007) 13:210–30. doi: 10.1111/j.1083-6101.2007.00393.x

61. Story M, French S. Food advertising and marketing directed at children and adolescents in the US. *Int J Behav Nutr Phys Act.* (2004) 1:3. doi: 10.1186/1479-5868-1-3

62. Cinelli M, De Francisci Morales G, Galeazzi A, Quattrociocchi W, Starnini M. The echo chamber effect on social media. *Proc Natl Acad Sci USA.* (2021) 118:e2023301118. doi: 10.1073/pnas.2023301118

63. Kim Y. Eating as a transgression: Multisensorial performativity in the carnal videos of mukbang (eating shows). *Int J Cult Stud.* (2021) 24:107–22. doi: 10.1177/1367877920903435

64. Hwang KO, Ottenbacher AJ, Green AP, Cannon-Diehl MR, Richardson O, Bernstam EV, et al. Social support in an Internet weight loss community. *Int J Med Inform.* (2010) 79:5–13. doi: 10.1016/j.ijmedinf.2009.10.003

65. Erdem H, Sisik A. The reliability of bariatric surgery videos in youtube platform. *Obes Surg.* (2018) 28:712–6. doi: 10.1007/s11695-017-2911-3

66. Mejova Y. Information sources and needs in the obesity and diabetes Twitter discourse. *Proceedings of the In DH'18: 2018 International Digital Health Conference, April 23–26*. New York, NY: ACM (2018). p. 21–9.

67. Jane M, Hagger M, Foster J, Ho S, Kane R, Pal S. Effects of a weight management program delivered by social media on weight and metabolic syndrome risk factors in overweight and obese adults: a randomised controlled trial. *PLoS One.* (2017) 12:e0178326. doi: 10.1371/journal.pone.0178326

68. Marsh HW. *Self Description Questionnaire-I (SDQI): APA Psyctests*. Campbelltown, NSW: University of Western Sydney (1990).

69. Harter S. *Manual For The Self-Perception Profile For Adolescents*. Denver, CO: University of Denver (1988).

70. Wichstrøm L. Harters self-perception profile for adolescents – reliability, validity, and evaluation of the question format. *J Pers Assess.* (1995) 65(1):100–16. doi: 10.1207/s15327752jpa6501_8

71. Probst M, Vandereycken W, Van Coppenolle H, Vanderlinden J. The body attitude test for patients with an eating disorder: psychometric characteristics of a new questionnaire. *Eat Disord.* (1995) 3(2):133–44. doi: 10.1080/10640269508249156

72. Frison E, Eggermont S. Exploring the relationships between different types of Facebook use, perceived online social support, and adolescents' depressed mood. *Soc Sci Comput Rev.* (2016) 34:153–71. 0894439314567449 doi: 10.1177/

73. Buhrmester D, Furman W. *The Network of Relationships Inventory: Relationship Qualities Version*. Richardson, TX: University of Texas at Dallas (2008). doi: 10.13072/midss.387

74. Rosen L, Whaling K, Carrier L, Cheever N, Rokkum J. The media and technology usage and attitudes scale: an empirical investigation. *Comput Hum Behav.* (2013) 29:2501–11. doi: 10.1016/j.chb.2013.06.006

75. Thompson JK, van den Berg P, Roehrig M, Guarda AS, Heinberg LJ. The sociocultural attitudes towards appearance scale-3 (SATAQ-3): development and validation. *Int J Eat Disord.* (2004) 35(3):293–304. doi: 10.1002/eat.10257

76. Schaefer LM, Thompson JK. The development and validation of the physical appearance comparison scale-revised (PACS-R). *Eat Behav.* (2014) 15(2):209–17. doi: 10.1016/j.eatbeh.2014.01.001

77. Hart TA, Flora DB, Palyo SA, Fresco DM, Holle C, Heimberg RG. Development and examination of the social appearance anxiety scale. *Assessment.* (2008) 15(1):48–59. doi: 10.1177/1073191107306673

78. Mendelson BK, Mendelson MJ, White DR. Body-esteem scale for adolescents and adults. *J Pers Assess.* (2001) 76(1):90–106. doi: 10.1207/S15327752JPA7601_6

79. Stotland S, Zuroff DCA. new measure of weight locus of control: the dieting beliefs scale. *J Pers Assess.* (1990) 54(1–2):191–203. doi: 10.1080/00223891.1990.9673986

80. Cash TF. *The Multidimensional Body-Self Relations Questionnaire Users' Manual*. (1994). Available online at: http://www.body-images.com/assessments/mbsrq.html (accessed July 13, 2022).

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Calreticulin and PDIA3, two markers of endoplasmic reticulum stress, are associated with metabolic alterations and insulin resistance in pediatric obesity: A pilot study

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Our aim was to evaluate the markers of endoplasmic reticulum (ER) stress among children and adolescents with obesity in relation to metabolic alterations. Calreticulin (CALR) and PDIA3 circulating levels were assessed on 52 pediatric subjects—26 patients with obesity and 26 normal weight controls (4–18 years)—enrolled in a pilot study. Clinical and metabolic evaluations were performed (BMI-SDS, insulin, and glucose at fasting and during an oral glucose tolerance test, lipid profile, blood pressure), and metabolic syndrome was detected. PDIA3 was higher ($p < 0.02$) and CALR slightly higher in children with obesity than in controls. PDIA3 was related positively to the Tanner stages. Both PDIA3 and CALR were positively associated with insulin resistance, cholesterol, and triglycerides and the number of criteria identifying metabolic syndrome and negatively with fasting and post-challenge insulin sensitivity. Our preliminary findings suggest the existence of a link between ER stress and metabolic changes behind obesity complications even at the pediatric age. CALR and PDIA3 could be early markers of insulin resistance and dyslipidemia-related ER stress useful to stratify patients at high risk of further complications.

KEYWORDS

obesity, ER stress, pediatrics, insulin, lipids

1 Introduction

Obesity is a growing public health issue due to its prevalence and numerous associated comorbidities in children and adolescents (1–4). Inflammation, oxidative stress, lipotoxicity, and endoplasmic reticulum (ER) stress are all identified as obesity triggers for its complications (5).

Some hypotheses have been proposed in order to clarify the mechanisms leading to proinflammatory response activation of visceral adipose tissue (VAT) (6), considering the influence of several mechanisms on adipose tissue, including the hypoxia-inducible factor (HIF) activation (7, 8), the production of reactive oxygen species (ROS) (9), the increased concentration of free fatty acids (FFAs) leading to lipotoxicity, and the role of proinflammatory cytokines (6). This lays the foundation for the development of complications such as insulin resistance and type 2 diabetes mellitus (T2DM) (10).

The excess of nutrients and the increased metabolism of FFAs, characteristics of obesity, can lead to the involvement of the endoplasmic reticulum (ER), since it plays a crucial role in controlling lipid metabolism by regulating lipid synthesis, modification, and secretion (11).

When the ER is subjected to excessive workload, it enters a stress situation (ER stress); thus, to protect cellular functionality, unfolded protein response (UPR) is activated, with the aim of solving ER stress by acting on cellular metabolism, through a process of retro-translocation in the cytoplasm, the addition of ubiquitin, and demolition by the proteasome (11). However, when the protein load is excessive and the ER stress is very high, the UPR leads to the activation of cellular death mechanisms. This situation can be observed in obesity when several chronic conditions such as hyperglycemia, hyperinsulinemia, and elevation of FFA plasma levels and proinflammatory cytokines promote the activation of the cell death pathway mediated by ER stress (12). Indeed, an increase in the expression of proteins involved in cytoskeletal remodeling, inflammation, cellular senescence, oxidative stress, and ER stress can be observed particularly in those patients showing signs of metabolic decompensation, such as insulin resistance or T2DM (13–16). It seems that some of the proteins involved in the abovementioned processes may represent valid markers of "progression" of the disease and could therefore contribute to the identification of complications at an early stage. Among these, calreticulin (CALR) and PDIA3, two important proteins located in the ER and involved in cellular response to ER stress, whose increase is part of the attempt of cells to cope with some of the deleterious effects of cell stress, such as protein misfolding and ROS damage, are worthy of a more in-depth analysis (13–18) (Figure 1).

CALR was initially described as an ER protein, regulating calcium homeostasis and the folding of glycoproteins (17). Today, CALR is recognized as a chaperone with various functions; it is involved in the regulation of cell proliferation processes, phagocytosis, apoptosis, adhesion, and adaptive and innate immune responses. Acting synergistically with calnexin (CNX), another chaperone of the ER, CALR becomes part of the quality control mechanism that monitors the state of glycosylation and three-dimensional arrangement of proteins transiting the ER (17, 19).

CALR functions become fundamental to resolve the accumulation of proteins within the ER and ensure a return to cellular homeostasis. In these conditions and following the activation of the UPR, there is an increase in the synthesis of this protein, and IRE-1 (an enzyme recruited as a first response) mediates the processing and activation of XBP-1, a transcription factor that acts on the genes of several chaperones, including CALR and CNX themselves (19, 20). It is through this mechanism that an increased CALR expression is observed in tissues subjected to strong cellular stress, just like in the case of VAT, subcutaneous adipose tissue (SAT), and the liver of obese subjects, in which CALR is found in high concentrations (14, 21).

PDIA3 is among the genes whose transcription is promoted by XBP-1 (19, 20). PDIA3 is present mainly but not exclusively in the ER, where its main function is to ensure and mediate the formation of the correct intramolecular disulfide bonds in nascent proteins. Indeed, PDIA3 is a stress response protein aimed at avoiding and solving situations of cellular stress (22), and its expression is increased in these cases, especially following protein accumulation that results in ER stress and UPR (14–16, 21).

This protein has four domains, with an active site that provides its oxidation-reductive property, while the C-terminal possibly enables protein retention in the ER and could act as a nuclear localization sequence and a portion responsible for the interaction and binding with CALR and CNX, contributing to normal folding and proper protein glycosylation interaction (22). PDIA3 is also involved in various functions in other cellular compartments, with studies supporting its involvement in signal transduction from the cell surface, the regulation of nuclear processes such as DNA repair, and the promotion of phagocytosis and autophagy (18).

Based on the above, we aimed to assess if an association among CALR, PDIA3, and metabolic risk factors in pediatric obesity exists since ER stress proteins could be precocious markers of metabolic impairment and no data exist in this age on these molecules.

2 Materials and methods

2.1 Population and clinical parameters

Fifty-two subjects aged between 4 and 18 were enrolled. Twenty-six case subjects were recruited in a pilot study from patients attending the Division of Pediatric Endocrinology of

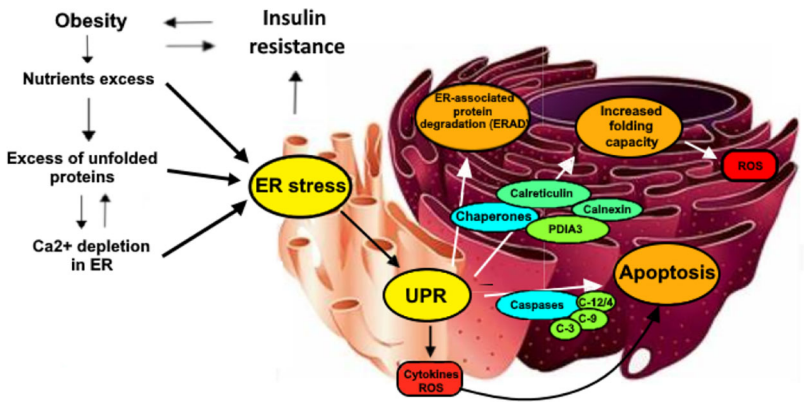

FIGURE 1
Some of the main mechanisms underlying the relationship between obesity, insulin resistance al URP-related chaperones activity. Nutrients intake, when is higher than cellular needs, causes an overproduction of unfolded proteins that require to be processed by ER. In these situations, ER activates multiple mechanisms to reduce stress sources. First, Insulin resistance is provoked by the cell to reduce the insulin-mediated protein transcription, lately being ad additional factor for the development of obesity. Secondly, ER activates UPR, increasing the activity of several enzymes such as caspases that are responsible of apoptotic processes in addition to ROS and inflammatory cytokines production. URP has also the role of increasing protein degradation (ERAD) and contemporarily of boosting folding capacity through oxidative protein folding, which is partially performed by the activation of the PDIA3/Calreticulin/Calnexin chaperones complex..

our hospital. As a control group, 26 normal weight age-matched subjects were enrolled. The control group consisted of outpatients of our hospital, joining the project "The prevention game begins as a child" (CE 95/12). The inclusion criteria for patients were the presence of obesity according to the International Obesity Task Force curves (IOTF) (23). For both groups, we excluded subjects with cardiorespiratory diseases, endocrine diseases (hypo- or hyperthyroidism, growth hormone deficiency, type 1 diabetes, adrenal insufficiency, hypercortisolism), chronic diseases, and genetic causes of obesity, as well as those under prescription drugs that could alter glucose and/or lipid metabolism.

All patients were subjected to clinical assessment, including Tanner criteria for pubertal development, defining puberty at stages 2–5 (24). Weight, height, and waist circumference were measured by the medical staff, and BMI and BMI *Z*-score were calculated. Patients and controls were categorized into weight classes according to the 2012 IOTF (23).

Blood pressure was measured and stratified by gender and age, as suggested by the National High Blood Pressure Education Program (NHBPEP) Working Group of the American Academy of Pediatrics (AAP) (25).

2.2 Biochemical evaluation

Blood samples were collected at fasting in the morning. We evaluated glucose and insulin, total cholesterol, HDL, triglycerides, PDIA3, and CALR. All patients were subjected to an oral glucose tolerance test (OGTT), evaluating glucose and insulin levels every 30 min for 120 min (26). From the OGTT, the indexes regarding insulin resistance (HOMA-IR) and insulin sensitivity (QUICKI and Matsuda index, ISI) as well as the insulinogenic index were calculated. The formulas are reported in our previous papers (27, 28).

LDL levels were obtained from Friedwald's formula, calculated for subjects with triglyceride levels below 150 mg/dl.

Patients were also evaluated for the presence of cardiovascular risk factors considered in the definition of metabolic syndrome using the limit values of the modified criteria of the National Cholesterol Education Program (NCEP) and the Adult Treatment Panel (ATP) III (>90 percentile triglycerides by sex and age; <10 percentile HDL by sex and age; IFG/IGT) (26).

2.3 CALR and PDIA3 analysis

CALR or PDIA3 concentration was evaluated in plasma samples using the Human CALR or PDIA3 ELISA kit [Elabsicence (Houston, Texas, USA)]. Briefly, a 100-µl dilution consisting of standard, blank, and sample was placed into the appropriate wells (in duplicate). Samples were incubated for 90 min at 37°C. Liquid was decanted from each well, and 100 µl of Biotinylated Detection Ab working solution was added to each well immediately. The samples were incubated for 1 h at 37°C. The liquid was decanted and 350 µl of wash buffer was added to each well and removed after 1 min (repeated three

times). Then, 100 µl of HRP conjugate working solution was added to each well, and the plate was incubated for 30 min at 37° C. The liquid was removed and each was washed three times as reported above. Then, 90 µl of substrate reagent was added to each well, and the samples were incubated for about 15 min at 37°C (protected from light). Fifty microliters of stop solution was added to each well, and optical density (OD value) was evaluated with a microplate reader set to 450 nm [Spark multimode microplate reader; Tecan (Seestrasse, Switzerland)].

CALR or PDIA3 concentration in each sample was calculated through the standard curve, obtained by serial dilution of a CALR or PDIA3 stock solution of 10 ng/ml. The sensitivity of the kit was estimated to be 0.10 ng/ml, the analytical range was between 0.16 and 10 ng/ml, and the coefficient of variation was <10%.

2.4 Statistical analysis

Due to the small sample of patients, a non-parametric approach was used. Data were expressed as median (IQR) or percentage (%). Differences between the two groups were analyzed using the Mann–Whitney *U* test. Correlation analysis was performed using Spearman's *r*.

Statistical significance was set at p <0.05. The statistical analysis was performed using SPSS for Windows V.26.0 (SPSS Inc., Chicago, IL, USA).

3 Results

This pilot study was carried out involving 52 subjects (26 healthy subjects and 26 with obesity) matched for sex and pubertal stage. Six of them were excluded because of incomplete clinical information. The clinical data are reported in Table 1. Of the children with obesity, 65.2% were obese and 34.8% were morbidly obese.

Children with obesity had a higher level of PDIA3 (0.212, 0.187–0.465 ng/ml, $p < 0.05$) than normal weight patients (0.188, 0.167–0.222 ng/ml), corrected by sex and age. PDIA3 significance was lost when correcting also for BMI and pubertal stage. CALR levels, on the other hand, were comparable between normal weight patients (0.230, 0.206–0.273 ng/ml) and those with obesity (0.233, 0.145–0.422 ng/ml).

3.1 Metabolic features

Glucose and insulin levels at 0 min ($p < 0.01$) and 120 min ($p < 0.001$) and HOMA-IR ($p < 0.001$) were higher, while HDL ($p < 0.001$), QUICKI ($p < 0.001$), and ISI ($p < 0.001$) were lower in obese subjects compared to normal weight subjects. The data are reported in Table 2.

3.2 Correlation analysis

3.2.1 CALR

CALR was positively correlated with systolic ($r = 0.329$; $p < 0.05$) and diastolic blood pressure ($r = 0.374$; $p < 0.05$), acanthosis score ($r = 0.684$; $p < 0.001$), non-HDL cholesterol ($r = 0.347$; $p < 0.03$), triglycerides ($r = 0.488$; $p < 0.003$), number of criteria identifying metabolic syndrome ($r = 0.705$; $p < 0.001$), fasting insulin ($r = 0.415$; $p < 0.005$), HOMA-IR ($r = 0.368$; $p < 0.005$), and insulinogenic index ($r = 0.292$; $p < 0.05$). CALR was, instead, negatively correlated with HDL ($r = -0.390$; $p < 0.01$), QUICKI ($r = -0.368$; $p < 0.01$), and ISI ($r = -0.421$; $p < 0.02$) (Table 3).

TABLE 1 Clinical characteristics of the population.

Variable		Obese	Normal weight	*p*-value
Gender	Male	12 (52.2%)	5 (40.3%)	
	Female	11 (47.8%)	18 (59.7%)	
Puberty	Prepubertal	8 (34.8%)	4 (17.4%)	
	Pubertal	15 (65.2%)	19 (82.6%)	
Age (years)		11.4 (8.8; 13.4)	12.6 (9.9; 13.2)	
Height (cm)		155.3 (137.8; 161)	152.5 (145; 163.1)	
Weight (kg)		69.1 (52; 81.5)	40 (34; 48.2)	$p < 0.001$
BMI (kg/m^2)		28.4 (26.1; 32.4)	17 (15.7; 18.2)	$p < 0.001$
BMI *Z*-score (kg/m^2)		2.03 (1.9; 2.5)	−0.94 (−1.32; −0.40)	$p < 0.001$
Waist (cm)		92 (78; 99)	69 (63.5; 72.5)	$p < 0.001$
Waist/heigh (cm)		0.58 (0.54; 0.61)	0.38 (0.01; 0.44)	$p < 0.001$

Data are expressed as median (IQR) or percentage (%).
BMI, body mass index.

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TABLE 2 Metabolic and biochemical characteristics of obese and normal weight individuals.

	Obese (N = 23)	Normal weight (N = 23)	p-value
Systolic blood pressure (mmHg)	123 (112; 132)	120 (110; 125)	
Systolic blood pressure (mmHg)	80 (70; 86)	80 (70; 80)	
Total cholesterol (mg/dl)	141 (121; 158)	142 (127; 164.5)	
HDL (mg/dl)	40 (36; 54)	60 (49; 65)	$p < 0.001$
LDL (mg/dl)	82 (63; 100)	73 (61; 96)	
Non-HDL-c (mg/dl)	97 (78; 120)	84 (72; 105.5)	
TG (mg/dl)	63 (38; 91)	44 (40.5; 54)	$p < 0.04$
AST (mg/dl)	24 (19; 28)	25 (23.5; 29)	
ALT (mg/dl)	21 (18; 30)	19 (17.5; 21.5)	
Glucose at 0 min (mg/dl)	89 (84; 95)	85 (75; 89.5)	$p < 0.01$
Glucose at 120 min (mg/dl)	117 (106; 123)	91 (86.5; 108.5)	$p < 0.001$
Insulin at 0 min (UI/ml)	16.8 (9.3; 24.6)	9.5 (6.3; 11.3)	$p < 0.001$
Insulin at 120 min (UI/ml)	70.7 (43.7; 135.5)	24.5 (17.4; 37.4)	$p < 0.001$
HOMA-IR	3.7 (2.2; 5.8)	2.0 (1.3; 2.3)	$p < 0.001$
QUICKI	0.31 (0.29; 0.34)	0.34 (0.33; 0.36)	$p < 0.001$
ISI	3.42 (1.87; 4.41)	6.14 (5.53; 8.32)	$p < 0.001$
Insulinogenic index	1.99 (0.89; 2.93)	1.32 (1.19; 1.91)	
CALR (ng/ml)	0.233 (0.145; 0.422)	0.230 (0.206; 0.273)	
PDIA3 (ng/ml)	0.212 (0.187; 0.465)	0.188 (0.167; 0.222)	$p < 0.05$

Descriptive characteristics are expressed as median (IQR).
HDL, high-density lipoprotein; LDL, low-density lipoprotein; TG, triglyceride; AST, aspartate aminotransferase; ALT, alanine aminotransferase; HOMA-IR, insulin resistance index; QUICKI, quantitative insulin-sensitivity check index; ISI, insulin sensitivity index.

3.2.2 PDIA3

As shown in Table 3, PDIA3 levels were positively correlated with weight ($r = 0.325$; $p < 0.001$), BMI ($r = 0.303$; $p < 0.03$) and BMI Z-score ($r = 0.378$, $p < 0.008$), waist circumference ($r = 0.292$; $p < 0.05$), Tanner stages ($r = 0.328$; $p < 0.05$), acanthosis score ($r = 0.342$; $p < 0.05$), number of criteria identifying metabolic syndrome ($r = 0.485$; $p < 0.001$), LDL ($r = 0.305$; $p < 0.05$), non-HDL cholesterol ($r = 0.438$; $p < 0.008$), triglycerides ($r = 0.664$; $p < 0.001$), fasting insulin ($r = 0.296$; $p < 0.02$), and HOMA-IR ($r = 0.273$; $p < 0.06$). PDIA3 was found to be only inversely correlated with HDL ($r = -0.433$; $p < 0.008$) and QUICKI ($r = -0.283$; $p < 0.06$).

4 Discussion

Obesity is associated with several comorbidities, all related to the establishment of chronic inflammation, oxidative stress, lipotoxicity, and endoplasmic reticulum (ER) stress. Our preliminary data suggest that CALR and PDIA3, two key molecules of ER stress, could be related to insulin resistance and altered lipid profile in pediatric obesity.

One of the mechanisms involved in the development of obesity complications is ER stress, triggered by an overload of lipids and proteins among other mechanisms. ER stress markers have been found elevated in the visceral adipose tissue of adult patients with obesity, and it seems that these proteins could be markers also in their circulating form. Two of the most studied molecules are CALR and PDIA3, starting from their relation with cancer (18, 29–32). The markers of ER stress are poorly investigated especially at a young age. Therefore, we aimed to explore their modulation in children and adolescents with obesity.

First, preliminary data from our study showed higher circulating levels of PDIA3, but not CALR, in obese pediatric subjects compared to those with normal weight. The correlation of PDIA3 with BMI, weight, and waist circumference can be explained by the release of ER stress markers in the adipose tissue, increasing simultaneously with the increased number of adipocytes. PDIA3 is also an important regulator of postnatal bone and muscle growth, particularly during the pubertal growth spurt (33), possibly explaining also the correlation of PDIA3 with the Tanner stage. However, because our population is mainly in the mid of puberty, the strength of the association could be influenced by this age and puberty distribution.

Second, in our cohort, we observed associations between circulating levels of both markers and cardiometabolic risk factors. PDIA3 was directly correlated with a high concentration of LDL cholesterol, and both PDIA3 and CALR were directly correlated with non-HDL cholesterol and triglycerides and inversely correlated with low HDL cholesterol

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TABLE 3 Significant correlation of CALR and PDIA3.

	CALR	Spearman's *r*	*p*-value	PDIA3	Spearman's *r*	*p*-value
Weight (kg)				+	0.325	0.001
BMI (kg/m^2)				+	0.303	0.03
BMI *Z*-score (kg/m^2)				+	0.378	0.008
Waist circumference (cm)				+	0.292	0.05
Diastolic blood pressure (mmHg)	+	0.329	0.05			
Systolic blood pressure (mmHg)	+	0.317	0.05			
Tanner stages				+	0.328	0.05
Acanthosis	+	0.684	0.001	+	0.342	0.05
LDL (mg/dl)				+	0.305	0.05
HDL (mg/dl)	−	−0.390	0.01	−	−0.433	0.008
Non-HDL cholesterol (mg/dl)	+	0.347	0.03	+	0.438	0.008
Triglycerides (mg/dL)	+	0.488	0.003	+	0.664	0.001
Number of criteria identifying metabolic syndrome	+	0.705	0.001	+	0.485	0.001
Fasting insulin (UI/ml)	+	0.415	0.005	+	0.296	0.02
HOMA-IR	+	0.368	0.01	+	0.273	0.06
Insulinogenic index	+	0.292	0.05			
QUICKI	−	−0.368	0.01	−	−0.283	0.06
ISI	−	−0.421	0.02			

BMI, body mass index; HOMA-IR, insulin resistance index; QUICKI, quantitative insulin-sensitivity check index; ISI, insulin sensitivity index; +, positive correlation; −, negative correlation.

levels. Our results are in line with recent literature, reporting an increased lipid turnover as a cause of ER malfunction (12, 34). Furthermore, both molecules were involved in cholesterol assembly and turnover (35, 36).

PDIA3 and CALR were also correlated directly with insulin levels and HOMA-IR and inversely with ISI and QUICKI, surrogate indexes of insulin sensitivity. Also, both markers were found to be related with acanthosis nigricans, which is one of the clinical manifestations of insulin resistance, due to the presence of keratinocytes and fibroblasts able to respond to insulin through the expression of IGF1-R (37, 38). Literature studies confirm how ER stress is directly involved in the development of insulin resistance during obesity such as in a pediatric population (39–41). Our results strengthen these findings and underline how insulin resistance and ER stress are strictly related in a precocious stage of metabolic derangement. This was also confirmed by the direct association of CALR and PDIA3 with the number of metabolic alterations typical of metabolic syndrome. Our findings were in line with similar results in adults with metabolic syndrome (42). Furthermore, CALR expression increased during an OGTT in healthy adults, suggesting that it could be a sensible ER stress marker of chronic and acute dysglycemia in both children and adults (42). Furthermore, a decreased CALR expression in adipose tissue after Roux-en-Y gastric bypass in obese adult patients has been shown, suggesting a link with the decrease in chronic inflammation and metabolic derangement (43).

This study has the limitation of having a small number of samples analyzed, which is secondary to the experimental difficulties faced in the development of the dosage of PDIA3 and CALR since their range for pediatric age is not yet known. Because of the lack of data in the literature that correspond to the results of our study, analysis for the expansion of the sample is needed to confirm our preliminary findings.

In conclusion, this pilot study aimed to establish the association between ER stress and metabolic complications in obese children and adolescents. The results suggest the existence of an important link between ER stress and metabolic changes behind obesity complications even in pediatric age. CALR and PDIA3 could be early markers of insulin resistance and dyslipidemia-related ER stress, being useful to stratify patients at high risk of further complications over time.

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

Ethical review and approval was not required for the study on human participants in accordance with the local legislation and institutional requirements. Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

Author contributions

Conceptualization: FP, SB, and MC. Methodology: VA, CP, MG, FP, SB, and MC. Software: DS. Validation: RR and MaC. Formal analysis: VA, DS, and FP. Investigation: VA, RR, VM, and ST. Data curation: MC. Writing—original draft preparation: VA, FGC, and FP. Writing—review and editing: SB and MC. Supervision: SB, MaC, and FP. Funding acquisition: FP, SB, and MC. All authors discussed the results and contributed to the final manuscript. All authors affirm that the present work is original, has not been published previously, and has not been submitted elsewhere for consideration of print or electronic publication. Each person listed as an author participated in the work in a substantive manner, in accordance with ICMJE authorship guidelines, and is prepared to take public responsibility for it.

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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. Chooi YC, Ding C, Magkos F. The epidemiology of obesity. *Metabolism* (2019) 92:6–10. doi: 10.1016/j.metabol.2018.09.005

2. Bray GA. Medical consequences of obesity. *J Clin Endocrinol Metab* (2004) 89:2583–9. doi: 10.1210/jc.2004-0535

3. Abarca-Gómez L, Abdeen ZA, Hamid ZA, Abu-Rmeileh NM, Acosta-Cazares B, Acuin C, et al. 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) 390:2627–42. doi: 10.1016/S0140-6736(17)32129-3

4. Afshin A, Forouzanfar MH, Reitsma MB, Sur P, Estep K, Lee A, et al. Health effects of overweight and obesity in 195 countries over 25 years. *N Engl J Med* (2017) 377:13–27. doi: 10.1056/nejmoa1614362

5. Longo M, Zatterale F, Naderi J, Parrillo L, Formisano P, Raciti GA, et al. Adipose tissue dysfunction as determinant of obesity-associated metabolic complications. *Int J Mol Sci* (2019) 20:2358. doi: 10.3390/ijms20092358

6. Blüher M. Adipose tissue inflammation: a cause or consequence of obesity-related insulin resistance?. *Clin Sci* (2016) 130:1603–14. doi: 10.1042/CS20160005

7. Trayhurn P, Wang B, Wood IS. Hypoxia in adipose tissue: A basis for the dysregulation of tissue function in obesity?. *British J Nutr* (2008) 100:227–35. doi: 10.1017/S0007114508971282

8. Singh P, Hoffmann M, Wolk R, Shamsuzzaman ASM, Somers VK. Leptin induces c-reactive protein expression in vascular endothelial cells. *Arterioscler Thromb Vasc Biol* (2007) 27(9):e302-7. doi: 10.1161/ATVBAHA.107.148353

9. Savini I, Catani MV, Evangelista D, Gasperi V, Avigliano L. Obesity-associated oxidative stress: Strategies finalized to improve redox state. *Int J Mol Sci* (2013) 14:10497–538. doi: 10.3390/ijms140510497

10. Lyons CL, Kennedy EB, Roche HM. Metabolic inflammation-differential modulation by dietary constituents. *Nutrients* (2016) 8:247. doi: 10.3390/nu8050247

11. Schröder M. Endoplasmic reticulum stress responses. *Cell Mol Life Sci* (2008) 65:862–94. doi: 10.1007/s00018-007-7383-5

12. Hotamisligil GS. Endoplasmic reticulum stress and the inflammatory basis of metabolic disease. *Cell* (2010) 140:900–17. doi: 10.1016/j.cell.2010.02.034

13. Boden G, Duan X, Homko C, Molina EJ, Song W, Perez O, et al. Increase in endoplasmic reticulum stress–related proteins and genes in adipose tissue of obese, insulin-resistant individuals. *Diabetes* (2008) 57:2438–44. doi: 10.2337/db08-0604

14. Caira S, Iannelli A, Sciarrillo R, Picariello G, Renzone G, Scaloni A, et al. Differential representation of liver proteins in obese human subjects suggests novel biomarkers and promising targets for drug development in obesity. *J Enzyme Inhib. Med Chem* (2017) 32:672–82. doi: 10.1080/14756366.2017.1292262

15. Murri M, Insenser M, Bernal-Lopez MR, Perez-Martinez P, Escobar-Morreale HF, Tinahones FJ. Proteomic analysis of visceral adipose tissue in pre-obese patients with type 2 diabetes. *Mol Cell Endocrinol* (2013) 376:99–106. doi: 10.1016/j.mce.2013.06.010

16. Alfadda AA, Benabdelkamel H, Masood A, Moustafa A, Sallam R, Bassas A, et al. Proteomic analysis of mature adipocytes from obese patients in relation to aging. *Exp Gerontol.* (2013) 48:1196–203. doi: 10.1016/j.exger.2013.07.008

17. Trombetta ES, Helenius A. Conformational requirements for glycoprotein reglucosylation in the endoplasmic reticulum. *J Cell Biol* (2000) 148:1123–30. doi: 10.1083/jcb.148.6.1123

18. Turano C, Gaucci E, Grillo C, Chichiarelli S. PDIA3/GRP58: A protein with multiple functions. *Cell Mol Biol Lett* (2011) 16:539–63. doi: 10.2478/s11658-011-0022-z

19. Schröder M, Kaufman RJ. The mammalian unfolded protein response. *Annu Rev Biochem* (2005) 74:739–89. doi: 10.1146/annurev.biochem.73.011303.074134

20. Calfon M, Zeng H, Urano F, Till JH, Hubbard SR, Harding HP, et al. IRE1 couples endoplasmic reticulum load to secretory capacity by processing the XBP-1 mRNA. *Nature* (2002) 415:92–6. doi: 10.1038/415092a

21. Boden G, Lebed B, Schatz M, Homko C, Lemieux S. Effects of acute changes of plasma free fatty acids on intra-myocellular fat content and insulin resistance in healthy subjects. *Diabetes* (2001) 50:1612–7. doi: 10.2337/diabetes.50.7.1612

22. Silvennoinen L, Myllyharju J, Ruoppolo M, Orrù S, Caterino M, Kivirikko KI, et al. Identification and characterization of structural domains of human PDIA3: Association with calreticulin requires several domains. *J Biol Chem* (2004) 279:13607–15. doi: 10.1074/jbc.M313054200

23. Cole TJ, Lobstein T. Extended international (IOTF) body mass index cut-offs for thinness, overweight and obesity. *Pediatr Obes* (2012) 7:284–94. doi: 10.1111/j.2047-6310.2012.00064.x

24. Tanner JM. *Growth at adolescence. with a general consideration of the effects and environmental factors upon growth and maturation from birth to maturity; 2d ed*. Oxford: Blackwell Scientific Publications (1962).

25. Flynn JT, Kaelber DC, Baker-Smith CM, Blowey D, Carroll AE, Daniels SR, et al. Clinical practice guideline for screening and management of high blood pressure in children and adolescents. *Pediatrics* (2017) 140(3):e20171904. doi: 10.1542/peds.2017-1904

26. Cruz ML, Goran MI. The metabolic syndrome in children and adolescents. *Curr Diab. Rep* (2004) 4:53–62. doi: 10.1007/s11892-004-0012-x

27. Ricotti R, Solito A, Zani EM, Caputo M, Genoni G, Barone-Adesi F, et al. The relationship between cortisol and IGF-I influences metabolic alteration in pediatric overweight and obesity. *Eur J Endocrinol* (2020) 182:255–64. doi: 10.1530/EJE-19-0792

28. Genoni G, Menegon V, Monzani A, Archero F, Tagliaferri F, Mancioppi V, et al. Healthy lifestyle intervention and weight loss improve cardiovascular dysfunction in children with obesity. *Nutrients* (2021) 13:130. doi: 10.3390/nu13041301

29. Raghavan M, Wijeyesakere SJ, Peters LR, Del Cid N. Calreticulin in the immune system: Ins and outs. *Trends Immunol* (2013) 34:13–21. doi: 10.1016/j.it.2012.08.002

30. Charonis AS, Michalak M, Groenendyk J, Agellon LB. Endoplasmic reticulum in health and disease: The 12th interna-tional calreticulin workshop, Delphi, Greece. *J Cell Mol Med* (2017) 21:3141–9. doi: 10.1111/jcmm.13413

31. Kageyama S, Isono T, Iwaki H, Wakabayashi Y, Okada Y, Kontani K, et al. Identification by proteomic analysis of calreticulin as a marker for bladder cancer and evaluation of the diagnostic accuracy of its detection in urine. *Clin Chem* (2004) 50:857–66. doi: 10.1373/clinchem.2003.027425

32. Leys CM, Nomura S, LaFleur BJ, Ferrone S, Kaminishi M, Montgomery E, et al. Expression and prognostic significance of prothymosin-α and PDIA3 in human gastric cancer. *Surgery* (2007) 141:41–50. doi: 10.1016/j.surg.2006.05.009

33. Linz A, Knieper Y, Gronau T, Hansen U, Aszodi A, Garbi N, et al. ER stress during the pubertal growth spurt results in impaired long-bone growth in chondrocyte-specific PDIA3 knockout mice. *J Bone Miner. Res* (2015) 30:1481–93. doi: 10.1002/jbmr.2484

34. Bravo R, Parra V, Gatica D, Rodriguez AE, Torrealba N. Endoplasmic reticulum and the unfolded protein response. *Int Rev Cell Mol Biol* (2013) 301:215–90. doi: 10.1016/B978-0-12-407704-1.00005-1

35. Linnik KM, Herscovitz H. Multiple molecular chaperones interact with apolipoprotein b during its maturation: The network of endoplasmic reticulum-resident chaperones (ERp72, GRP94, calreticulin, and BiP) interacts with apolipoprotein b regardless of its lipidation state. *J Biol Chem* (1998) 273:21368–73. doi: 10.1074/jbc.273.33.21368

36. Stillemark P, Borén J, Andersson M, Larsson T, Rustaeus S, Karlsson KA, et al. The assembly and secretion of apolipoprotein b-48-containing very low-density lipoproteins in McA-RH7777 cells. *J Biol Chem* (2000) 275:10506–13. doi: 10.1074/jbc.275.14.10506

37. Maffeis C. *L'obesità del bambino: Aspetti clinici e fisiopatologici*. Torino: Centro Scientifico editore (2009).

38. Ahrens W, Pigeot I, Pohlabeln H, De Henauw S, Lissner L, Molnár D, et al. Prevalence of overweight and obesity in European children below the age of 10. *Int J Obes* (2014) 38:S99–S107. doi: 10.1038/ijo.2014.140

39. Özcan U, Cao Q, Yilmaz E, Lee AH, Iwakoshi NN, Özdelen E, et al. Endoplasmic reticulum stress links obesity, insulin action, and type 2 diabetes. *Sci (80-.)* (2004) 306:457–61. doi: 10.1126/science.1103160

40. Czaja MJ. JNK regulation of hepatic manifestations of the metabolic syndrome. *Trends Endocrinol Metab* (2010) 21:707–13. doi: 10.1016/j.tem.2010.08.010

41. Jalali S, Aghasi M, Yeganeh B, Mesaeli N. Calreticulin regulates insulin receptor expression and its downstream PI3 ki-nase/Akt signalling pathway. *Biochim Biophys Acta - Mol Cell Res* (2008) 1783:2344–51. doi: 10.1016/j.bbamcr.2008.08.014

42. Sage AT, Holtby-Ottenhof S, Shi Y, Damjanovic S, Sharma AM, Werstuck GH. Metabolic syndrome and acute hyperglycemia are associated with endoplasmic reticulum stress in human mononuclear cells. *Obesity* (2012) 20:748–55. doi: 10.1038/oby.2011.144

43. Ferraz-Bannitz R, Welendorf CR, Coelho PO, Salgado WJr, Nonino CB, Beraldo RA, et al. Bariatric surgery can acutely modulate ER-stress and inflammation on subcutaneous adipose tissue in non-diabetic patients with obesity. *Diabetol Metab Syndr* (2021) 13(1):19. doi: 10.1186/s13098-021-00623-w

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Obesity in adolescents may be associated with limitations in daily activities and an increased level of anxiety in patients and their parents – preliminary results of a pilot study

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Obesity is a chronic disease, that in adolescents may lead to serious consequences affecting somatic and mental health. This study aimed to assess the prevalence of depressive symptoms and anxiety in adolescents with obesity and their parents. The relationships between depressive and anxiety symptoms and the somatic consequences of obesity were also analyzed.

Material and Methods: 19 patients with obesity (BMI Z-SCORE 2.1-5.5), at the age 16-17, and their parents answered validated questionnaires (Children's Depression Inventory 2, The State-Trait Anxiety Inventory), and a survey assessing everyday functioning.

Results: There were no significant differences in the occurrence of symptoms of depression in children and their parents: for the overall scale score of T-score (p=0.331), for the emotional problems (p=0.281) subscale, and the functional problems (p=0.147) subscale. The comparison of the results between boys and girls revealed no significant differences. A significantly higher level of anxiety was found in parents of children who gained weight in the year preceding the study (p = 0.046), and both in children and parents of children with metabolic-associated fatty liver disease – MAFLD (p=0.022 and p=0.007). According to adolescents, obesity affects the most leisure activities.

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Conclusion: Obesity, like any chronic disease, can have a significant impact on the emotional state of children and adolescents as well as the possibility of realizing interests and spending free time. Much more important than depressive disorders are anxiety disorders concerning both patients and their parents.

KEYWORDS

obesity, depression, anxiety, adolescents, children, MAFLD

Introduction

Obesity is a chronic disease that often occurs from early childhood, leading to the development of a cascade of other health problems (1). According to data published in 2017, the percentage of children with obesity increased eightfold between the years 1975 and 2016, and the total number in 2016 was 41 million (2). The negative impact of obesity on the somatic development of children and adolescents has been documented in many studies. It has been proven that even in the youngest age groups there is a risk of developing metabolic complications, such as arterial hypertension, liver steatosis, obstructive sleep apnea, vitamin D deficiency, and progressive disorders of glucose, lipid, and uric acid metabolism (3–5). Less is known about the association of obesity in the developmental age with mental health outcomes. The prevalence and future risk of anxiety, symptoms of depression, and everyday functioning in the family and school environment remain unclear (6).

The studies published so far and their meta-analyzes revealed that children and adolescents with obesity are at risk of the development of depressive symptoms or even overt depression, that may persist into adulthood (6, 7). That risk was higher for female patients (6, 8, 9). In some detailed studies, the authors also drew attention to disorders of the emotional state of adolescents with obesity other than depression, including anxiety. It has been shown that unrecognized and untreated emotional problems can lead to the development of mental health deterioration and eating disorders (10). To date, the relationship between the occurrence of metabolic complications of obesity and the occurrence of anxiety disorders, depressive symptoms, and everyday functioning in adolescents has not been analyzed. Some single studies have been conducted in adults, but the results are inconclusive (11, 12). Even less attention has been paid to the emotional problems of parents of obese children. Although the child's disease, very often in the family context of obesity, can have a significant impact on parents' emotional functioning (13). Parents of obese children seem to be at particular risk of developing anxiety and depression symptoms because, in addition to the treatment of a child with a chronic, difficult disease, they are often obese themselves.

This preliminary study aimed to assess the prevalence of depressive and anxiety symptoms in adolescents with obesity and their parents. The impact of medical factors, such as concomitant metabolic disorders, complications of obesity, and results of the body weight reduction on the symptoms of depression and anxiety, and selected aspects of everyday functioning were also analyzed.

Material and methods

The preliminary study included 19 children (74% female), at the mean age of 16 (range 15-17 years) with obesity and their parents (14) The study used validated questionnaires (Children's Depression Inventory 2, CDI2, and The State-Trait Anxiety Inventory, STAI, both in the Polish language version). These questionnaires were completed separately by the parent (n=19) and the patient. The method used to measure depression has a high-reliability coefficient. Cronbach's α-coefficient measuring the reliability of the children's and parents' results for the tested sample and subscales ranged between 0.71 and 0.79, which proves that the scale is sufficiently reliable for the tested sample (15). The STAI questionnaire consists of two independent parts, each containing 20 statements. The part STAI X1 and STAI X2. X1 can evaluate anxiety considered as the current emotional state. This part of the questionnaire is a very sensitive tool. It enables one to trace the dynamics of anxiety even in short time intervals. X2, concerns anxiety understood as a personality trait (16). High levels of anxiety X1 and X2 defined has been defined for the value of the state scale: 8-10, and a high level of depressive symptoms was defined for the ten scale values above 64. Additionally, the participants of the study were asked about subjective feelings about the influence of the disease on key aspects of their lives: relationship with peers, school performance, way of spending free time, hobbies, and relations with siblings. The adolescents referred to each issue in the range from 1 to 5, where 1.2 meant that the disease did not affect the examined issues, and 4.5 - the disease's impact on the examined factors. The middle answer was neutral and it was difficult to express the sentence. Such an answer was classified more as a positive

answer, which in the case of this study means that the disease does not influence the examined issues. The study also analyzed the medical data regarding the occurrence of biochemical disorders typical of the metabolic syndrome (disorders of glucose metabolism, elevated triglycerides ≥ 1.7 mmol/L, and low HDL <1.0 mmol/L) (3), vitamin D deficiency, presence of biochemical and ultrasonographical presence of metabolic-associated fatty liver disease (MAFLD). The results of obesity treatment, defined as the change in body weight in the 12 months preceding the study, were also analyzed.

Statistics

All statistical calculations were performed using the statistical package StatSoft Inc. STATISTICA (data analysis software system) version 13.3. The Shapiro-Wilk W test was used to check whether the quantitative variable was derived from a population with a normal distribution. The significance of differences between two groups in the model of unrelated variables was tested by Student's t-test or Mann-Whitney U-test (when the assumptions of the parametric test were not met). The significance of differences between two groups in the model of related variables was tested by Student's t-test or Wilcoxon's test (when the assumptions of the parametric test were not met). Correlation analysis was used to determine the relationship, strength, and direction between ordinal variables by calculating the Spearman correlation coefficient. Fisher's exact test was used to examine correlations for dichotomous characteristics, and Pearson's chi-squared test for independence was used for categorical variables. The decision on the hypotheses to be verified was made by assuming a significance level of 0.05.

The odds ratio was used to assess the chance of an event occurring in one group to the chance of it occurring in another group.

Results

The coefficient of age variation was 5.5% (less than 10%, not statistically significant). The mean BMI Z-SCORE was 3.5 (range 2.1-5.5) (14). In the study group, 74% of patients were already treated with the combined lifestyle intervention, including constant supervision by a dietician. Nevertheless, approximately 74% of participants in the year preceding the study increased their body weight. Odds ratio analysis showed, however, that dietician care increased the chance of losing weight almost six times compared to being left without such care. The most common concomitant problem was vitamin D deficiency (n=15; 79%). Impaired fasting glucose was diagnosed in one patient (5%), impaired glucose tolerance in 4 (21%), elevated triglycerides in 4 (21%), low HDL-cholesterol in 3 (16%), MAFLD features in 6 (32%) (see Table 1). The analysis of the responses showed that obesity has not been shown to affect relationships with peers. According to teenagers, obesity affects the most spending free time and pursuing a hobby. However, even for these variables, half of the respondents believe that the disease is not an obstacle in terms of leisure activities. Interestingly, almost 16% of respondents believe that obesity has a negative impact on school performance and the same number of young people believe that it limits the possibility of developing their interests (Table 2). There were no significant differences in the occurrence of symptoms of depression in children and their parents: for the overall scale score of T-score (p=0.331), for the emotional problems (p=0.281) subscale, and for the functional problems (p=0.147) subscale. The comparison of the results between boys and girls revealed no significant differences. There were also no significant differences in the level of depression (emotional problems, functional problems, and overall score CDI) between adolescents who increased their weight and those who lost weight (p = 0.103, p = 0.171; p = 0.085). The concomitant complications of obesity also were insignificant in terms of the occurrence of depression symptoms: impaired glucose tolerance (p=0.898, p=0.909; p=0.866); dyslipidemia (p=0.664, p=0.704; p=0.770), MAFLD (p=0.823, p=0.911; p=0.804), and vitamin D deficiency (p=0.921, p=0.203; p=0.471). The relationship between the results of the depression symptoms and BMI Z-SCORE, HOMA-IR, and vitamin D measured with the Spearman correlation was statically insignificant (p> 0.05). The results obtained from the STAI test allowed for the selection of participants with a high level of anxiety (a permanent trait - X2) and a high level of "transient" anxiety (X1). There were no significant differences in the distribution of sten X1 values between children and their parents (T=33.5; p=0.402). Significant differences were found in the distribution of the sten X2 values (T=17.0; p=0.008). There were no differences in anxiety (X1, X2 children, X2 parents) levels between boys and girls (p=0.546;

TABLE 1 Characteristic of the study group.

	mean (range) [SD]	number (%) of te results out of range
Fasting glucose [mmol/L]	4.8 (3.6-5.8) [0.52]	1 (5%)
Glucose 120' (oral glucose tolerance test) [mmol/L]	6.5 (4.8-9.4) [1.4]	4 (21%)
Triglicerydes [mmol/L]	1.27 (0.58-2.81) [0.57]	4 (21%)
HDL cholesterol [mmol/L]	1.2 (0.75-1.45) [0.22]	3 (16%)
25(OH)D [ng/mL]	19.95 (7.4-38) [9.7]	15 (79%)

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TABLE 2 The influence of the disease on key aspects of adolescent's lives.

variabile	Realtionship with peers		School performance		Way of spending free time		Hobby		Relations with siblings	
Answer (score 1-5)	n_i	ω_i [%]	n_i	ω_i [%]	n_i	ω_i [%]	n_i	ω_i [%]	n_i	ω_i [%]
1	7	3ω6.8	6	31.6	4	21.1	4	21.1	11	57.9
2	2	10.5	8	42.1	6	35.0	7	36.8	6	31.6
3	8	42.1	2	10.5	5	21.7	5	26.3	1	5.3
4	2	10.5	2	10.5	4	21.7	2	10.5	1	5.3
5			1	5.3			1	5.3		

p=0.738; p=0.212, respectively). However, a significantly higher level of X2 anxiety was found in parents of children who gained weight in the year preceding the study (p = 0.046). The chance of a high level of anxiety as a feature of X2 for parents of children who have increased their body weight was more than two times higher than for that who decreased weight. Statistically significant higher level of X1 and X2 parents was noticed in patients with MAFLD (p=0.022 and p=0.007). Additionally, the odds ratio analysis showed a more than ten times higher risk of a high level of anxiety (X2 children) in patients with MAFLD). The other concomitant complications of obesity were not significant for parameters of anxiety (X1, X2 children, X2 parents): impaired glucose tolerance (p=0.254, p=0.158; p=0.378); dyslipidemia (p=0.432, p=0.216; p=0.215), and vitamin D deficiency (p=0.803, p=0.794; p=0.582). The relationships between the anxiety levels X1, X2 children, X2 parents, BMI Z-SCORE, HOMA-IR, and vitamin D levels measured with the Spearman monotonic correlation coefficient were statically insignificant (p> 0.05).

Discussion

Obesity itself can be a stressful state due to the high prevalence of weight stigma. This interaction between excessive body weight and emotional state may be bi-directional leading to a vicious cycle of stress and obesity (8, 17). In this study, we showed that according to adolescents with obesity, the disease most significantly limits the ability to pursue interests, hobbies, and leisure activities. Among obese adolescents diminished physical activity is a common problem, which itself leads to weight gain. It is worth noticing, that if patients with perceive obesity as a significant obstacle to physical activity, then putting pressure on them can cause frustration and even increase emotional problems. A detailed analysis of that problem was carried out by Jodkowska et al., obtaining similar results and concluding that in overcoming the barriers to physical activity in obese adolescents, one should aim to comprehensively reduce body weight and support health-oriented motivation (18). Contrary to most of the studies published so far, in this study we have not shown a significant predominance in the incidence of depressive symptoms in girls (6, 8, 9). Overall, in the present study, rates of depression in both adolescents and their parents were relatively low. Although it is commonly repeated, that obesity is related to depression, that thesis is not fully supported by research findings. Most of the studies relating depression to obesity in adolescents have generated inconsistent results. Among overweight children drawn from specialist clinics, approximately 24% are estimated to present symptoms of depression (19). However, it is worth noting that children and adolescents drawn from specialist clinics are not representative of children in the community and may overestimate the risk (20). In an excellent community-based study including over 6,000 participants, Wardle et al. showed that in adolescents, regardless of gender, socioeconomic status, or ethnicity, reports of depressive symptoms are not significantly higher in obese than normal-weight groups (21). In the population of adolescents with obesity, the problem of anxiety seems to be much more important. That issue should be analyzed in obese teenagers themselves and their parents. Although the results of the present study obtained in adolescents themselves did not show any significant correlation between treatment outcomes as measured by weight change and the occurrence of anxiety or depressive disorders, in the case of their parents, anxiety was more severe in those whose children did not lose weight. No similar, clear relationship was observed for depressive symptoms. However, the influence of a small number of participants on this result cannot be ruled out. It certainly requires further research. The problem of perceiving one's own child's obesity and the concerns about it has been analyzed by BeLue et al. Caregivers in this study consistently reported concern for their overweight children's mental health and problem behavior (13). Obesity in adolescents may have consequences for caregivers and families, even if it is not a significant problem for the patients themselves. On this basis, one could speculate that reducing the child's body weight, may be beneficial to reduce anxiety in parents. And as a further consequence, perhaps may improve the attitude towards treatment. In this context, the next observation from the present study concerning the role of a dietitian in the therapeutic process is of particular importance. Regular surveillance of the patient significantly increases the chance of losing weight. Similar results were obtained in the previous study (22). Therefore, it can be speculated that dietary habits may indirectly influence the psychological effects of treatment. This preliminary observation

should become the subject of further research. Another important new observation in this study is the association of the severity of anxiety with the occurrence of organ complications of obesity, i.e. MAFLD. The risk of a high level of anxiety was 10 times higher in patients with MAFLD, than in those without that disorder. So far, the research has clearly distinguished between the somatic and psychological effects of obesity in adolescents. This analysis is the first attempt to combine both aspects. At present, we do not have sufficient evidence to show the direction of the relationship between a higher level of anxiety and the occurrence of MAFLD. Nevertheless, this observation is not entirely new. Some recent studies on the incidence of emotional, depressive, and anxiety disorders in adult patients with the liver disease found similar associations of incompletely explained origins (23). Some researchers have suggested the influence of chronic stress on the development of liver damage in people with obesity. According to this theory, as in type 2 diabetes and cardiovascular disease, stress involves both behavioral and biological responses, which activate the hypothalamic –pituitary – adrenal axis, resulting in elevated levels of cortisol and

pro-inflammatory biomarkers that could be involved in the development of MAFLD (24, 25). Also, pro-inflammatory pathways, immune dysregulation, and systemic or multi-organ inflammation are considered to mediate pathophysiological interaction between certain mental health disorders (25, 26). Finally, there are also established relationships between obesity and mental health that may be connected to changes in gut microbiota, resulting in such comorbidity. Recently, this mechanism of MAFLD development has also been suggested. According to this theory, perhaps both metabolic disorders and symptoms of anxiety and depression have a similar source in people with obesity (27). Undoubtedly further studies are needed to unravel the mechanism(s) of MAFLD and their psychological manifestations.

Although the results of the study turned out to be innovative and surprising, the work itself has significant limitations. The first one is the size of the group. As far as it is a pilot study with a small number of participants, unequal gender distribution, substantial variation in BMI Z-SCORE, and metabolic characteristics, the results need to be confirmed in a larger group of participants. Another is conducting a study in a clinic, on patients admitted to the hospital. A greater value of the results could be obtained by conducting the study in a larger community and comparing the results in people with obesity and normal body weight.

Conclusions

Obesity, like any chronic disease, can have a significant impact on the emotional state of children and adolescents as well as the possibility of realizing interests and spending free time. Much more important than depressive disorders are anxiety disorders concerning both patients and their parents.

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 study was approved by the local Jagiellonian University Ethics Committee (No 1072.6120.34.2021, date MAR-17-2021.). Written informed consent to participate in this study was provided by the participants' legal guardian/ next of kin.

Author contributions

MW MM-S, DG, MP, AS, JS: conceived the project, DG, MP, AS, MW, DD, AK-K: collected data, DG, MW, MM-S, EM: wrote the original draft. DG, MW, MM-S, AK-K were responsible for conducting the systematic review. DG, EM performed the statistical analysis. JS: participated in revising the work for important intellectual content. All authors contributed to the article and approved the submitted version.

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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.

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References

1. Majcher A, Czerwonogrodzka-Senczyna A, Kądziela K, Rumińska M, Pyrżak B. Development of obesity from childhood to adolescents. *Pediatr Endocrinol Diabetes Metab* (2021) 27(2):70–5. doi: 10.5114/pedm.2021.105297

2. Abarca-Gómez L, Abdeen ZA, Hamid ZA, Abu-Rmeileh NM, Acosta-Cazares B, Acuin C, et al. 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

3. Zimmet P, Alberti KGM, Kaufman F, Tajima N, Silink M, Arslanian S, et al. The metabolic syndrome in children and adolescents ? an IDF consensus report. *Pediatr Diabetes* (2007) 8(5):299–306. doi: 10.1111/j.1399-5448.2007.00271.x

4. Calderón JM, Álvarez-Pitti J, Cuenca I, Ponce F, Redon P. Development of a minimally invasive screening tool to identify obese pediatric population at risk of obstructive sleep Apnea/Hypopnea syndrome. *Bioengineering* (2020) 7(4):131. doi: 10.3390/bioengineering7040131

5. Rumińska M, Majcher A, Pyrżak B, Czerwonogrodzka-Senczyna A, Brzewski M, Demkow U. Cardiovascular risk factors in obese children and adolescents. *Adv Exp Med Biol* (2015) 878:39–47. doi: 10.1007/5584_2015_168

6. Sutaria S, Devakumar D, SS Y, Das S, Saxena S. Is obesity associated with depression in children? systematic review and meta-analysis. *Arch Dis Child* (2019) 104(1):64–74. doi: 10.1136/archdischild-2017-314608

7. Fox CK, Gross AC, Rudser KD, Foy AMH, Kelly AS. Depression, anxiety, and severity of obesity in adolescents. *Clin Pediatr (Phila)* (2016) 55(12):1120–5. doi: 10.1177/0009922815615825

8. Mannan M, Mamun A, Doi S, Clavarino A. Prospective associations between depression and obesity for adolescent males and females- a systematic review and meta-analysis of longitudinal studies. *PloS One* (2016) 11(6):e0157240. doi: 10.1371/journal.pone.0157240

9. Boutelle KN, Hannan P, Fulkerson JA, Crow SJ, Stice E. Obesity as a prospective predictor of depression in adolescent females. *Health Psychol* (2010) 29(3):293–8. doi: 10.1037/a0018645

10. Sagar R, Gupta T. Psychological aspects of obesity in children and adolescents. *Indian J Pediatr* (2018) 85(7):554–9. doi: 10.1007/s12098-017-2539-2

11. Phillips CM, Perry IJ. Depressive symptoms, anxiety and well-being among metabolic health obese subtypes. *Psychoneuroendocrinology* (2015) 62:47–53. doi: 10.1016/j.psyneuen.2015.07.168

12. Jokela M, Hamer M, Singh-Manoux A, Batty GD, Kivimäki M. Association of metabolically healthy obesity with depressive symptoms: pooled analysis of eight studies. *Mol Psychiatry* (2014) 19(8):910–4. doi: 10.1038/mp.2013.162

13. BeLue R, Francis LA , Colaco B. Mental health problems and overweight in a nationally representative sample of adolescents: Effects of race and ethnicity. *Pediatrics* (2009) 123(2):697–702. doi: 10.1542/peds.2008-0687

14. Kułaga Z, Litwin M, Tkaczyk M, Palczewska I, Zajączkowska M, Zwolińska D, et al. Polish 2010 growth references for school-aged children and adolescents. *Eur J Pediatr* (2011) 170(5):599–609. doi: 10.1007/s00431-010-1329-x

15. Hair JF, Babin BJ, Anderson RE, Black WC. Multivariate data analysis. 8th ed. Cengage Learning (2019).

16. Wrzesniewski K. *Inwentarz stanu i cechy leku STAI: polska adaptacja STAI: podrecznikk*. Pracownia Testów Psychologicznych Polskiego Towarzystwa Psychologicznego (2011).

17. Tomiyama AJ. Stress and obesity. *Annu Rev Psychol* (2019) 70(1):703–18. doi: 10.1146/annurev-psych-010418-102936

18. Jodkowska M, Oblacińska A, Nałęcz H, Mazur J. Perceived barriers for physical activity in overweight and obese adolescents and their association with health motivation. *Dev Period Med* (2017) 21(3):248–58. doi: 10.34763/devperiodmed.20172103.248258

19. Britz B, Siegfried W, Ziegler A, Lamertz C, Herpertz-Dahlmann B, Remschmidt H, et al. Rates of psychiatric disorders in a clinical study group of adolescents with extreme obesity and in obese adolescents ascertained *via* a population based study. *Int J Obes* (2000) 24(12):1707–14. doi: 10.1038/sj.ijo.0801449

20. Fitzgibbon ML, MR S, Kirschenbaum DS. Obese people who seek treatment have different characteristics than those who do not seek treatment. *Health Psychol* (1993) 12(5):342–5. doi: 10.1037/0278-6133.12.5.342

21. Wardle J, Williamson S, Johnson F, Edwards C. Depression in adolescent obesity: cultural moderators of the association between obesity and depressive symptoms. *Int J Obes* (2006) 30(4):634–43. doi: 10.1038/sj.ijo.0803142

22. Kozioł-Kozakowska A, Wójcik M, Furtak A, Januś D, Starzyk JB. A comparison of the impact of two methods of nutrition-behavioral intervention on selected auxological and biochemical parameters in obese prepubertal children–crossover preliminary study. *Int J Environ Res Public Health* (2019) 16(16):2841. doi: 10.3390/ijerph16162841

23. Colognesi M, Gabbia D, de Martin S. Depression and cognitive impairment–extrahepatic manifestations of NAFLD and NASH. *Biomedicines* (2020) 8(7):229. doi: 10.3390/biomedicines8070229

24. Kang D, Zhao D, Ryu S, Guallar E, Cho J, Lazo M, et al. Perceived stress and non-alcoholic fatty liver disease in apparently healthy men and women. *Sci Rep* (2020) 10(1):38. doi: 10.1038/s41598-019-57036-z

25. Shea S, Lionis C, Kite C, Atkinson L, SS C, HS R, et al. Non-alcoholic fatty liver disease (NAFLD) and potential links to depression, anxiety, and chronic stress. *Biomedicines* (2021) 9(11):1697. doi: 10.3390/biomedicines9111697

26. Tsigos C, Kyrou I, Kassi E, Chrousos GP. *Stress: Endocrine physiology and pathophysiology*. Feingold KR, et al, editors Cengage Learning (2020).

27. Agustí A, García-Pardo MP, López-Almela I, Campillo I, Maes M, Romaní-Pérez M, et al. Interplay between the gut-brain axis, obesity and cognitive function. *Front Neurosci* (2018) 12. doi: 10.3389/fnins.2018.00155

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Nonalcoholic fatty liver disease in children with obesity– observations from one clinical centre in the Western Pomerania region

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Nonalcoholic fatty liver disease (NAFLD) is a growing health problem in the pediatric population, due to the constantly increasing percentage of children with obesity. The objective of the study was to assess the occurrence of NAFLD based on ultrasound (USG) analysis and the use of alanine aminotransferase (ALT) in children with overweight and obesity depending on glucose tolerance. Medical records of 228 consecutive patients aged 2-18 years with overweight and obesity were reviewed retrospectively. Based on the oral glucose tolerance test children were divided into groups according to the severity of carbohydrate metabolism disorders. ALT, lipid parameters and insulin sensitivity indices HOMA, Matsuda and Quicki were analyzed. NAFLD was diagnosed (based on the USG) in 51 patients (23.61%) - the incidence in the impaired glucose tolerance (IGT) and type 2 diabetes (T2DM) group was significantly higher when compared to ones without glucose intolerance. Because of extreme values of metabolic parameters in TDM2 children being outliers, they were not considered in the statistical analysis of the study. 22 (11.58%) patients had elevated ALT values, of which 12 (54.55%) had hepatic steatosis features on ultrasound. 72.73% (n=32) patients with fatty liver features on USG had ALT values considered normal with cut-off point 42 U/l accepted in this study. Almost every fourth obese child in the study group presents features of fatty liver in ultrasound examination. Although ultrasound is not recommended by North American *Society* For Pediatric Gastroenterology, Hepatology &Nutrition(NASPGHAN) for the diagnosis of NAFLD in children, it allows identifying a high percentage of children with features of fatty liver. This percentage increases significantly in children with glucose intolerance.

KEYWORDS

NAFLD, fatty liver, obesity, ALT, glucose intolerance

Introduction

The increasing number of overweight and children with obesity causes nonalcoholic fatty liver disease (NAFLD) to be one of the most common pathologies of this organ (1). Its occurrence can lead to hepatic fibrosis and cirrhosis (2). In the last decades NAFLD has been the leading cause for liver transplants in adult patients. Due to the severity of the issue, in 2017 NASPGHAN has published recommendations regarding the diagnosis and treatment of nonalcoholic fatty liver disease in children (3).

Pathogenesis of nonalcoholic fatty liver disease's development is complex (4). The emphasis is put both on genetic, as well as metabolic factors (insulin resistance, hyperglycemia, dyslipidemia, cytokine overproduction, oxidative stress). Accumulation of triglycerides in the liver is caused by three factors: lipolysis in the adipose tissue, *de novo* synthesis caused by excess carbohydrate consumption in the diet and dietary lipids. In the setting of overnutrition and obesity, hepatic fatty acid (FA) metabolism is altered, commonly leading to the accumulation of triglycerides within hepatocytes, and to a clinical condition known as NAFLD (5). It has been proven, that the association between dietary fructose and NAFLD is stronger than with glucose. In the recent years the consumption of fructose has significantly increased in the form of high-fructose corn syrup (HFCS) and mixtures of fructose and glucose monosaccharides (6, 7). Incorrect eating habits and excess carbohydrate consumption lead to insulin resistance, which is proven to be one of the important risk factors for NAFLD (2, 8). Bugianesi et al. suggest, that an increased flow of free fatty acids to the liver caused by peripheral insulin resistance causes its steatosis (9). In the adipose tissue of patients with diabetes, disruptions in translocation and activation of glucose transporters have been found. Their lower number and incorrect function have also been found in obese patients. The European Association for the Study of the Liver-The European Association for the Study of Diabetes and The European Association for the Study of Obesity (EASL-EASD-EASO) recommendations include active seeking for impaired glucose tolerance in patients with NAFLD, as well as for NAFLD in type 2 diabetes patients (10).

Alanine and aspartate aminotranferases (AST) are markers of hepatic cell damage, ALT more so than AST. ALT isawidely used marker of hepatic steatosis. The upper limit for children in different age groups allowing to assess the risk of NAFLD is debatable (11, 12). Sensitivity of ALT alone is lower than when using imaging studies and histopathology. NASPHGAN recommendations suggest alanine aminotransferase assessment as a screening test. The laboratory norm has been established using ALT levels in healthy children in accordance to age, and set as 22 U/l for girls and 26 U/l for boys (3). According to those guidelines, norms set by local laboratories should not be taken into account. ALT levels exceeding the norm twofold for longer than 3 months should encourage further investigation, where NAFLD and other chronic inflammatory liver diseases. Studies evaluating the usefulness of aspartate transaminase and γ-glutamyltransferase (GGTP) as a diagnostic marker of NAFLD in children have not yet been performed. However, some authors emphasize, that elevated levels of ALT, AST and GGTP are associated with worse histopathology of the liver biopsies. Elevated AST and GGTP levels with normal ALT may suggest liver diseases other than NAFLD (13). Ultrasonography alone, without checking markers in the serum, is not considered a screening test.

NASPHAN recommendations authors emphasize, that the screening test, i.e. ALT level assessment, should be performed from the age of 9-11 in children with obesity with body mass index(BMI) $\geq 85^{th}$ percentile and additional risk factors, such as central obesity, insulin resistance, prediabetes and diabetes, dyslipidemia, sleep apnea or family history of NAFLD. Due to the fact, that an increasing fraction of the pediatric population is at risk of being overweight or obese because of changes in eating habits and insufficient physical activity, early diagnosis of NAFLD is necessary. The search for the best markers and risk factors for the disease lies in the interests of many investigators. In the recent years an increasing number of publications concerning this problem has appeared.

Our hypothesis is that simple, non-invasive methods of routine investigation may be useful and sufficient to identify severe metabolic complications in children with obesity. The aim of the study is to assess the incidence of hepatic steatosis by the way of routine investigations in overweight children, including abdominal ultrasound (USG) and alanine aminotransferase (ALT) levels, in correlation to different grades of carbohydrate metabolism disorders.

Patients and methods

Medical records of 228 consecutive patients in the age of 2-18 years with overweight and obesity were retrospectively analyzed. The study was approved by the Pomeranian Medical University Bioethical Committee on April 14 2004, decision number BN-001/67/04. None of the children were previously diagnosed because of overweight. Neither glucose metabolism disorders nor clinical signs of diabetes have been observed in the children before. All those children were hospitalised in one day clinic of the referential Pediatric Endocrinology Clinic for the Western Pomerania region, inhabited by almost 2 million people, over the course of four years.

Obesity and overweight were defined with BMI SDS. To establish norms for Polish population for body weight, height ant BMI percentile charts by Palczewska and Niedźwiecka were used (14). Children with secondary causes of obesity (genetic, endocrine disorders e.g. hypothyroidism, Cushing's disease etc.,

iatrogenic obesity) as well as children with active hepatotropic virus infections, autoimmune liver diseases and congenital metabolic defects were excluded from the study. Children with secondary causes of obesity (genetic, endocrine disorders e.g. hypothyroidism, Cushing's disease etc., iatrogenic obesity) were excluded from the study. The primary aim of the study was to assess the risk of obesity-related metabolic complications. Anthropometric parameters were measured (body height, body weight, BMI) as well as puberty stage according to Tanner's scale. Based on the results of oral glucose tolerance test (OGTT) the study group was divided in four groups: with no carbohydrate metabolism disorders, with impaired fasting glucose (IFG), with impaired glucose tolerance (IGT) and with type 2 diabetes (T2DM). In OGTT both glucose and insulin levels were analyzed in 0 and 120 minute of the test. In the whole study group an abdominal ultrasound with hepatic steatosis assessment was performed, as well as laboratory studies consisting of hepatic enzymes levels, i.e. alanine and aspartate aminotransferase (ALT and AST), lipid profile, i.e. cholesterol level, high-density lipoprotein (HDL), low-density lipoprotein (LDL) and triglycerides (TG). Insulin resistance indices have been calculated: homeostasis model assessment of insulin resistance (HOMA-IR) (15, 16), Matsuda index, quantitative insulin sensitivity check index (Quicki). To assess the hepatocyte function ALT and AST serum levels were measured, using kinetic method. The norm has been set as below 39 U/l for AST and below 42 U/l for ALT. Total cholesterol levels were assessed using the enzymatic-colorimetric method with cholesterol esterase. TG levels were assessed using the enzymatic-colorimetric method with phosphoglycerol esterase. HDL levels were measured using direct method with enzymes modified with poliethylene glycol. LDL levels were measured using direct method with catalase. All lipid profile analyses were performed on Roche Cobas C501 device. Total cholesterol levels below 170 mg/dl were accepted as normal, 170-199 mg/dl as high normal and above 199 mg/dl as high. HDL cholesterol levels higher than 35 mg/dl have been considered as normal, while the values equal and below 35 mg/dl as low. LDL levels below 110 mg/dl were considered as normal, levels of 110-129 mg/dl as high normal and above 129 mg/dl as high. Reference values for triglyceride levels were different for girls and boys. For girls values lower than 130 mg/dl were considered normal, whereas in boys it was values below 120 mg/dl. Values 130-150 mg/dl and 120-150 mg/dl were considered high normal for girls and boys respectively, while values above 150 mg/dl were considered high for both sexes.

To measure insulin resistance indirect methods were used. Based on glucose and insulin serum levels measured during OGTT insulin resistance and insulin sensitivity indices were calculated:

Quantitative Insulin Sensitivity Check Index (Quicki) = 1/ log fasting insulin (μIU/mL) + log fasting glucose (mg/dl).Values below < 0.34 suggest insulin resistance.

Homeostasis Model Assessment of Insulin Resistance (HOMA-IR) = fasting glucose (mg/dl) x fasting insulin (μIU/mL)/405. It is assumed, that values >2.5 for I and V stage of the puberty according to Tanner score suggest insulin resistance, and for II-IV stage of the puberty values >3.5 suggest insulin resistance.

Matsuda Index = 100000/√fasting insulin (μIU/mL) x fasting glucose (mg/dl) x mean OGTT glucose (mg/dl) x mean OGTT insulin (μIU/mL).Values<7.3 suggest insulin resistance.

Abdominal ultrasound was performed in all subjects. The examinations were performed on Philips Pure Wave CX50 using convex C5-1 for older children and C8-5 for younger children. All examinations were performed on the same device by two experienced ultrasonographists. Hepatic steatosis was graded using US-FLI semi-quantitative method (17), however the subjects were not divided according to the grade of steatosis, only the presence or absence thereof was considered.

All children with obesity hospitalised in the Clinic took part in dietary schooling and had access to psychologic support. Children's eating habits were analyzed using authors' proprietary nutrition questionnaire. Eating habits were rated based on 24-hour eating history, repeated 3 times (3 days of the week chosen by the child's mother, including 1 day off school). To assess the portion sizes, "Album of photographs of food and dishes" issued by National Food and Nutrition Institute (NFNI) was used (18). Children below the age of 13 filled the questionnaire with their mothers, while children above 13 did it alone or with parental help. To calculate the caloric intake and basic macronutrients of the children's diet, a computer program DIETA.5 created by NFNI was used. Caloric demand was calculated for every patient individually in accordance to NFNI nutrition norms, considering ideal body weight, age, sex and physical activity level (19)]. Suggested macronutrient ratio of total daily caloric intake, i.e. proteins (12.5%), digestible carbohydrates (57.5%), simple carbohydrates (15%), overall fats (30%) and saturated fats (10%) was used to create dietary recommendations for subjects in this study. Because of extreme values of metabolic parameters in TDM2 children being outliers, they were not considered in the statistical analysis of the study.

Methods of statistical analysis

Based on the gathered results a database was created in Microsoft Excel. Further statistical analysis was performed in the Stata 11.0 program. The Kolmogorov-Smirnov test was used to verify the normality of distributions of continuous variables. Those variables were then described as mean values and standard deviations. Statistical differences between two groups were then checked with t-Student test and Mann-Whitney test. Non-continuous variables were described as quantity and frequency. The analysis of relationships between non-continuous variables was carried out using Pearson's χ^2 test or Fisher's test. To analyze

the relationships between continuous variables Pearson's correlation was used. The results were described as probability – p, correlation coefficient – r, and regression lines. To estimate the risk of a pathology depending on its risk factors a model of logit regression was used. The results were described by calculating the odds ratio (OR) along with 95% confidence interval. Probability was calculated in this model using Pearson's χ^2 test or two-sided Fisher's exact test. If OR>1, the analyzed factor increases the risk. If OR<1, the analyzed factor decreases the risk. For the whole model R values (multidimensional correlation coefficient) and p (probability) were also calculated. For all of the performed analyses, statistical significance was considered at p values ≤ 0.05.

Results

The study group consisted of 228 children hospitalised because of overweight and obesity. Within this group there were 118 girls (51.75%) and 110 boys (48.25%). The average age was 10.4±3.6 years.122 of the children were prepubertal (53.51%).

The average body weight of the examined children was 62.7 ± 23.9 kg, while the average body weight SDS was 2.98 ± 0.91. The average BMI body mass index value was 27.83 ± 4.7 kg/m^2, the average BMI SDS 2.98 ± 0.74. In 83.77% (n = 191) subjects there were no changes in glucose tolerance, 3.07% (n = 7) subjects were diagnosed with IFG, 10.97% (n = 25) with IGT and 2.19% (n = 5) with T2DM (Table 1).All children with T2DM were newly diagnosed upon admission.

Because of extreme values of metabolic parameters in TDM2 children (as seen in Table 1), they were excluded as outliers from further statistical analysis. Based on ultrasound imaging, hepatic steatosis was diagnosed in 48 patients (22.54%), of which there were 25 (22.52%) girls and 23 (22.55%) boys. The average age of subjects with steatosis was 10.71 ± 3.63 years. In the study group 11.58% (n=22) of the subjects had incorrect ALT values, whereas in the group with hepatic steatosis in USG examination ALT levels were elevated in 27.27% (n=12) of the subjects. In the whole study group the percentage of boys with elevated ALT was 14.89% (n=14), the percentage of girls was 8.33% (n=8). In 54.55% (n=12) of children with elevated alanine aminotransferase levels hepatic steatosis was found in ultrasound.

In the group of children without glucose intolerance (GI), the sonographic features of fatty liver were found in 19.57% (n = 36) of the subjects. In the group with IGT, this percentage

TABLE 1 Subjects characteristic by glucose tolerance status.

	Allsubjects	Obesity*	IFG	IGT	T2DM	p	R
n (%)	228	191 (83.77)	7 (3.07)	25 (10.96)	5 (2.19)		
Sex – Male, n (%)	110 (48.25)	99 (51.83)	5 (71.43)	11 (44)	2 (40.00)		
Sex – Female, n (%)	118 (51.75)	92 (48.17)	2 (28.57)	14 (56.00)	3 (60.00)		
Age (years)	10.4 ± 3.6	10.4 ± 3.58	12.33 ± 2.78	10.88 ± 3.14	17.34 ± 0.53	<0.0001	0.31
Weight (kg)	62.7 ± 23.9	60.10 ± 23.15	77.96 ± 16.28	71.73 ± 25.53	94.26 ± 13.78	0.0005	0.28
Weight SD	2.98 ± 0.91	2.96 ± 0.90	2.8 ± 0.92	3.23 ± 1.00	2.71 ± 0.99	0.4505	0.11
Height (m)	1.47 ± 0.19	1.45 ± 0.19	1.63 ± 0.10	1.54 ± 0.19	1.67 ± 0.06	0.0016	0.26
Height SD	1.33 ± 1.23	1.30 ± 1.18	1.77 ± 1.22	1.67 ± 1.48	-0.07 ±1.29	0.0238	0.20
BMI (kg/m^2)	27.83 ± 4.70	27.38 ± 4.34	29.92 ± 8.94	29.43 ± 4.74	33.84 ± 4.77	0.0023	0.25
BMI SD	2.98 ± 0.74	2.98 ± 0.74	2.65 ± 1.01	3.08 ± 0.72	2.82 ± 0.62	0.5621	0.10
Fatty liver on ultrasound. n (%)	51 (23.61)	36 (19.57)	3 (42.86)	9 (40.91)	3 (100)		
ALT (U/l)	25.9 ± 19.1	24.00 ± 15.41	22.83 ± 16.23	27.88 ± 12.79	85.80 ± 48.27	<0.0001	0.51
AST (U/l)	26.3 ± 10.8	25.50 ± 9.19	20.50 ± 7.97	26.33 ± 8.41	58.00 ± 21.97	<0.0001	0.48
Cholesterol (mg/dl)	166.8 ± 36.4	164.26 ± 34.48	164.29 ± 49.63	179.88 ± 37.67	207.00 ± 57.24	0.0177	0.21
HDL-C (mg/dl)	47.0 ± 14.5	47.25 ± 14.76	45.86 ± 7.95	47.91 ± 14.93	36.80 ± 5.22	0.4490	0.11
LDL-C (mg/dl)	101.5 ± 28.3	100.0 ± 27.27	98.14 ± 40.81	112.09 ± 31.10	111.40 ± 32.23	0.2195	0.14
TG (mg/dl)	105.9 ± 69.5	97.80 ± 50.34	142.57 ± 152.28	122.15 ± 54.71	284.00 ± 226.03	<0.0001	0.42
Glucose 0' OGTT (mg/dl)	86.0 ± 8.2	84.46 ± 7.25	104.39 ± 4.03	90.36 ± 7.00	97.14 ± 2.62	<0.0001	0.51
Insulin 0' OGTT (µIu/ml)	15.3 ± 11.9	13.74 ± 8.78	43.77 ± 39.56	17.43 ± 6.27	26.90 ± 2.02	<0.0001	0.47
Glucose 120' OGTT (mg/dl)	114.7 ± 27.2	106.13 ± 15.99	120.90 ± 11.59	150.17 ± 11.37	224.90 ± 20.11	<0.0001	0.82
Insulin 120' OGTT (µIu/ml)	91.4 ± 89.1	69.96 ± 52.61	121.21 ± 81.82	198.14 ± 148.11	262.56 ± 161.03	<0.0001	0.56
HOMA-IR	3.36 ± 3.01	2.90 ± 1.94	11.53 ± 10.95	3.91 ± 1.56	6.44 ± 0.34	<0.0001	0.52
Quicki	0.33 ± 0.03	0.34 ± 0.03	0.29 ± 0.04	0.32 ± 0.02	0.29 ± 0.00	<0.0001	0.37
Matsuda	5.74 ± 4.06	6.43 ± 4.12	2.52 ± 1.68	2.76 ± 1.19	1.40 ± 0.35	<0.0001	0.37

* Obesity, children with obesity with no carbohydrates metabolism disorders; BMI, body mass index; Glucose 0' OGTT, fasting glucose; Insulin 0' OGTT, fasting insulin; Glucose 120' OGTT, glucose after 2 hours of oral glucose tolerance test; Insulin 120' OGTT, insulin after 2 hours of oral glucose tolerance test.

increased to 40.91% (n = 9) subjects and in the group with IFG amounted to 42.86% (n = 3) children (Table 1). ALT levels were above the norm in 10.37% (n=17) of the subjects without carbohydrate metabolism disorders, 15.00% (n=3) of the IGT group and 33.33% (n=2) of the IFG group. When analyzing the occurrence of metabolic complications among the examined children, according to the pubertal development, the presence of sonographic features of fatty liver was found in 18.85% (n=23) of the prepubertal subjects. During the puberty, this percentage increased to 25.00% (n=17), and in postpubertal subjects it was 34.78% (n=8). Considering the values of liver enzymes in the prepubertal subjects abnormal ALT was noted in 11.76% (n=12) of children. During the puberty, this value increased to 8.06% (n=5), while in postpubertal subjects it was 19.23% (n=5). Taking into consideration the carbohydrate metabolism disturbances among children in the prepubertal stage 1.64% (n=2) had abnormal fasting glucose and 8.20% (n=10) had impaired glucose tolerance. In the group of pubertal children, these disorders were present in 6.76% (n=5) and 14.86% (n=11), respectively. Type 2 diabetes was found in none of these groups. All subjects with DM2 were diagnosed in postpubertal children. Moreover, in the post puberty group, glucose tolerance disorders were found in 12.5% of the examined children.

To asses potential cardiovascular risk, we analyzed blood pressure. Differences in systolic and diastolic blood pressure in patients depending on the concentration of alanine aminotransferase and the presence of fatty liver features on ultrasound did not show statistical significance.

Depending on the presence of hepatic steatosis there were significant differences in the values of alanine aminotransferase (p=0.0006; R 0.25), glycaemia after 2 hours of OGTT (p=0.0059; R 0.20), fasting insulin (p= 0.0006; R 0.23) and all insulin resistance indices (Table 2).

The risk of abnormal liver ultrasound imaging expressed as OR in the IGT group was 2.85 times higher (95% Cl 1.13-7.18, p=0.027) compared to children with obesity without carbohydrate metabolism disorders (Figure 1). In case of insulin resistance expressed as HOMA-IR risk of fatty liver

TABLE 2 Subjects characteristic by presence of fatty liver on ultrasound and alanine aminotransferase levels.

	Fatty liver on ultrasoundabsent	Fatty liver on ultrasoundpresent	p	R	ALT < 42U/l	ALT > 42 U/l	p	R
n (%)	165 (77.46)	48 (22.54)			168 (88.42)	22 (11.58)		
Sex – Male, n (%)	86 (77.48)	23 (22.55)			80 (85.11)	14 (14.89)		
Sex – Female, n (%)	79 (77.45)	25 (22.52)			88 (91.67)	8 (8.33)		
Age (years)	9.85 ± 3.46	10.71 ± 3.63	0.1355	0.10	10.21 ± 3.53	10.42 ± 4.15	0.7960	0.02
Weight (kg)	58.14 ± 22.56	69.20 ± 23.57	0.0034	0.20	61.54 ± 23.47	68.85 ± 26.77	0.1784	0.10
Weight SD	2.90 ± 0.89	3.28 ± 0.98	0.0111	0.17	2.97 ± 0.88	3.45 ± 1.14	0.0205	0.17
Height (m)	1.44 ± 0.19	1.50 ± 0.19	0.0538	0.13	1.46 ± 0.19	1.48 ± 0.18	0.6865	0.03
Height SD	1.28 ± 1.21	1.58 ± 1.15	0.1257	0.11	1.32 ± 1.13	1.75 ± 1.41	0.0977	0.12
BMI (kg/m^2)	26.84 ± 4.32	29.86 ± 4.89	0.0001	0.27	27.58 ± 4.47	30.39 ± 6.14	0.0089	0.19
BMI SD	2.91 ± 0.73	3.23 ± 0.81	0.0094	0.18	2.97 ± 0.74	3.42 ± 0.81	0.0081	0.19
Fatty liver on ultrasound, n (%)	–	–	–	–	32 (72.73)	12 (27.27)		
ALT (U/l)	22.41 ± 13.64	31.37 ± 18.06	0.0006	0.25	19.91 ± 7.35	58.43 ± 16.21	<0.0001	0.82
AST (U/l)	25.19 ± 8.91	26.98 ± 9.68	0.2593	0.08	23.73 ± 7.21	38.72 ± 11.65	<0.0001	0.52
Cholesterol (mg/dl)	163.5 ± 34.6	177.6 ± 36.6	0.0196	0.16	163.5 ± 33.0	179.9 ± 41.7	0.0351	0.15
HDL-C (mg/dl)	48.21 ± 15.21	44.21 ± 10.95	0.1080	0.11	47.47 ± 14.99	43.38 ± 10.92	0.2182	0.09
LDL-C (mg/dl)	98.69 ± 26.46	111.68 ± 32.98	0.0074	0.19	99.43 ± 26.59	114.85 ± 35.29	0.0154	0.18
TG (mg/dl)	95.02 ± 49.16	123.56 ± 74.55	0.0028	0.21	98.59 ± 52.81	130.19 ± 90.68	0.0182	0.17
Glucose 0' OGTT (mg/dl)	85.02 ± 8.08	87.79 ± 8.08	0.0384	0.14	85.10 ± 8.21	89.28 ± 7.78	0.0249	0.16
Insulin 0' OGTT (µIu/ml)	13.12 ± 8.33	19.53 ± 17.93	0.0006	0.23	14.46 ± 10.34	22.29 ± 22.26	0.0054	0.20
Glucose 120' OGTT (mg/dl)	108.95 ± 19.46	118.59 ± 23.72	0.0059	0.20	110.89 ± 19.93	118.43 ± 22.05	0.1099	0.12
Insulin 120' OGTT (µIu/ml)	75.25 ± 76.28	101.88 ± 78.72	0.0411	0.15	82.39 ± 81.61	113.92 ± 79.19	0.0971	0.13
HOMA-IR	2.80 ± 1.92	4.44 ± 4.95	0.0007	0.23	3.11 ± 2.43	5.24 ± 6.32	0.0030	0.21
Quicki	0.34 ± 0.03	0.32 ± 0.03	0.0002	0.25	0.33 ± 0.03	0.31 ± 0.03	0.0041	0.21
Matsuda	6.52 ± 4.25	4.44 ± 2.92	0.0022	0.22	6.04 ± 4.18	3.92 ± 2.36	0.0246	0.17

BMI – body mass index. Glucose 0' OGTT – fasting glucose. Insulin 0' OGTT – fasting insulin. Glucose 120' OGTT - glucose after 2 hours of oral glucose tolerance test. Insulin 120' OGTT - insulin after 2 hours of oral glucose tolerance test.

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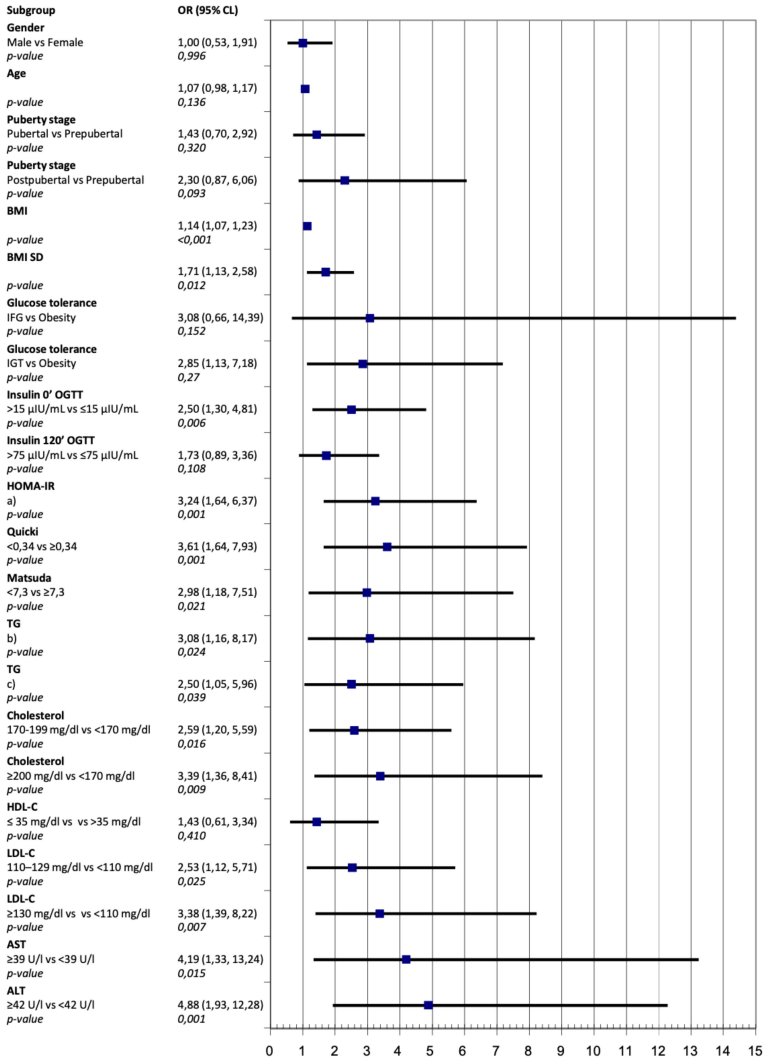

FIGURE 1
Odds ratio for fatty liver on ultrasound for different anthropometrics and biochemical variables.

increased 3.24 times (95% Cl 1.64-6.37, p=0.001), 3.61 times (95% Cl 1.64-7.93, p=0.001) for Quicki and 2.98 for Matsuda (95% Cl 1.18-7.51, p=0.021)

(Figure 1). In subjects with high ALT values this risk was 4.88 times higher (95% Cl 1.93-12.28, p=0.001) than in subjects with normal ALT values (Figure 1).

Logistic regression for ALT shows high odds ratio for children with fatty liver on ultrasound 4.88 (95% Cl 1.93-2.28; p=0.001), for Quicki 4.20 (95% Cl 1.20-14.76; p=0.025) and HOMA-IR 2.72 (95% Cl 1.06-7.02; p=0.038) (Figure 2).

ALT values significantly correlated with BMI SD (r 0.34; p<0.001), insulin after 2 hours of OGTT (r 0.25; p<0.001), insulin resistance indices Quicki (r -0.24; p<0.001), HOMA-IR (r 0.24; p<0,001), Matsuda (r -0.26; p<0.001) and with cholesterol (r 0.27; p<0.001), LDL-C (r 0.23; p<0.001) and triglycerides (r 0.23; p<0.001).

Analysis of the children's eating habits performed on part of the subjects (n=68) shows, that children's rations were not properly balanced. Average percentage of implementation of the norm for energy and main nutrients as well as their percentage share in the diet's energy value was exceeded in almost all analyzed nutrients except for digestible carbohydrates. Energy value of the diets was significantly varied. Average calorie intake as a percentage of suggested norm was about 30% higher

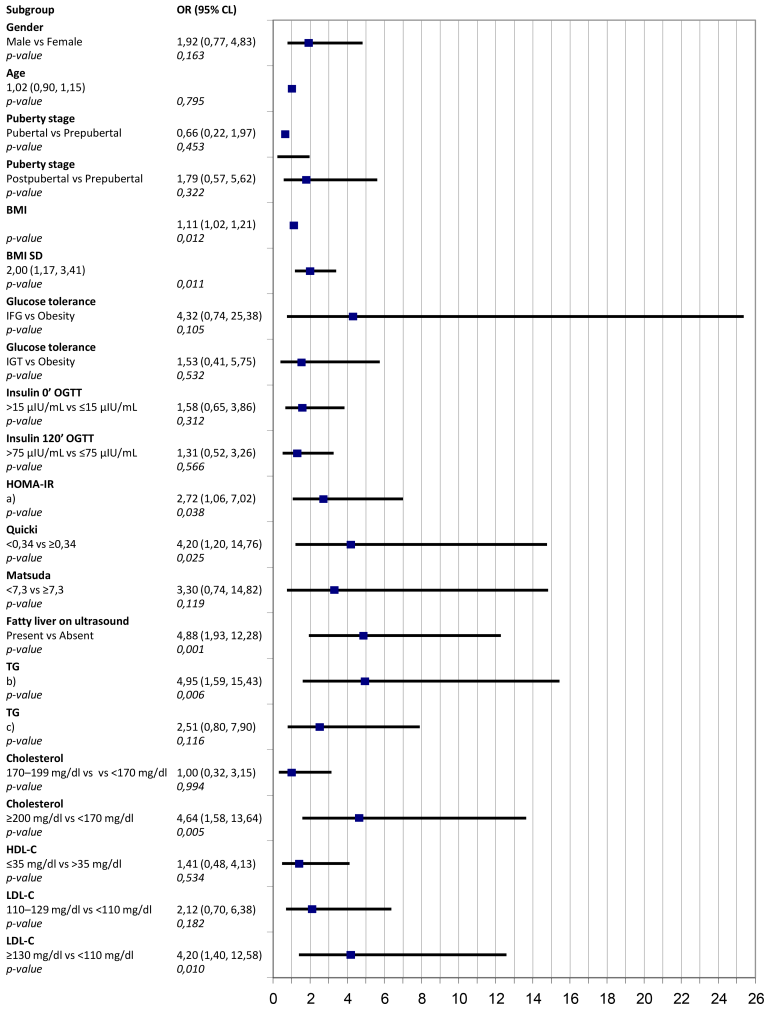

FIGURE 2
Odds ratio for abnormally high levels of alanine aminotransferase for different anthropometrics and biochemical variables.

than the caloric demand in 75% of the respondents. Energy intake from proteins on the suggested level of 12.5% was implemented only in less than 25% of children. Energy from fat made up for 32.2 ± 6.3% of the caloric intake (with the recommendations being 30%). Saturated fat intake exceeded the recommended norm on average by 144.1 ± 52.6%, with only 25% of analyzed children being close to the recommended goal.

Discussion

According to data provided by CDC (Center for Disease Control and Prevention) in the last 30 years the number of children with obesity has increased twofold, while the number of obese teenagers has tripled. WHO (World Health Organization) estimations say, that in 2019 there were 38 million overweight children below the age of 5 in the world, almost half of which lived in Asia.

In the Polish population, similar to other countries in Central-Eastern Europe, which were subjected to economic and social changes, there is a visible increase in prevalence of obesity in the pediatric population. In 2007-2009 a study was performed on a random, representative population sample in the age of 7-18. The study was performed as a part of the OLAF project. According to norms set by this study, overweight and obesity was observed in 18.7% boys and 14.1% girls. In Western Pomerania a screening study concerning the prevalence of overweight and obesity was performed among children in the

age 8-9 (own data, soon to be published). 4903 children have been screened, of which 22.8% had overweight and obesity – 16.5% were overweight and 6.3% were obese.

Obesity in children is a cause of many metabolic complications, including carbohydrate metabolism disorders, lipid disorders, hypertension and NAFLD. Some authors discuss a possibility of association of certain obesity phenotypes with a different cardiovascular risk (20). It may be argued, that the same phenotypes may be associated with an increased risk of NAFLD development.

Early data publish in NEJM in 2002 suggest the prevalence of impaired glucose tolerance in 25% of children with obesity in the age of 4-10 and 21% obese teenagers in the age of 11-18. In this study "silent type 2 diabetes" was diagnosed in 4% of obese teenagers (21). Italian studies on increasing incidence of carbohydrate metabolism disorders in children with obesity show that those disorders were found in 12.4% of patients. The most common disorder was impaired glucose tolerance, found in 11.2% of patients. It was more common in teenagers than in children (14.8% vs. 4.1%). "Silent T2DM" was diagnosed in two teenagers (0.4%) (22).

In presented analysis impaired fasting glucose was present in 3.07% (n=7) of the subjects, impaired glucose tolerance in 10.97% (n=25); in 2.19% (n=5) of subjects based on OGTT type 2 DM was diagnosed, which may be considered "silent T2DM". Despite varying data concerning different populations, increasing prevalence of carbohydrate metabolism disorders in children with obesity remains a fact. To quantify the grade of insulin resistance, HOMA-IR, Quicki and Matsuda indices were used in this study, as well as fasting insulin levels. As expected, HOMA-IR was high in children with type 2 DM. Surprisingly high values of HOMA-IR were also found in children with impaired fasting glucose (Table 1). What certainly draws attention while analyzing biochemical studies' results of patients with carbohydrate metabolism disorders are elevated aminotransferase levels, especially ALT, which was highest in children with T2DM (Table 1). Because other biochemical parameters in T2DM children were also significantly different from the rest of the analyzed group, they were considered outliers and excluded from further statistical analysis.

Another important complication of obesity is nonalcoholic fatty liver disease. It is defined as accumulation of fat in the liver in the absence of excessive alcohol consumption or other liver pathologies (23). Spectrum of the disease ranges from steatosis to steatohepatitis, which is characterized by hepatocellular inflammation and injury, progressing to fibrosis and eventually cirrhosis (24, 25). NAFLD is one of the most common causes of chronic liver disease in childhood (26).

The incidence of NAFLD is hard to estimate, both in the adult and pediatric population. The golden standard of diagnosing NAFLD is liver biopsy (25), which difficult to use in everyday practice – hence the search for other, less invasive methods, such us checking the serum levels of inflammation markers and assessing the liver in imaging studies such as ultrasonography or *magnetic resonance imaging* (MRI). In light of the most recent guidelines, the idea of liver biopsy being the golden standard in NAFLD diagnostics need to be re-evaluated – while it indeed remains the diagnostic gold standard for the assessment of fibrosis and inflammation, it is not recommended in simple hepatosteatosis (27). Moreover, biopsies have known limitations and possible disadvantages which ring especially true in the pediatric population - a risk of complications due to the procedure being invasive, sampling variability because of the small sample obtained, and the heterogeneous distribution of histological changes in liver parenchyma. As pointed out by the authors, routine liver biopsies performed specifically to confirm NAFLD seem unwarranted due to the prevalence of the condition, and may even be considered unethical. While liver biopsy remains the only diagnostic tool to confirm non-alcoholic steatohepatitis (NASH), its prevalence among NAFLD patients is quite low - estimated at 1.5%-6.5%. Considering these points, authors suggest, that liver biopsies should only be performed in patients at high risk of the progressive type of the disease (28, 29).

In existing publications, the prevalence of NAFLD was estimated to be somewhere in a wide range between 1.7% to 85% of children with obesity (30–33). Such a wide range depends on the analyzed population and chosen diagnostic methods. In an analysis by American authors considering 408 children with obesity in the mean age of 13.2, NAFLD was found in 26% of children, more often in boys 29.4% than girls 22.6% (34). Meta-analysis presented by Anderson et al. estimates the incidence of NAFLD in the general population at 7.6%, and 34.2% in children with obesity (35).

In available literature several studies can be found, where biopsy was used as a diagnostic criterium. Schwimmer et al. published a report in which based on autopsy studies of 742 children in the age of 2-19 hepatic steatosis was found in 9.6% of children with normal weight and 38% of children with obesity (36). One of the imaging studies used to assess the grade of hepatic steatosis is magnetic resonance (MRI), but because of the costs of the study ultrasonography is used more often (35, 37). Hernaez et al. in their analysis have found ultrasonography to be accurate and reliable for detection of moderate-sever fatty liver (sensitivity 84.8%, specificity 93.6% compared to histology) (38). As opposed to adult recommendations by EASL-EASD-EASO (10), NASPHGAN does not recommend ultrasound as a screening for NAFLD in children. It opens the area for further discussion, as ultrasound, being a cheap, simple, non-invasive and repetitive examination, which would make it a suitable choice for the pediatric population. It is also undeniable, that the constant technological progress in the radiology department improves the result achieved by the examinations.

In our study hepatic steatosis was found in 22.54% of the subjects. This prevalence is lower than previously reported by

other authors. It may be caused by the young age of the whole study group and the fact, that 122 of the children were prepubertal. Even though they were already overweight, they may have not yet presented with NAFLD as a complication of obesity.

As reported by Younossi, glucose intolerance, and especially T2DM, is an accelerator of NAFLD and a predictor of severe fibrosis and mortality (39). In our study we tried to assess the fraction of children with NAFLD according to the grade of glucose metabolism disorders. In children without carbohydrate metabolism disorders features of fatty liver were found in 19.57% of the subjects, while in children with glucose intolerance 40.91% of the subjects were affected. All of the children with type 2 diabetes were diagnosed with hepatic steatosis. Increasing percentage of children with hepatic steatosis in groups with glucose intolerance undoubtedly shows a correlation between carbohydrate metabolism disorders and fatty lived disease. Odds ratio (OR) calculations show, that the risk of hepatic steatosis is, 2.85 times higher in children with impaired glucose tolerance.

In our study there is no noticeable difference in occurrence of hepatic steatosis in ultrasound examination when it comes to children's sex (22.55% vs 22.52%), which is consistent with the study by Prokopowiczet al. (40)

In pediatric populations, a meta-analytic study showed that the pooled NAFLD prevalence is higher in boys than in girls in general populations and obese clinical cohorts (41). The study also revealed significant variance across the published reports, which is partly explained by the technique used to diagnose NAFLD (ultrasound versus aminotransferases) and by failure to consider pubertal stages when sex hormone levels change dramatically in a sex-specific manner (41). According to the meta-analysis by Anderson et al. (35), the incidence of hepatic steatosis based only on USG is similar in both sexes, whereas considering only ALT, it is more common in boys. Our findings in the presented study support similar results. Based on ultrasound imaging the prevalence of hepatic steatosis doesn't differ between the sexes, while based on ALT it is more common in boys, that difference is however not statistically significant.

Many studies support the central role of insulin resistance in the development of fatty liver (42). The presence of liver steatosis is an important marker of multiorgan insulin resistance, independently of BMI, body fat percentage, and visceral fat mass (43). In particular, NAFLD has been found to be associated with insulin resistance in liver, skeletal muscles and adipose tissue in children and adolescents with obesity (44).

In our study high values of insulin resistance indices QUICKI, HOMA-IR, Matsuda significantly increase the risk of hepatic steatosis (OR 3.61; 3.24 and 2.98 respectively, $p<0.001$). In the presence of elevated fasting insulin levels, the risk of hepatic steatosis was also significantly higher – OR 2.50 ($p=0.006$).

According to NASPGHAN guidelines, serum ALT levels should be used as a screening tool for assessing the risk of NAFLD, with normal values <22 U/l for girls and <26 U/l for boys (3). In our analysis local laboratory's norm was used, with values above 42 U/l determined as too high. In the study group 11.58% of children (n=22) had elevated ALT levels. In the group with fatty liver features in USG examination those values were elevated 27.27% of children (n=12). In the group of children with high ALT levels 54.55% of children (n=12) had features of fatty liver. It must be noted, however, that the mean value of ALT levels in the whole study group was 24.37±15.16 U/l, which exceeds the values given as reference by NASPGHAN in all analyzed subgroups. Especially high levels were observed in the group of children with impaired glucose tolerance Table 1). In previously cited study by Yu et al. optimal cut-off points for ALT have been established according to sex as 42 U/l for boys and 30 U/l for girls (34).

In our analysis in 10.37% of the subjects with no carbohydrate metabolism disorders ALT values were abnormal. The percentage of children with abnormal ALT levels in IGT group was 15.00%, in IFG group 33.33% and in T2DM group 80.00%. Meta-analysis published by Anderson et al. shows, that in 14 clinical studies concerning children with obesity abnormal ALT values were observed in 13.7% of children (6.2 to 27.6%) (35). It is emphasized, that using ALT to diagnose NAFLD may cause overdiagnosis of this ailment in persons with normal body weight, as well as underestimation of this problem in patients with obesity.

In the proprietary study ALT levels correlated with insulin after 2 hours of OGTT, insulin resistance indices as well as triglyceride and cholesterol levels.

The assessment of odds ratio shows, that the highest risk of abnormal ALT values concerns children with high insulin resistance indices, hypertriglyceridemia and hypercholesterolemia. OR values for elevated ALT levels and fatty liver features in sonograms is 4.88 (95%Cl 1.93-12.28, $p=0.001$). Considering adipose tissue is a hormonally active tissue, some adipokines may be associated with the metabolic state of the child – as such, the leptin/adiponectin ratio may be a useful indicator of insulin resistance (45).

Routine work with a child with obesity at risk of NAFLD always involves cooperation with a dietitian. In our analysis, declared energy consumption presented as a percentage of the norm were higher by about 30% in relation to the caloric need in as many as 75% of the subjects. Declared consumption of simple carbohydrates amounted to 112.4±68.9% of the norm.

An analysis of quantity and quality of products consumed by the child, as well as eating habits correction are crucial. Latest studies show, that diet rich in saccharose and HFCS not only increases the risk of NAFLD, but also NASH (46, 47). Excessive consumption of fructose and saccharose correlates with epidemic increase of obesity, metabolic syndrome and NAFLD occurrence (48). Long-term excessive fructose consumption leads to leptin resistance and, as a consequence, obesity (49). Anika Nier et al. in their study concerning the effect of fructose

consumption and nutrition patterns on the development of NAFLD in children with obesity emphasize, that children with obesity and early symptoms of NAFLD have a higher energy intake in comparison to children without hepatic steatosis, mostly caused by fructose and saccharose consumption (6). The most recommended way of preventing and treating obesity is still taking measures to change the patient's lifestyle, including modification of the eating habits and increasing physical activity.

What we consider to be the strength of the study is it being performed on a group of children, considering the scarcity of publications on the subject of NAFLD in the pediatric population. Furthermore, the group consisting of consecutive children from one region of Poland guarantees the subjects' homogeneity (only Caucasian origin), without placing bias on the study group by selecting specific patients. The most important limitation of our study is a relatively low number of subjects in each sub-group, with type 2 diabetes. It is known though, that for type 2 diabetes to develop a longer history of obesity is needed. In our opinion, supported by other authors' research, those patients require special attention. In the future they are possible candidates for combined pharmacological or bariatric treatment (50).

The study was also performed on a group of children in a wide range of age (2-18), which may cause the results to be skewed by the effect of sex hormones in older children. Metabolic complications of obesity, such as fatty liver, hypertransaminasemia and disturbances in glucose metabolism, are significantly more common in older children and with advanced puberty. DM2 only occurred in children with Tanner stage V.

Only ALT and AST were used as markers of liver function. However, they are considered to be the first routinely performed assessment of liver function in children, who (if required) went on to be diagnosed further. Furthermore, studies evaluating GGTP as a marker of hepatic steatosis in children have not yet been performed. One of the aims of the study was to assess if routinely performed laboratory tests, i.e. ALT and AST, could be considered sufficient to diagnose NAFLD in those children. What needs to be noted is that the study is a clinical analysis performed by pediatric endocrinologists in children diagnosed because of obesity, with no assumptions being made as for the presence of liver dysfunction beforehand, which is why the tests performed during diagnostics are not meant to specifically focus on that subject. The list of possible complications of obesity is immeasurably long, meaning that diagnosing each and every system in the patient's body during one visit in the outpatient's clinic is technically impossible, which forces physicians to look for the most effective screening studies possible.

One of the screening tests that may be an indicator of both NAFLD and cardiovascular risk may be waist to height ratio. In the analysis of Umano and al (51). it has been proven that WHR can be a good screening anthropometric parameter for the assessment of NAFLD. Blood pressure measurement is essential to assess cardiovascular risk in obese subjects. In our analysis differences in systolic and diastolic blood pressure in patients depending on the concentration of alanine aminotransferase and the presence of fatty liver features on ultrasound did not show statistical significance.

Further research is required with other variables considered, such as dividing the children in sub-groups according to the stage of liver steatosis. Novel diagnostic methods may also play a bigger role in NAFLD diagnostics, however further research is necessary, as not all innovative techniques will gain success in the clinical and research area. Reevaluation of ALT normative values seems necessary, which requires them to be included in later studies.

Conclusions

Almost every fourth child with obesity in the study group presents features of fatty liver in ultrasound examination. Although ultrasound is not recommended by NASPGHAN for the diagnosis of NAFLD in children, it allows identifying a high percentage of children with features of fatty liver. This percentage increases significantly in children with glucose intolerance. Routinely used ALT reference values may be responsible for a high percentage of subjects with normal ALT levels despite the presence of hepatic steatosis in ultrasound.

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 Pomeranian Medical University Bioethical Committee on April 14 2004, decision number BN-001/67/04. Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

Author contributions

Conceptualization, KM, EP and MW; methodology, KM, EP, and AH-J; software, TJ and AB-M; validation, KM, EP and MW; formal analysis, KM and KS; investigation, KM, KS and EP; resources, KM, KS and AB-M; data curation, KM and KS; writing—original draft preparation, KM, EP and AH-J; writing—review and editing, TJ, EP, AB-M and MW; visualization, TJ and AB-M; supervision, EP and MW; project administration,

KM and EP. All authors contributed to the article and approved the submitted version.

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. Pacifico L, Nobili V, Anania C, Verdecchia P, Chiesa C. Pediatric nonalcoholic fatty liver disease, metabolic syndrome and cardiovascular risk. *World J Gastroenterol* (2011) 17(26):3082–91. doi: 10.3748/wjg.v17.i26.3082

2. Ranucci G, Spagnuolo MI, Iorio R. Children with obesity with fatty liver: Between reality and disease mongering. *World J Gastroenterol* (2017) 23(47):8277–82. doi: 10.3748/wjg.v23.i47.8277

3. Vos MB, Abrams SH, Barlow SE, Caprio S, Daniels SR, Kohli R, et al. NASPGHAN clinical practice guideline for the diagnosis and treatment of nonalcoholic fatty liver disease in children: Recommendations from the expert committee on NAFLD (ECON) and the north American society of pediatric gastroenterology, hepatology and nu. *J Pediatr Gastroenterol Nutr* (2017) 64 (2):319–34. doi: 10.1097/MPG.0000000000001482

4. Shaunak M, Byrne CD, Davis N, Afolabi P, Faust SN, Davies JH. Non-alcoholic fatty liver disease and childhood obesity. *Arch Dis Child.* (2021) 106(1):3–8. doi: 10.1136/archdischild-2019-318063

5. Alves-Bezerra M, Cohen DE. Triglyceride metabolism in the liver. *Compr Physiol* (2018) 8(1):1–22. doi: 10.1002/cphy.c170012

6. Nier A, Brandt A, Conzelmann IB, Özel Y, Bergheim I. Non-alcoholic fatty liver disease in overweight children: Role of fructose intake and dietary pattern. *Nutrients* (2018) 10(9):1–18. doi: 10.3390/nu10091329

7. Softic S, Stanhope KL, Boucher J, Divanovic S, Lanaspa MA, Johnson RJ, et al. Fructose and hepatic insulin resistance. *Crit Rev Clin Lab Sci* (2020) 57(5):308–22. doi: 10.1080/10408363.2019.1711360

8. Day CP. Pathogenesis of steatohepatitis. *Bailliere's Best Pract Res Clin Gastroenterol* (2002) 16(5):663–78. doi: 10.1053/bega.2002.0333

9. Bugianesi E, McCullough AJ, Marchesini G. Insulin resistance: A metabolic pathway to chronic liver disease. *Hepatology* (2005) 42(5):987–1000. doi: 10.1002/hep.20920

10. Marchesini G, Day CP, Dufour JF, Canbay A, Nobili V, Ratziu V, et al. EASL-EASD-EASO clinical practice guidelines for the management of non-alcoholic fatty liver disease. *J Hepatol* (2016) 64(6):1388–402. doi: 10.1016/j.jhep.2015.11.004

11. Pacifico L, Ferraro F, Bonci E, Anania C, Romaggioli S, Chiesa C. Upper limit of normal for alanine aminotransferase: Quo vadis? *Clin Chim Acta* (2013) 422:29–39. doi: 10.1016/j.cca.2013.03.030

12. Schwimmer JB, Dunn W, Norman GJ, Pardee PE, Middleton MS, Kerkar N, et al. SAFETY study: Alanine aminotransferase cutoff values are set too high for reliable detection of pediatric chronic liver disease. *Gastroenterology* (2010) 138 (4):1357–1364.e2. doi: 10.1053/j.gastro.2009.12.052

13. Schwimmer JB, Newton KP, Awai HI, Choi LJ, Garcia MA, Ellis LL, et al. Paediatric gastroenterology evaluation of overweight and children with obesity referred from primary care for suspected non-alcoholic fatty liver disease. *Aliment. Pharmacol Ther* (2013) 38(10):1267–77. doi: 10.1111/apt.12518

14. Palczewska I, Szilagyi-Pagowska I. Ocena rozwoju somatycznego dzieci i młodzieży. *Med Prakt. Pediatr* (2002) 3:140–69.

15. 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

16. Shashaj B, Luciano R, Contoli B, Morino GS, Spreghini MR, Rustico C, et al. Reference ranges of HOMA-IR in normal-weight and obese young caucasians. *Acta Diabetol* (2016) 53(2):251–60. doi: 10.1007/s00592-015-0782-4

17. Ballestri S, Nascimbeni F, Lugari S, Lonardo A, Francica GA. Critical appraisal of the use of ultrasound in hepatic steatosis. *Expert Rev Gastroenterol Hepatol* (2019) 13(7):667–81. doi: 10.1080/17474124.2019.1621164

18. Szponar L, Wolnicka K, Rychlik E. *Album fotografii produktów i potraw Album of photographs of food products and dishes*. Warszawa. Instytut Żywności Żywienia. (2000).

19. Jarosz M, Rychlik E, Stoś K, Wierzejska R, Wojtasik A, Charzewska J, et al. *Normy Żywienia dla populacji polski*. Warszawa: Insytut Żywności i Żywienia (2017).

20. Vecchié A, Dallegri F, Carbone F, Bonaventura A, Liberale L, Portincasa P, et al. Obesity phenotypes and their paradoxical association with cardiovascular diseases. *Eur J Intern Med* (2018) 48:6–17. doi: 10.1016/j.ejim.2017.10.020

21. Sinha R, Fisch G, Teague B, Tamborlane WV, Banyas B, Allen K, et al. Prevalence of impaired glucose tolerance among children and adolescents with marked obesity. *N Engl J Med* (2002) 346(11):802–10. doi: 10.1056/NEJMoa012578

22. Brufani C, Ciampalini P, Grossi A, Fiori R, Fintini D, Tozzi A, et al. Glucose tolerance status in 510 children and adolescents attending an obesity clinic in central Italy. *Pediatr Diabetes* (2010) 11(1):47–54. doi: 10.1111/j.1399-5448.2009.00527.x

23. Giorgio V, Prono F, Graziano F, Nobili V. Pediatric non alcoholic fatty liver disease: Old and new concepts on development, progression, metabolic insight and potential treatment targets. *BMC Pediatr* (2013) 13(1):13–40. doi: 10.1186/1471-2431-13-40

24. Loomba R, Sirlin CB, Schwimmer JB, Lavine JE. Advances in pediatric nonalcoholic fatty liver disease. *Hepatology* (2009) 50(4):1282–93. doi: 10.1002/hep.23119

25. Janczyk W, Socha P. Non-alcoholic fatty liver disease in children. *Clin Res Hepatol Gastroenterol* (.2012) 36(3):297–300. doi: 10.1016/j.clinre.2012.03.026

26. Di Sessa A, Cirillo G, Guarino S, Marzuillo P, Miraglia del Giudice E. Pediatric non-alcoholic fatty liver disease: Current perspectives on diagnosis and management. *Pediatr Heal Med Ther* (2019) 10:89–97. doi: 10.2147/phmt.s188989

27. Chalasani N, Younossi Z, Lavine JE, Charlton M, Cusi K, Rinella M, et al. The diagnosis and management of nonalcoholic fatty liver disease: Practice guidance from the American association for the study of liver diseases. *Hepatology* (2018) 67(1):328–57. doi: 10.1002/hep.29367

28. GBD 2015 Obesity Collaborators, Afshin A, Forouzanfar MH, Reitsma MB, Sur P, Estep K, et al. Health effects of overweight and obesity in 195 countries over 25 years. *N Engl J Med* (2017) 377(1):13–27. doi: 10.1056/nejmoa1614362

29. Lind L, Johansson L, Ahlström H, Eriksson JW, Larsson A, Risérus U, et al. Comparison of four non-alcoholic fatty liver disease detection scores in a Caucasian population. *World J Hepatol* (2020) 12(4):149–59. doi: 10.4254/wjh.v12.i4.149

30. Rocha R, Cotrim HP, Bitencourt AGV, Barbosa DBV, Santos AS, Almeida AdM, et al. Nonalcoholic fatty liver disease in asymptomatic Brazilian adolescents. *World J Gastroenterol* (2009) 15(4):473–7. doi: 10.3748/wjg.15.473

31. Jimenez-Rivera C, Hadjiyannakis S, Davila J, Hurteau J, Aglipay M, Barrowman N, et al. Prevalence and risk factors for non-alcoholic fatty liver in children and youth with obesity. *BMC Pediatr* (2017) 17(1):1–7. doi: 10.1186/s12887-017-0867-z

32. Xanthakos SA, Jenkins TM, Kleiner DE, Boyce TW, Mourya R, Karns R, et al. High prevalence of nonalcoholic fatty liver disease in adolescents undergoing bariatric surgery. *Gastroenterology* (2015) 149(3):623e8–634.e8. doi: 10.1053/j.gastro.2015.05.039

33. Mohamed RZ, Jalaludin MY, Anuar Zaini A. Predictors of non-alcoholic fatty liver disease (NAFLD) among children with obesity. *J Pediatr Endocrinol Metab* (2020) 33(2):247–53. doi: 10.1515/jpem-2019-0403

34. Yu EL, Golshan S, Harlow KE, Angeles JE, Durelle J, Goyal NP, et al. Prevalence of nonalcoholic fatty liver disease in children with obesity. *J Pediatr* (2019) 207:64–70. doi: 10.1016/j.jpeds.2018.11.021

35. Anderson EL, Howe LD, Jones HE, Higgins JPT, 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

36. Schwimmer JB, Deutsch R, Kahen T, Lavine JE, Stanley C, Behling C. Prevalence of fatty liver in children and adolescents. *Pediatrics* (2006) 118(4):1388–93. doi: 10.1542/peds.2006-1212

37. Bohte AE, Van Werven JR, Bipat S, Stoker J. The diagnostic accuracy of US, CT, MRI and 1H-MRS for the evaluation of hepatic steatosis compared with liver biopsy: A meta-analysis. *Eur Radiol* (2011) 21(1):87–97. doi: 10.1007/s00330-010-1905-5

38. Hernaez R, Lazo M, Bonekamp S, Kamel I, Brancati FL, Guallar E, et al. Diagnostic accuracy and reliability of ultrasonography for the detection of fatty liver: A meta-analysis. *Hepatology* (2011) 54(3):1082–90. doi: 10.1002/hep.24452

39. Younossi ZM. Non-alcoholic fatty liver disease – a global public health perspective. *J Hepatology. Eur Assoc Study Liver* (2019) 70(3):531–44. doi: 10.1016/j.jhep.2018.10.033

40. Prokopowicz Z, Malecka-Tendera E, Matusik P. Predictive value of adiposity level, metabolic syndrome, and insulin resistance for the risk of nonalcoholic fatty liver disease diagnosis in children with obesity. *Can J Gastroenterol Hepatol* (2018) 2018:9465784. doi: 10.1155/2018/9465784

41. Suzuki A, Lonardo A, Nascimbeni F, Ballestri S, Fairweather D, Win S, et al. Sex differences in nonalcoholic fatty liver Disease : State of the art and identification of research gaps. *Hepatology* (2019) 70(4):1457–69. doi: 10.1002/hep.30626

42. Fabbrini E, Magkos F, Mohammed BS, Pietka T, Abumrad NA, Patterson BW, et al. Intrahepatic fat, not visceral fat, is linked with metabolic complications of obesity. *Proc Natl Acad Sci U. S. A* (2009) 106(36):15430–5. doi: 10.1073/pnas.0904944106

43. D'Adamo E, Cali AMG, Weiss R, Santoro N, Pierpont B, Northrup V, et al. Central role of fatty liver in the pathogenesis of insulin resistance in obese adolescents. *Diabetes Care* (2010) 33(8):1817–22. doi: 10.2337/dc10-0284

44. Perseghin G, Bonfanti R, Magni S, Lattuada G, De Cobelli F, Canu T, et al. Insulin resistance and whole body energy homeostasis in obese adolescents with fatty liver disease. *Am J Physiol - Endocrinol Metab* (2006) 291(4):697–703. doi: 10.1152/ajpendo.00017.2006

45. Frühbeck G, Catalán V, Rodríguez A, Ramírez B, Becerril S, Salvador J, et al. Involvement of the leptin-adiponectin axis in inflammation and oxidative stress in the metabolic syndrome. *Sci Rep* (2017) 7:6619. doi: 10.1038/s41598-017-06997-0

46. Abdelmalek MF, Suzuki A, Guy C, Unalp-Arida A, Colvin R, Johnson RJ, et al. Increased fructose consumption is associated with fibrosis severity in patients with nonalcoholic fatty liver disease. *Hepatology* (2010) 51(6):1961–71. doi: 10.1002/hep.23535

47. Jegatheesan P, De Bandt JP. Fructose and NAFLD: The multifaceted aspects of fructose metabolism. *Nutrients* (2017) 9(3):1–13. doi: 10.3390/nu9030230

48. Mager DR, Patterson C, So S, Rogenstein CD, Wykes LJ, Roberts EA. Dietary and physical activity patterns in children with fatty liver. *Eur J Clin Nutr* (2010) 64(6):628–35. doi: 10.1038/ejcn.2010.35

49. Fang YL, Chen H, Wang CL, Liang L. Pathogenesis of non-alcoholic fatty liver disease in children and adolescence: From "Two hit theory" to "Multiple hit model. *World J Gastroenterol* (2018) 24(27):2974–83. doi: 10.3748/wjg.v24.i27.2974

50. Leitner DR, Frühbeck G, Yumuk V, Schindler K, Micic D, Woodward E, et al. Obesity and type 2 diabetes: Two diseases with a need for combined treatment strategies - EASO can lead the way. *Obes Facts.* (2017) 10:483–92. doi: 10.1159/000480525

51. Umano GR, Grandone A, Di Sessa A, Cozzolino D, Pedullà M, Marzuillo P, et al. Pediatric obesity-related non-alcoholic fatty liver disease: waist-to-height ratio best anthropometrical predictor. *Pediatr Res* (2021) 90:166–70. doi: 10.1038/s41390-020-01192-w

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Insulin resistance in school-aged girls with overweight and obesity is strongly associated with elevated white blood cell count and absolute neutrophil count

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Background: The primary objective of the study was to discuss the sex differences in insulin resistance-induced changes in metabolic and inflammatory markers in school-aged children with overweight and obesity.

Methods: A cross-sectional study of 800 children aged seven and twelve years was performed. Questionnaires, anthropometric data and fasting blood samples were collected.

Results: Children with overweight and obesity showed statistically significant differences in multiple metabolic and inflammatory markers compared with children with normal BMI. The correlation coefficient (*r*) between white blood cell count, absolute neutrophil count, fasting plasma insulin, HOMA-IR, HOMA-β, triglyceride, HDL-C, triglyceride/HDL ratio, alanine transaminase, serum uric acid, systolic blood pressure and BMI were higher in all children, but the linear relationships between white blood cell count, absolute neutrophil count and BMI were stronger in girls with overweight and obesity than in boys with overweight and obesity. Subsequently, HOMA-IR was shown to be more strongly associated with increased white blood cell count and absolute neutrophil count in school-aged girls with overweight and obesity by partial correlation analysis and the multiple linear regression analysis.

Conclusions: Elevated white blood cell count and absolute neutrophil count in children with overweight and obesity, especially girls, can serve as markers of insulin resistance.

KEYWORDS

pediatric obesity, sex, insulin resistance, white blood cell count, absolute neutrophil count

Introduction

Obesity of children and adolescents is a worldwide health problem that is becoming more prevalent in low- and middle-income countries, as in many high-income countries. According to the assessment of the World Obesity Federation, 206 million children and adolescents aged 5-19 will be obese by 2025, and by 2030 there will be 254 million. In 42 countries with an estimated 1 million children with obesity in 2030, China ranks first, followed by India, the United States, Indonesia and Brazil, and only seven of the top 42 are high-income countries (1). About a fifth of all children are overweight or obese according to the Chinese sex-age-specific BMI cutoff points in China. By 2030, the prevalence of overweight and obesity in school-age children may come to 31.8%, or about 58.92 million (2). Obesity is often associated with insulin resistance and is the leading cause of insulin resistance in children. The main cause of children's insulin resistance is the typical lipid distribution pattern, that is, increased deposition of lipids in insulin sensitive tissue such as the liver, skeletal muscle and viscera. This lipid deposition pattern is also associated with the infiltration of immune system cells into intra-abdominal tissues, inducing systemic low-grade inflammation (3).

Reavan GM argues that insulin resistance is crucial in the pathogenesis of type 2 diabetes, hypertension, and coronary heart disease (4). Insulin resistance manifests as hyperinsulinemia and is a driver of dyslipidemia, high blood pressure and altered glucose metabolism (5). Therefore, insulin resistance can cause changes in multiple metabolic and systemic inflammatory markers (6–8). However, this effect may differ between boys and girls, and there are few such studies. Therefore, the aim of the present study was to determine if sex differences existed in insulin resistance-induced changes in metabolic and inflammatory markers in school-aged children who were overweight and obese.

Materials and methods

Study design and samples

This cross-sectional study was carried out in Kaifeng, Henan Province from October to November 2019 and July to September 2020, during which time it was interrupted due to the COVID-19 outbreak. Five primary schools were selected by cluster sampling from 33 primary schools of a district in Kaifeng. According to the data from last year's school physical examination, 1,297 children aged 7-12 years were investigated by simple random sampling. Eight hundred children completed questionnaires, anthropometric measures and laboratory tests, but the remainder did not take fasting blood samples because their parents or guardians did not agree or had no time to attend.

Inclusion criteria for the study samples were:

-All participants participated in the study voluntarily.

-They were students in grade 2 to 6 of primary school.

-No metabolic or endocrine disease.

Exclusion criteria for the study samples were:

-Any pathological changes, such as endocrine, metabolic or inadequate renal function, which may contribute to changes in dietary habits and nutrient intake.

-Infectious diseases and treatment with antibiotics.

Written informed consents were acquired from parents or guardians. This study was approved by the Ethics Committee of Henan University.

Questionnaire survey

The demographic characteristics, lifestyle, diet, home environment, maternal pregnancy, feeding patterns in infancy and other risk factors were assessed with standardized questionnaires for students and parents. Under the guidance of well-trained investigators, the student questionnaire was completed in the school. The parents' questionnaire was taken home and filled in by the parents. After all questionnaires are collected, if there are any problems, the investigators would call the parents to verify.

Anthropometric studies

The height, weight and waist circumference of children wearing light clothing without shoes were measured with standard methods, and the data were accurate to 0.1 cm and 0.1 kg. The electronic sphygmomanometer (Omron HEM-7136) was used to measure the systolic and diastolic blood pressure for three times with an interval of 30 seconds. The average value was calculated for analysis. Body mass index (BMI) was calculated by dividing a child's weight (in kilograms) by height (in square meters). The diagnosis of overweight and obesity is based on Chinese sex- and age-specific BMI criteria, that is, a BMI at or above the 85^{th} and 95^{th} percentile, respectively (9). The age-specific BMI Z-scores were calculated using WHO AnthroPlus software. According to BMI Z-scores, children were classified as: normal weight with Z-scores from −2 to +0.99, overweight from 1 to 1.99, obese from 2 to 2.99, and very obese ≥ 3 (10, 11).

Hematological and clinical biochemical studies

Overnight fasting blood samples were collected for measuring blood routine examination, blood lipid level, liver

function, kidney function, fasting blood glucose, fasting insulin and C-reactive protein. Insulin resistance and β-cell function were calculated from fasting blood glucose and fasting insulin by the homeostasis model assessment of insulin resistance (HOMA-IR index) (12). The neutrophil-to-lymphocyte ratio was determined by dividing the absolute neutrophil count by the absolute lymphocyte count, the platelet-to-lymphocyte ratio determined by dividing the platelet count by the absolute lymphocyte count, and the triglyceride/HDL ratio determined by dividing the triglyceride level by the high-density lipoprotein cholesterol (HDL-C) level (13–15).

Statistical analysis

None of the quantitative variables were normally distributed. Quantitative variables and categorical variables are summarized as median (interquartile range) and number (percentage), respectively. Differences between participants with and without overweight/obesity or male and female were evaluated by the nonparametric Wilcoxon test for quantitative data and the chi-square test for categorical data. Although the quantitative variables in this study do not conform to a normal distribution, they can be regarded as approximately a normal distribution due to the large sample size. So the partial correlation analysis was used to analyze the linear relationship between BMI and the clinical indicators and between HOMA-IR and the clinical indicators adjusted for confounding factors. Multiple linear regression analysis showed a sex difference in the association of HOMA-IR and the white blood cell count/absolute neutrophil count in children who were overweight and obese.

All analyses were performed with SPSS 26.0 (IBM, Armonk, NY, USA), and a two-tailed $P<0.05$ was the level of statistically significant.

Results

Clinical characteristics of population

800 school-age children (474 boys and 326 girls) aged between 7 to 12 years were included in this study. According to the BMI Z-score established by the WHO, there were 161 children with normal weight and 639 children with overweight and obesity. According to Chinese sex- and age-specific BMI criteria, 181 children are of normal BMI and 619 are overweight or obese. There was no statistical difference between the two classification methods (χ^2 = 1.49, P=0.223). The classification method used in this study is Chinese criteria.

The clinical characteristics of the normal BMI and overweight/obesity participants are presented in Table 1. In addition to absolute lymphocyte counts, absolute basophils count, fasting plasma glucose, aspartate transaminase, blood urea nitrogen and serum creatinine, other clinical indicators were statistically different between children with normal BMI and those who were overweight and obese.

Linear relationship between BMI and clinical indicators

Partial correlation analysis was used to show a linear relationship between BMI and clinical indicators adjusted for age and sex (Table 2). Because the correlation coefficients r between white blood cell count, absolute neutrophil count, fasting plasma insulin, HOMA-IR, HOMA-β, triglyceride, HDL-C, triglyceride/HDL ratio, alanine transaminase, serum uric acid, systolic blood pressure and BMI were stronger (r>0.3, P<0.05), these linear relationships were further explored for sex differences.

Sex differences in the linear relationships between BMI, HOMA-IR and clinical indicators

Sex differences in the linear relationships between white blood cell count, absolute neutrophil count, fasting plasma insulin, HOMA-IR, HOMA-β, triglyceride, HDL-C, triglyceride/HDL ratio, alanine transaminase, serum uric acid, systolic blood pressure and BMI adjusted for age were analyzed in school-age children with normal BMI and who were overweight and obese. We found the linear relationships between white blood cell count, absolute neutrophil count and BMI that were stronger in girls than in boys who were overweight and obese (Table 3), but these sex differences were not seen in normal BMI children (Supplementary Table 1).

In this study, HOMA-IR was found to be higher in girls who were overweight and obese than in boys who were overweight and obese (P<0.001), with a HOMA-IR in girls of 3.06 (2.12-4.76) and in boys of 2.56 (1.68-3.93). A more pronounced linear relationship was found between white blood cell count, absolute neutrophil count and HOMA-IR in girls who were overweight and obese (Table 4).

Sex differences in the association between HOMA-IR and white blood cell count/absolute neutrophil count

Multiple linear regression analysis showed that HOMA-IR was associated with white blood cell count (β=0.18, P<0.001) and absolute neutrophil count (β=0.15, P<0.001) when adjusted for age in girls who were overweight and obese, but the association was less pronounced in boys who were overweight and obese (Table 5).

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TABLE 1 Clinical characteristics of school-age children with normal BMI and overweight/obesity.

Variables	Normal BMI (n=181)	Overweight/obesity (n=619)	*P*
Sex			
Male	96 (20.25)	378 (79.75)	0.053
Female	85 (26.07)	241 (73.93)	
Age (years)	9.40 (8.70-10.65)	10.20 (9.20-10.90)	<0.001
BMI Z-score	1.63(0.67-2.56)	2.08 (1.40-2.54)	<0.001
White blood cell count (10^9/L)	6.03 (5.23-6.91)	7.15 (6.08-8.44)	<0.001
Absolute neutrophil count (10^9/L)	2.92 (2.32-3.58)	3.79 (3.05-4.73)	<0.001
Absolute lymphocyte counts (10^9/L)	2.53 (2.19-2.95)	2.64 (2.18-3.18)	0.065
Absolute monocyte count (10^9/L)	0.34 (0.28-0.41)	0.38 (0.32-0.47)	<0.001
Absolute eosinophil count (10^9/L)	0.11 (0.07-0.19)	0.13 (0.09-0.21)	0.028
Absolute basophils count (10^9/L)	0.03 (0.02-0.03)	0.03 (0.02-0.04)	0.781
Red blood cell count (10^{12}/L)	4.74 (4.55-4.92)	4.84 (4.65-5.05)	<0.001
Hemoglobin concentration (g/L)	135.00 (129.25-140.00)	136.00 (131.00-141.00)	0.042
Hematocrit (%)	40.75 (39.13-42.08)	41.30 (39.90-42.70)	<0.001
Mean corpuscular volume (fL)	85.70 (83.90-87.88)	85.00 (82.80-87.30)	0.005
Mean corpuscular hemoglobin (pg)	28.40 (27.73-29.10)	28.10 (27.30-28.80)	<0.001
Mean corpuscular hemoglobin concentration (g/L)	332.00 (328.00-335.00)	330.00 (325.00-334.00)	<0.001
Platelet count (10^9/L)	287.00 (248.25-323.00)	312.00 (274.00-354.25)	<0.001
Neutrophil-to-lymphocyte ratio	1.16 (0.91-1.44)	1.42 (1.11-1.81)	<0.001
Platelet-to-lymphocyte ratio	114.79 (93.39-136.48)	119.25 (100.32-141.90)	0.004
Fasting plasma insulin (mIU/L)	5.81 (4.14-8.15)	12.38 (8.23-19.16)	<0.001
Fasting plasma glucose (mmol/L)	4.93 (4.73-5.21)	4.98 (4.75-5.29)	0.227
HOMA-IR	1.28 (0.90-1.88)	2.74 (1.80-4.25)	<0.001
HOMA-β	78.59 (59.15-112.49)	164.53 (111.57-270.58)	<0.001
Total cholesterol (mmol/L)	3.84 (3.39-4.29)	4.08 (3.63-4.56)	<0.001
Triglyceride (mmol/L)	0.81 (0.62-1.08)	1.13 (0.84-1.53)	<0.001
High-density lipoprotein cholesterol (mmol/L)	1.38 (1.24-1.57)	1.22 (1.09-1.37)	<0.001
Low-density lipoprotein cholesterol (mmol/L)	2.23 (1.92-2.51)	2.41 (2.13-2.78)	<0.001
Triglyceride/HDL ratio	0.57 (0.43-0.75)	0.94 (0.64-1.35)	<0.001
Total bilirubin (μmol/L)	10.99 (9.08-13.52)	10.01 (8.20-12.53)	0.001
Aspartate transaminase (U/L)	22.85 (20.20-26.10)	23.00 (19.30-28.10)	0.697
Alanine transaminase (U/L)	12.25 (10.33-15.60)	18.30 (13.60-31.30)	<0.001
Blood urea nitrogen (mmol/L)	4.07 (3.48-4.97)	4.03 (3.33-4.70)	0.314
Serum creatinine (μmol/L)	43.90 (40.53-48.50)	44.05 (40.50-48.10)	0.928
Serum uric acid (μmol/L)	298.17 (255.15-353.71)	366.24 (316.76-423.21)	<0.001
C-reactive protein (mg/L)	4.08 (3.80-4.37)	4.39 (2.21-4.47)	0.001
Systolic blood pressure (mmHg)	101.33 (95.67-107.33)	109.00 (102.67-116.25)	<0.001
Diastolic blood pressure (mmHg)	70.33 (66.33-74.33)	72.33 (67.42-77.67)	0.001

BMI, body mass index; HOMA-β, homeostasis model assessment of beta-cell function; HOMA-IR, homeostasis model assessment of insulin resistance.

Discussion

We determined if sex differences existed in insulin resistance-induced changes in metabolic and inflammatory markers in children who were overweight and obese through data from physical measurements and haematological tests of 800 school-age children in this study. We found that HOMA-IR was more strongly associated with increased white blood cell count and absolute neutrophil count in school-aged girls who were overweight and obese.

Insulin resistance is defined in physiological terms as requiring higher concentrations of insulin to trigger the physiological effects formerly induced by lower concentrations. Obesity is the leading cause of insulin resistance in children, and insulin resistance is closely associated with multiple cardiovascular risk factors and metabolic disorders, such as

TABLE 2 Partial correlation analysis of BMI and clinical indicators adjusted for age and sex in all children.

Variables	BMI	
	r	*P*
White blood cell count (10^9/L)	0.32	<0.001
Absolute neutrophil count (10^9/L)	0.31	<0.001
Absolute monocyte count (10^9/L)	0.26	<0.001
Absolute eosinophil count (10^9/L)	0.003	0.926
Red blood cell count (10^{12}/L)	0.20	<0.001
Hemoglobin concentration (g/L)	0.06	0.107
Hematocrit (%)	0.14	<0.001
Mean corpuscular volume (fL)	-0.20	<0.001
Mean corpuscular hemoglobin (pg)	-0.18	<0.001
Mean corpuscular hemoglobin concentration (g/L)	-0.19	<0.001
Platelet count (10^9/L)	0.26	<0.001
Neutrophil-to-lymphocyte ratio	0.16	<0.001
Platelet-to-lymphocyte ratio	0.06	0.097
Fasting plasma insulin (mIU/L)	0.54	<0.001
HOMA-IR	0.50	<0.001
HOMA-β	0.56	<0.001
Total cholesterol (mmol/L)	0.14	<0.001
Triglyceride (mmol/L)	0.37	<0.001
High-density lipoprotein cholesterol (mmol/L)	-0.37	<0.001
Low-density lipoprotein cholesterol (mmol/L)	0.21	<0.001
Triglyceride/HDL ratio	0.41	<0.001
Total bilirubin (μmol/L)	-0.14	<0.001
Alanine transaminase (U/L)	0.34	<0.001
Serum uric acid (μmol/L)	0.44	<0.001
C-reactive protein (mg/L)	0.10	0.006
Systolic blood pressure (mmHg)	0.40	<0.001
Diastolic blood pressure (mmHg)	0.22	<0.001

BMI, body mass index; HOMA-β, homeostasis model assessment of beta-cell function; HOMA-IR, homeostasis model assessment of insulin resistance.

dyslipidemia, impaired glucose tolerance, type 2 diabetes, hyperuricemia, and elevated transaminases. Currently, there is no universally accepted definition of insulin resistance because there is no standardized analytical method for measuring plasma insulin. The "gold standard" method for measuring systemic insulin sensitivity is the euglycemic-hyperinsulinemic clamp (16). However, due to the complexity of the procedure, this methodology is used only in scientific research but not in clinical application. So HOMA-IR has been established and widely used as a substitute indicator of whole body insulin resistance (17).

In addition to routine blood tests and metabolism-related indicators, this study also assessed the association of novel markers with insulin resistance, such as C-reactive protein, neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio and triglyceride/HDL ratio. Fasting insulin levels are higher in children who were overweight and obese when compared to subjects with normal BMI, and the levels of HOMA-IR and HOMA-B were also higher. By partial correlation analysis, white blood cell count and absolute neutrophil count were more strongly correlated with BMI and HOMA-IR in girls who were overweight and obese. Subsequently, multiple linear regression analysis also demonstrated that HOMA-IR significantly increased white blood cell counts and absolute neutrophil count in girls who were overweight and obese.

Men and women have different energy needs, and there are sex differences in human metabolism (18). The biological differences between men and women lead to different physiological responses to exercise, including height, weight, fat mass, lean muscle mass and hormone levels. During prolonged exercise, women showed a greater ability to oxidize lipids as a fuel source, while men oxidized more protein and carbohydrates (19). Adipose tissue, considered a major storage site for excess energy, is now recognized as an endocrine organcapable of producing and releasing bioactive compounds

TABLE 3 Sex differences in partial correlation analysis of BMI and clinical indicators adjusted for age in children with overweight and obesity.

Variables	Boy's BMI (n=378)		Girl's BMI (n=241)	
	r	*P*	*r*	*P*
White blood cell count (10^9/L)	0.17	0.001	0.37	<0.001
Absolute neutrophil count (10^9/L)	0.14	0.007	0.35	<0.001
Fasting plasma insulin (mIU/L)	0.47	<0.001	0.48	<0.001
HOMA-IR	0.43	<0.001	0.44	<0.001
HOMA-β	0.49	<0.001	0.52	<0.001
Triglyceride (mmol/L)	0.28	<0.001	0.29	<0.001
High-density lipoprotein cholesterol (mmol/L)	-0.32	<0.001	-0.28	<0.001
Triglyceride/HDL ratio	0.33	<0.001	0.32	<0.001
Alanine transaminase (U/L)	0.31	<0.001	0.35	<0.001
Serum uric acid (μmol/L)	0.35	<0.001	0.46	<0.001
Systolic blood pressure (mmHg)	0.33	<0.001	0.38	<0.001

BMI, body mass index; HOMA-β, homeostasis model assessment of beta-cell function; HOMA-IR, homeostasis model assessment of insulin resistance.

TABLE 4 Sex differences in partial correlation analysis of HOMA-IR and clinical indicators adjusted for age in children with overweight and obesity.

Variables	Boy's HOMA-IR (n=378)		Girl's HOMA-IR (n=241)	
	r	*P*	*r*	*P*
White blood cell count (10^9/L)	0.12	0.025	0.27	<0.001
Absolute neutrophil count (10^9/L)	0.09	0.071	0.29	<0.001
Triglyceride (mmol/L)	0.31	<0.001	0.25	<0.001
High-density lipoprotein cholesterol (mmol/L)	-0.10	0.043	-0.13	0.039
Triglyceride/HDL ratio	0.27	<0.001	0.22	0.001
Alanine transaminase (U/L)	0.23	<0.001	0.23	<0.001
Serum uric acid (μmol/L)	0.17	0.001	0.14	0.028
Systolic blood pressure (mmHg)	0.29	<0.001	0.32	<0.001

HOMA-IR, homeostasis model assessment of insulin resistance.

TABLE 5 Sex differences in multiple linear regression analysis of HOMA-IR and white blood cell count/absolute neutrophil count adjusted for age in children with overweight and obesity.

Independent variable	White blood cell count (10^9/L)	Absolute neutrophil count (10^9/L)
	β (95%CI), *P*	*β* (95%CI), *P*
Male (n=378)		
HOMA-IR	0.09 (0.01-0.16), 0.024	0.05 (-0.004-0.11), 0.069
Age	-0.04 (-0.22-0.15), 0.711	0.01 (-0.14-0.16), 0.892
Female (n=241)		
HOMA-IR	0.18 (0.10-0.27), <0.001	0.15 (0.09-0.21), <0.001
Age	-0.10 (-0.30–0.11), 0.349	0.04 (-0.11-0.19), 0.643

HOMA-IR, homeostasis model assessment of insulin resistance.

involved in chronic inflammatory and pathological metabolic processes associated with obesity (20). Recent evidence have shown that excess adipose tissue is tightly associated with increased adipokine release, immune cell infiltration, and the progress of low-grade systemic inflammation from childhood to adulthood (21). Multiple studies have shown that the major cellular component of adipose tissue is adipocytes, which are sustained by an extracellular matrix interspersed with preadipocytes, fibroblasts, endothelial cells and immune cells (22, 23). Especially, the strong local presence of leukocytes such as macrophages, mast cells, natural killer cells, neutrophils, monocytes, and T and B lymphocytes led human adipose tissue defined as an immune organ that maintained delicate immune homeostasis (24, 25).

A complete blood count is an inexpensive and readily available blood test. Obesity is associated with hematologic abnormalities (26). Herishanu et al. analyzed 327 patients with persistent leukocytosis in a hematological clinic and found that 15% of the patients were asymptomatic and obese, most of whom were middle-aged females with mild leukocytosis, it is characterized by increased neutrophilia with elevated acute-phase reactants (C-reactive protein and erythrocyte sedimentation rate) (27). Raghavan et al. similarly noted that BMI was associated with white blood cell count and neutrophil count within the physiological range in obese women (28). Obesity-related leukocytosis is significantly predominant in women, and the etiology of this leukocytosis may be multifactorial. Sex-specific pathways of inflammation that affect obesity and metabolic syndrome have been identified. In patients with metabolic syndrome, women have lower concentrations of anti-inflammatory adiponectin. However, in men, metabolic syndrome is associated with increased monocyte-derived circulating cytokines (mainly IL-6) and hyperresponsive circulating immune cells (29). In addition, in females, inflammation may be limited by estrogen (30). Importantly, we found that in school-aged girls who were overweight and obese, increased white blood cell count and absolute neutrophil count were strongly associated with increased HOMA-IR.

There are some limitations in the current study, including the cross-sectional study design, the lack of assessment of a wider range of inflammatory biomarkers, such as IL-6 and TNF-α, and the physical changes of secondary sexual characteristics during puberty were not evaluated.

Conclusions

Although adipose tissue-induced inflammation is low-grade, it has a negative effect on distal organ function through insulin resistance, which may be responsible for complications associated with obesity. Our findings indicated that elevated white blood cell count and absolute neutrophil count in children who were overweight and obese, and especially in girls, can serve as markers of insulin resistance. In the future, further metabolomics and proteomics experiments may be able to explain the mechanism of insulin resistance in children with obesity.

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 Ethics Committee of Henan University. Written informed consent to participate in this study was provided by the participants' legal guardian/next of kin.

Author contributions

LZ and H-MH contributed to the design and the data analysis of the study, and drafted the manuscript. NQ, Z-TZ, KZ, YL, H-BC, and J-NX collected and managed data. All authors approved the final article and approved the submitted version.

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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

Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fendo.2022.1041761/full#supplementary-material

References

1. Jebeile H, Kelly AS, O'Malley G, Baur LA. Obesity in children and adolescents: Epidemiology, causes, assessment, and management. *Lancet Diabetes Endocrinol* (2022) 10(5):351–65. doi: 10.1016/S2213-8587(22)00047-X

2. Wang Y, Zhao L, Gao L, Pan A, Xue H. Health policy and public health implications of obesity in China. *Lancet Diabetes Endocrinol* (2021) 9(7):446–61. doi: 10.1016/S2213-8587(21)00118-2

3. Ighbariya A, Weiss R. Insulin resistance, prediabetes, metabolic syndrome: What should every pediatrician know? *J Clin Res Pediatr Endocrinol* (2017) 9(Suppl 2):49–57. doi: 10.4274/jcrpe.2017.S005

4. Reaven GM. Pathophysiology of insulin resistance in human disease. *Physiol Rev* (1995) 75(3):473–86. doi: 10.1152/physrev.1995.75.3.473

5. Reaven G. Metabolic syndrome: Pathophysiology and implications for management of cardiovascular disease. *Circulation* (2002) 106(3):286–8. doi: 10.1161/01.cir.0000019884.36724.d9

6. Vekic J, Zeljkovic A, Stefanovic A, Jelic-Ivanovic Z, Spasojevic-Kalimanovska V. Obesity and dyslipidemia. *Metabolism* (2019) 92:71–81. doi: 10.1016/j.metabol.2018.11.005

7. Yaribeygi H, Farrokhi FR, Butler AE, Sahebkar A. Insulin resistance: Review of the underlying molecular mechanisms. *J Cell Physiol* (2019) 234(6):8152–61. doi: 10.1002/jcp.27603

8. Gasmi A, Noor S, Menzel A, Doşa A, Pivina L, Bjorklund G. Obesity and insulin resistance: Associations with chronic inflammation, genetic and epigenetic factors. *Curr Med Chem* (2021) 28(4):800–26. doi: 10.2174/0929867327666200824112056

9. Group of China obesity task force. Body mass index reference norm for screening overweight and obese in Chinese children and adolescents. *Zhonghua Liu Xing Bing Xue Za Zhi.* (2004) 25(2):97–102.

10. de Onis M, Onyango AW, Borghi E, Siyam A, Nishida C, Siekmann J. Development of a WHO growth reference for school-aged children and adolescents. *Bull World Health Organ.* (2007) 85(9):660–7. doi: 10.2471/blt.07.043497

11. Monasor-Ortolá D, Quesada-Rico JA, Nso-Roca AP, Rizo-Baeza M, Cortés-Castell E, Martínez-Segura A, et al. Degree of accuracy of the BMI z-score to determine excess fat mass using DXA in children and adolescents. *Int J Environ Res Public Health* (2021) 18(22):12114. doi: 10.3390/ijerph182212114

12. 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

13. Rodríguez-Rodríguez E, López-Sobaler AM, Ortega RM, Delgado-Losada ML, López-Parra AM, Aparicio A. Association between neutrophil-to-Lymphocyte ratio with abdominal obesity and healthy eating index in a representative older Spanish population. *Nutrients* (2020) 12(3):855. doi: 10.3390/nu12030855

14. Rodríguez-Rodríguez E, Salas-González MD, Ortega RM, López-Sobaler AM. Leukocytes and neutrophil-lymphocyte ratio as indicators of insulin resistance in Overweight/Obese school-children. *Front Nutr* (2022) 8:811081. doi: 10.3389/fnut.2021.811081

15. von Bibra H, Saha S, Hapfelmeier A, Müller G, Schwarz PEH. Impact of the Triglyceride/High-density lipoprotein cholesterol ratio and the hypertriglyceremic-waist phenotype to predict the metabolic syndrome and insulin resistance. *Horm Metab Res* (2017) 49(7):542–9. doi: 10.1055/s-0043-107782

16. 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

17. Minh HV, Tien HA, Sinh CT, Thang DC, Chen CH, Tay JC, et al. Assessment of preferred methods to measure insulin resistance in Asian patients with hypertension. *J Clin Hypertens (Greenwich).* (2021) 23(3):529–37. doi: 10.1111/jch.14155

18. Brennan L, Gibbons H. Sex matters: A focus on the impact of biological sex on metabolomic profiles and dietary interventions. *Proc Nutr Soc* (2020) 79(2):205–9. doi: 10.1017/S002966511900106X

19. Bassett AJ, Ahlmen A, Rosendorf JM, Romeo AA, Erickson BJ, Bishop ME. The biology of sex and sport. *JBJS Rev* (2020) 8(3):e0140. doi: 10.2106/JBJS.RVW.19.00140

20. Calcaterra V, Vandoni M, Rossi V, Berardo C, Grazi R, Cordaro E, et al. Use of physical activity and exercise to reduce inflammation in children and adolescents with obesity. *Int J Environ Res Public Health* (2022) 19(11):6908. doi: 10.3390/nu11081788

21. Kumar S, Kelly AS. Review of childhood obesity: From epidemiology, etiology, and comorbidities to clinical assessment and treatment. *Mayo Clin Proc* (2017) 92(2):251–65. doi: 10.1016/j.mayocp.2016.09.017

22. Qi Y, Hui X. The shades of grey in adipose tissue reprogramming. *Biosci Rep* (2022) 42(3):BSR20212358. doi: 10.1042/BSR20212358

23. Rakotoarivelo V, Variya B, Ilangumaran S, Langlois MF, Ramanathan S. Inflammation in human adipose tissues-shades of Gray, rather than white and brown. *Cytokine Growth Factor Rev* (2018) 44:28–37. doi: 10.1016/j.cytogfr.2018.10.001

24. Kawai T, Autieri MV, Scalia R. Adipose tissue inflammation and metabolic dysfunction in obesity. *Am J Physiol Cell Physiol* (2021) 320(3):C375–91. doi: 10.1152/ajpcell.00379.2020

25. Russo L, Lumeng CN. Properties and functions of adipose tissue macrophages in obesity. *Immunology* (2018) 155(4):407–17. doi: 10.1111/imm.13002

26. Purdy JC, Shatzel JJ. The hematologic consequences of obesity. *Eur J Haematol* (2021) 106(3):306–19. doi: 10.1111/ejh.13560

27. Herishanu Y, Rogowski O, Polliack A, Marilus R. Leukocytosis in obese individuals: Possible link in patients with unexplained persistent neutrophilia. *Eur J Haematol* (2006) 76(6):516–20. doi: 10.1111/j.1600-0609.2006.00658.x

28. Raghavan V, Gunasekar D, Rao KR. Relevance of haematologic parameters in obese women with or without metabolic syndrome. *J Clin Diagn Res* (2016) 10 (5):EC11–6. doi: 10.7860/JCDR/2016/18779.7732

29. Ter Horst R, van den Munckhof ICL, Schraa K, Aguirre-Gamboa R, Jaeger M, Smeekens SP, et al. Sex-specific regulation of inflammation and metabolic syndrome in obesity. *Arterioscler Thromb Vasc Biol* (2020) 40(7):1787–800. doi: 10.1161/ATVBAHA.120.314508

30. Vasanthakumar A, Chisanga D, Blume J, Gloury R, Britt K, Henstridge DC, et al. Sex-specific adipose tissue imprinting of regulatory T cells. *Nature* (2020) 579 (7800):581–5. doi: 10.1038/s41586-020-2040-3

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Childhood obesity and central precocious puberty

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Childhood obesity is a major public health problem worldwide, and the relationship between obesity and central precocious puberty has long been confirmed, however, the mechanisms underlying this association remain elusive. This review provides an overview of the recent progress regarding how childhood obesity impacts on hypothalamic-pituitary-gonadal axis and pubertal onset, focusing on adipokines (leptin and ghrelin), hormone (insulin), and lipid (ceramide), as well as critical signaling pathways (AMPK/SIRT, mTOR) that integrate the peripheral metabolism and central circuits. Notably, prevention of obesity and CPP is beneficial for the adult life of the children, thus we further summarize the potential strategies in treating and preventing childhood obesity and CPP. The updated understanding of metabolic stress and pediatric endocrine disease will arise the attention of society, and also contribute to preventing more serious comorbidities in the later period of life in children.

KEYWORDS

childhood obesity, central precocious puberty, metabolic status, integration, prevention strategies

Introduction

Precocious puberty refers to the early onset of puberty, manifests as early secondary sexual characteristics and physical development, and is a pediatric endocrine disease. Precocious puberty can be mainly classified as central precocious puberty (CPP) and peripheral precocious puberty (PPP) according to whether the hypothalamic-pituitary-gonadal (HPG) axis is activated. The initiation of the HPG axis is usually considered true precocious puberty, therefore, CPP is the dominant diagnosis. The development of secondary sexual characteristics before the age of 8 in girls and 9 in boys is defined as CPP. CPP shows an obvious gender dimorphism, which is a conserved feature of puberty in higher mammals, possibly due to environmental and psychological factors. A recent systematic review and meta-analysis indicated that pooled prevalence is 25% in girls less than 6 years of age, although significant heterogeneity exists in different age groups (1). Females have a prominent population of Kiss1 neurons in the anterior ventral periventricular nucleus, which is crucial

for establishing the positive feedback between ovarian steroids and the gonadotropin-releasing hormone (GnRH) surge generator, facilitating the females more sensitive to the central regulatory effects of some metabolic signals than in males (2, 3).

Detailed descriptions of the HPG axis in controlling puberty can be found in many excellent reviews. In brief, GnRH neurons in the hypothalamus release GnRH pulse to the hypophyseal portal blood system, while the gonadotroph cells in the pituitary respond to the signal and release the gonadotrophins luteinizing hormone (LH) and follicle-stimulating hormone (FSH), which reach the gonads to drive puberty onwards. Therefore, the evaluation of LH peaks after GnRH testing is the gold standard in the biochemical diagnosis of CPP. The combined detection of secondary sexual characters, bone age, hormone levels, pelvic ultrasound and cranial MRI could increase the sensitivity and accuracy for CPP diagnosis (4, 5).

The development of CPP is both determined by genetic and environmental factors. The past decade has confirmed a series of core genes of CPP, such as the kisspeptin gene (KISS1), the kisspeptin receptor gene (KISS1R), the makorin ring finger protein 3 (MKRN3), and the Delta-like non-canonical Notch ligand 1 (DLK1). It is reported that DLK1 gene mutation is associated with the prevalence of family CPP, and MKRN3 gene loss-of-function mutations are the most prevalent genetic etiology of CPP. Apart from rare genetic variants, the occurrence of metabolic comorbidities rises as the dominant cause of CPP in recent years. The epidemic of childhood obesity serves as the major companion for CPP, suggesting the impact of nutritional and metabolic cues on the HPG axis. There is clear evidence to support the effects of higher childhood body mass index (BMI) on the onset and development of puberty in both boys and girls (6). Childhood obesity and PP are both important risk factors for metabolic syndrome, type 2 diabetes, and insulin resistance (7). Genome-wide association study (GWAS) in humans identified multiple BMI-increasing alleles that are also associated with earlier age of menarche, further confirming genetic co-regulation of childhood obesity and CPP (8). Notably, early timing of puberty directly affects the adult height, muscle, and fat mass accrual, induces various psychological symptoms, conduct problems, and is also linked to an increased risk of cardiovascular events, breast cancer, higher susceptibility to attention deficit-hyperactivity disorder ADHD (9), as well as various oncologic, cognitive, and behavioral disorders, and even reduced life expectancy (10–12). Therefore, PP has raised global public health concerns, and strategies that prevent PP are highlighted worldwide.

Metabolic basics of CPP

Reproduction is an energy-intensive, but essential process for the survival of all species. Therefore, a certain threshold of body fat storage is required to initiate and maintain reproductive function. Overweight women have served as fecundity icons dating from *Palaeolithic* Ages. Whereas individuals with anorexia or malnutrition are often related to delay or absence of puberty onset and perturbed fertility (13). The current generation of adolescents is growing up at a time of unprecedented ample food, whereby nutritional deficiency and food insecurity are eliminated, and overweight and obesity are burgeoning. Nutrition overwhelming in modern society not only causes metabolic burden, but also accelerates the process of puberty. Studies showed that BMI could affect the GnRH stimulation test, and the peak of LH stimulation is negatively correlated with BMI (14, 15).

The metabolic-reproductive interplay emphasizes the link between body energy reserves and reproductive function, and lays the basis for the impact of different metabolic disorders, ranging from anorexia to obesity and metabolic syndrome, on puberty and fertility. Hypothalamic circuits are responsible for the tight coupling between body energy status and puberty onset. Among these circuits, GnRH neurons operate as the final output pathway for the central control of the onset of puberty, and a plethora of metabolic hormones and neuropeptides are coupled and tightly regulated the function of GnRH neurons. Elimination of the inhibition on the HPG axis is dynamically balanced before the initiation of puberty. Interaction of kisspeptin and kisspeptin receptor, synchronized operation of Neurokinin-B, glutamate, leptin, and androgens are considered to be drivers of GnRH pulse generation, whereas endogenous opioid peptides such as dynorphin A, gamma-aminobutyric acid (GABA), MKRN3 are acted as inhibitors of GnRH release (16).

Clinical and experimental work further provide evidence to support the interaction of energy reserves and CPP. The age of menarche was approximately 17 years in the early 19th century, while it dramatically declined to approximately 13 years by the mid-20th century, largely attributed to improved nutrition, personal hygiene, and better socioeconomic conditions (6). Studies in different regions all reported that increased BMI is a predictor of early onset of puberty in girls (17–20). Notably, body composition is found to be more sensitive to earlier puberty onset than BMI (21). Furthermore, higher birthweight is also a predictor of younger ages at menarche and advanced breast development, and gestational glucose intolerance is associated with increased odds of offspring overweight/obesity in late adolescence (22, 23). Notably, children born small for gestational age are also prone to develop obesity, metabolic syndrome, as well as CPP, which might be explained by thrifty phenotype hypothesis (24, 25). Collectively, these findings further confirmed the link of metabolic status and CPP.

Effects of adipokines and hormones on CPP

Obesity is accompanied by a series of metabolic alterations, and different metabolic cytokines and hormones, such as leptin,

ghrelin, insulin, as well as certain central lipids may impact the HPG axis, and participate in the fine-tuning of puberty.

Leptin and ghrelin

Leptin is the first identified adipose cytokine, which is a peptide hormone (16kDa) encoded by the product of the obese (*ob*) gene, and secreted from the adipocytes into the circulation, the level of leptin is directly related to the amount of body fat stores. Leptin can pass the blood-brain barrier, and take the action at certain neurons in the hypothalamus. Physiologically, circulating leptin is a sensitive maker of metabolic status, and transports the signal to the control center (hypothalamus). Upon binding with leptin receptor (LepRb), leptin activates pathways such as JAK2/STAT3, PI3K/IRS/AKT, and SHP2/MAPK in the hypothalamus, and exerts anorexigenic and thermogenic functions to alleviate the metabolic burden in the peripheral. Simultaneously, the accumulation of phosphorylated STAT3 dimers induces the transcription of SOCS3, which inhibits the JAK2/STAT3 pathway. This efficient work of the leptin signaling feedback loop guarantees metabolic homeostasis. Accordingly, leptin deficiency (*ob/ob* mice) and leptin receptor deficiency (*db/db* mice) animals spontaneously develop into obesity and/or type 2 diabetes.

In addition to metabolic control, leptin is also a permissive factor for the activation of GnRH neurosecretion at puberty. Kisspeptins that are produced by hypothalamic Kiss1 neurons are fundamental GnRH regulators, and leptin deficiency has been found to decrease hypothalamic Kiss1 expression, whereas exogenous administration of leptin increases Kiss1 in rodent models of leptin deficiency (26). The clinical investigation reported that the serum leptin level is obviously higher in CPP girls than in the controls (27). Consistently, individuals with malnutrition have low levels of leptin with a delay in puberty onset. Leptin acts as the upstream afferent signal for GnRH neurosecretion, whereas GnRH neurons that lack functional LepRb require alternative afferent pathways to perform their regulatory actions.

Obese individuals exhibit high leptin concentrations due to the adipose tissue expansion, however, the high leptin concentrations failed to obtain the expected suppression of food intake and increased energy expenditure, a phenomenon that is termed leptin resistance (28). Leptin resistance is a hallmark of obesity, featuring a high concentration of circulating and central leptin. The lack of feedback of leptin signal in leptin resistance might persistently stimulate kiss1 expression, and destroy the balance of NKB and Dyn modulation on kisspeptin secretion (29).

Contrary to the anorectic action of leptin, the gut-derived peptide ghrelin is required for the orexigenic process. In both mice and humans, increased appetite is correlated with elevated levels of circulating ghrelin. Ghrelin directly acts upon the HPG axis by stimulating adreno-cortico-tropic-hormone (ACTH) synthesis and secretion in the anterior pituitary, or indirectly elevates ACTH through paracrine stimulation of hypothalamic corticotropin-releasing hormone synthesis and secretion. It is reported that plasma ghrelin is negatively correlated with BMI and body fat percentage, and circulating ghrelin levels are decreased in human obesity (30). Consistently, a progressive reduction in ghrelin levels has been observed during puberty, and GnRH analog (GnRHa) treatment in CPP girls further decreases the circulating ghrelin levels (31). However, the reason why ghrelin secretion decreases at puberty is not yet known.

Every other day fasting (EODF) is reported to delay the Di-(2-ethylhexyl) phthalate-induced puberty onset acceleration in female rats, accompanied by the decrease of serum leptin, luteinizing hormone and estradiol (32). Considering the locations of the central action of leptin and the Kiss1 neurons are consecutive and possibly overlapped, the accumulation of metabolic hormone in the hypothalamus might be an important integrator to explain childhood obesity and CPP.

Insulin

Insulin is another important hormone that affects both childhood obesity and CPP. Insulin is a pancreatic hormone, and secreted in response to the increase of blood concentration of glucose (hyperglycemia), to promote circulating glucose to enter effector cells. Reduced sensitivity of cells to insulin is termed insulin resistance, which often occurs in obesity and related T2DM. The inefficient glucose uptake in conditions of insulin resistance causes hyperglycemia, which further stimulates insulin secretion to compensate the insulin insensitivity. However, continuous hyperglycemia and insulin resistance in obesity has been shown to conduct stimulatory/permissive actions on the HPG axis. Actually, conditions of low or null insulin levels, such as uncontrolled diabetes, are usually associated with suppressed GnRH levels and reproductive activity (33). Mice lacking insulin receptors selectively in neurons are obese and show a delay in development due to GnRH deficiency (34). On the contrary, high insulin levels in female rodents and women show significantly increased LH secretion (35, 36).

During development, nutrient consumption promotes growth as well as the production of required hormones through insulin-like systems (37). The hormonal profile in peri-pubertal girls with obesity is characterized by hyperinsulinemia and higher HOMA-IR index, especially evident in early puberty (38). Upon obesity and insulin resistance, the compensated insulin secretion might accelerate the timing of puberty. In a cross-sectional study that included 79 girls with CPP and 37 girls with premature thelarche, Li et al. found that the serum insulin-like growth factor-1 (IGF-1), IGF binding protein-3 (IGFBP-3) levels are obviously a higher in

CPP girls compared with controls (39). Metformin is a widely used drug for treating T2DM and is also used for delaying sexual maturation in girls with CPP. Heterogeneous mice (UM-HET3) that were treated with metformin (i.p) between the ages of 15 and 56 days showed increased insulin sensitivity and normal sexual maturation in female pups, indicating insulin signaling and puberty are tightly integrated (40). Insulin also stimulates the synthesis of leptin in adipocytes, the two hormones might synergistically modulate puberty onset.

Molecular link of metabolic and reproductive circuits on CPP

The identification of *Kiss1*, and its receptor Gpr54 is considered to be a breakthrough in puberty. *Kiss1* governs the secretion of kisspeptin, which directly acts on GnRH neurons *in promoting* puberty activation. The Kiss1 system is sensitive to metabolic conditions, and an important transmitter for impacting puberty. Therefore, hypothalamic kisspeptin neurons have been postulated to be a key nodal nexus between metabolism and puberty (41). Childhood obesity is usually associated with an active Kiss1 system, and a series of molecular substrates are involved in this process.

AMPK/SIRT signaling

AMPK is a fundamental nutrition sensor that is essential for cellular energy homeostasis, alteration of cellular AMP/ATP ratio determines the status of AMPK. Upon energy deficiency, an increased AMP/ATP ratio directly activates AMPK *via* phosphorylation at Thr-172 of the α-subunit, attempting to restore the balance. Exercise is an efficient way to deplete energy, and the AMPK is kept in active status during exercise. In contrast, an overwhelming energy supply (including childhood obesity) will inhibit AMPK activity.

AMPK is widely expressed in metabolic organs and also co-expressed in Kiss 1 neurons, therefore the brain AMPK is likely to integrate metabolic/nutritional status and the onset of puberty in obesity. Chronic energy deficiency at puberty activates hypothalamic AMPK, and pharmacological or virogenetic activation of AMPK delays pubertal onset to a variable extent in rodent models (42). AMPK is found to inhibit the Kiss 1 gene, thus suppressing the function of GnRH neurons (43). On the contrary, childhood obesity inhibits AMPK activity (44), which might relieve the suppression of the Kiss 1 gene, and contribute to the development of CPP. Hypothalamic activation of AMPK along with the persistence of the repressive action of SIRT1 at the Kiss1 promoter, leads to reduced Kiss1 expression, whereas the eviction of SIRT1 from the Kiss1 promoter as well as AMPK suppression in conditions of overnutrition, transactivates Kiss 1 gene transcription and the GnRH secretion (45, 46).

Central AMPK activity also regulates peripheral metabolism. Estrogens (E2) are reported to inhibit hypothalamic AMPK through estrogen receptor alpha (ERα), which leads to the promotion of thermogenesis in brown adipose tissue in a feeding-independent manner. Genetic activation of AMPK in the VMH prevented E2-induced increase in brown adipose tissue-mediated thermogenesis and weight loss (47).

mTOR signaling

Mammalian target of rapamycin (mTOR) is atypical serine/threonine protein kinase, and is an evolutionally conserved protein. mTOR can integrate multiple intracellular signals (nutrition, energy, and growth factors), and is involved in the transcriptional and translational regulating processes.

In conditions of energy sufficiency, timely eviction of SIRT1 from the Kiss1 promoter, together with the presumable activation of mammalian target of rapamycin (mTOR), allows increased Kiss1 expression and the normal occurrence of puberty. Activation of PI3K/Akt/mTOR pathway in the hypothalamus is associated with the increased GnRH release and CPP in adolescent female rats, whereas inhibition of mTOR in the hypothalamus could block the activation of Kiss1, Grp54, and GnRH (48). In addition, activation of mTOR also induces Akt phosphorylation and kisspeptin release, which also contributes to GnRH secretion and CPP (49). The central concentration of kisspeptin also affects the mTOR pathway, it is reported that Kisspeptin 10 (Kp-10) maintains the activation of mTOR signaling (50). Accordingly, pharmacological inhibition of kisspeptin weakens mTOR pathway (51). Therefore, the interaction of mTOR and kisspeptin might further drive the initiation of CPP.

Hypothalamic ceremide

A central ceramide signaling pathway serves as an alternative mediator of childhood obesity and CPP. Different from kisspeptin's direct regulation of GnRH neurons, hypothalamic ceramide involves the paraventricular nucleus (PVN) and sympathetic ovarian innervation. Obese female rats with CPP show higher expression of serine palmitoyltransferase long-chain base subunit 1 (SPTLC1), a crucial component for *de novo* ceramide synthesis, the increased ceramide level further results ovarian sympathetic output, whereas blockade of ceramide synthesis normalized the timing of puberty and ovarian sympathetic tone (52). Consistently, another report revealed that early-onset obesity enhanced ceramide synthesis in PVN, which accelerates the maturation of the ovarian noradrenergic system, virogenetic suppression of SPTLC1 that inhibits ceremide synthesis, in turn reverses obesity-induced

CPP, indicating that central ceremide is crucial in integrating metabolic and neuronal circuits in CPP (53).

Treatment and prevention strategies for CPP

Puberty is a crucial biological process normally occurring at a specific time, therefore, the aim of CPP treatment is to normalize the course of puberty, preserve the adult height, and alleviate the associated complications. GnRH secretion is considered the initial drive for the development of CPP, therefore, GnRH analog (GnRHa) is the primary option for CPP. GnRHa treatment shows efficacy in suppressing gonadotrophin and slowing the progression of secondary sexual characteristics (54). However, whether the GnRHa treatment benefit adult height is still in conflict, and treatment with GnRHa has different effects on BMI according to baseline body composition (55). Patients with CPP are frequently obese due to hormonal and metabolic changes, and a decrease in BMI has been reported in these patients during GnRHa therapy (56). A study tracked 92 adult females that implemented GnRHa treatment in childhood, and found that these subjects had normal BMI and body composition, although the final height is not increased consistently (57). Another study evaluated the effects of GnRHa treatment in 94 girls with idiopathic CPP, and reported increased insulin resistance but normal BMI and lipid profile over 2 years after menarche (58). On the contrary, a study has followed up body composition longitudinally in girls show a gradual increase in adiposity, a decrease in muscle mass, and bone mineral density during GnRHa treatment, whereas bone mass was preserved after treatment (59). In patients with hypothalamic hamartoma, GnRHa treatment increases the mean BMI and the percentage of body fat mass in females, a possible reason is that GnRHa treatment causes an increase in appetite and consequently an elevation of the fat mass index in CPP girls (60, 61). Collectively, GnRHa therapy has a favorable effect on CPP, whereas further well-designed longitudinal investigations are needed to evaluate its long-term metabolic outcomes.

Prevention is the first and foremost strategy for CPP and obesity. A healthy lifestyle is crucial for the development of children. Away from "junk food" should be highlighted. Overconsumption of high-fat high-fructose food is the driving force of obesity, especially in children. It is reported that high-fructose corn syrup consumption (e.g., drinks and desserts) during childhood has stronger metabolic effects than in other generations, which is closely associated with CPP (62). An animal study revealed that EODF can delay puberty onset acceleration in female rats (32), indicating appropriate diet restrictions can be taken into consideration. In addition to diet control, exercise is another important approach for the prevention of CPP in children (63). Lack of out-door exercise may affect insulin sensitivity and vitamin D levels. A systematic meta-analysis comparing serum vitamin D levels between patients with CPP and controls revealed that vitamin D-deficient subjects are more likely to develop CPP, suggesting that CPP may be linked to vitamin D deficiency (64). However, a cross-sectional study fails to observe significant differences in serum 25(OH)D concentration between CPP girls and prepubertal controls (65). Although there are inconsistent findings in different reports, a recent trial reports that the frequency of CPP cases increased approximately three times during the COVID-19 pandemic due to prolonged stress, home quarantine, as well less excise (66). CPP and obesity also affect calcium-phosphate metabolism and adult height, therefore, adequate outdoor exercises are strongly recommended, and adequate sunshine and vitamin D are beneficial for children in all aspects.

Guaranteeing the quality and duration of sleep of the children is equally important for preventing obesity and CPP. The circadian timing system regulates a variety of biological actions including metabolism, hormone, immunity, and reproductive function. Sleep quality and duration determine the hypothalamic melatonin secretion, which impacts the transcription of kisspeptin and GnRH (67). In addition, sleep also promotes the secretion of growth hormones and attribute to the prevention of ADHD. Moreover, attention should also be paid to the psychological status of the children, avoiding negative emotions is one of the effective strategies in managing a healthy lifestyle for children.

Exogenous chemicals that interfere with the endocrine system are defined as endocrine disruptors. Endocrine disruptors may affect the synthesis, metabolism, and the action of endogenous hormones, leading to the dysregulation of normal physiological processes and promoting the development of disease (57). Endocrine disruptors are ubiquitous in the environment, and might also be risks for CPP. In a case-control study, Zhou et al. compared urinal metabolites and serum hormones of 30 precocious puberty girls with 46 age- and race-matched prepubertal females, and confirmed the association of CPP with phthalate esters exposure (68). Ubiquitously present bisphenol A is another threat to female CPP, possibly through Kiss1 activation in ARC (69, 70). Children that are exposed to antibiotics, especially fluoroquinolones and tetracyclines are reported to be positively associated with the occurrence of CPP (71). Pesticides that extensively used in farming act as endocrine-disrupting chemicals, and significantly affect the time of puberty onset (72). In addition, pheromones are also considered to promote hormone secretions and induce CPP (73). Collectively, keeping children from this environmental pollution helps prevent CPP and related diseases.

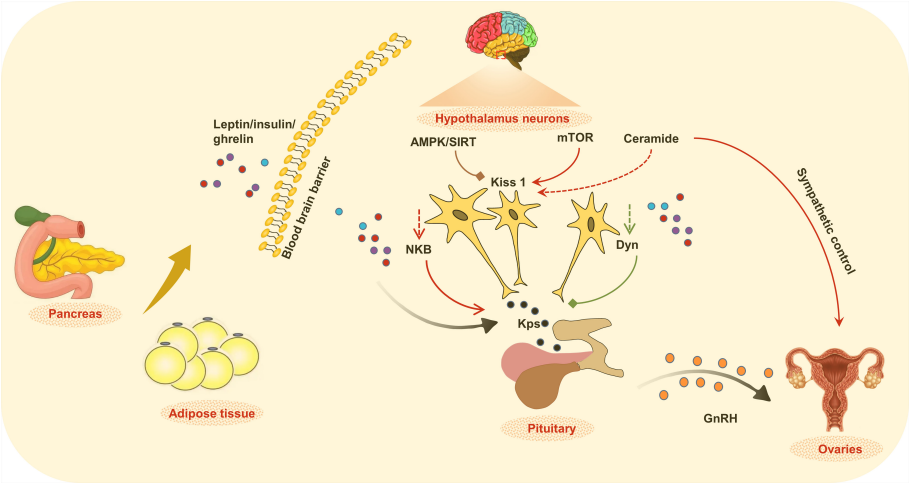

FIGURE 1
The regulation of metabolic and endocrine functions. Alteration of peripheral metabolic status in childhood obesity changed the production and release of cytokines, hormones, lipid, as well as energy sensitive molecules that integrate the control of metabolic signals and central circuits, which together affect the initial of puberty.

Conclusions and perspective

The obesity epidemic and CPP are associated with a series of metabolic and endocrine diseases, and might also impact the quality of life in adults. In this review, we have focused on the peripheral cytokines, hormones, lipids, as well as energy-sensitive molecules to integrating the metabolic and endocrine functions during CPP (Figure 1). Understanding the role of childhood obesity in CPP will draw attention to treating and preventing the disorder in children. Considering the complex interaction of the two systems, and the genetic, psychological and environmental impact on CPP, we propose that prevention should be considered to be the foremost strategy for childhood obesity and CPP (Figure 2). Maintaining physical and mental

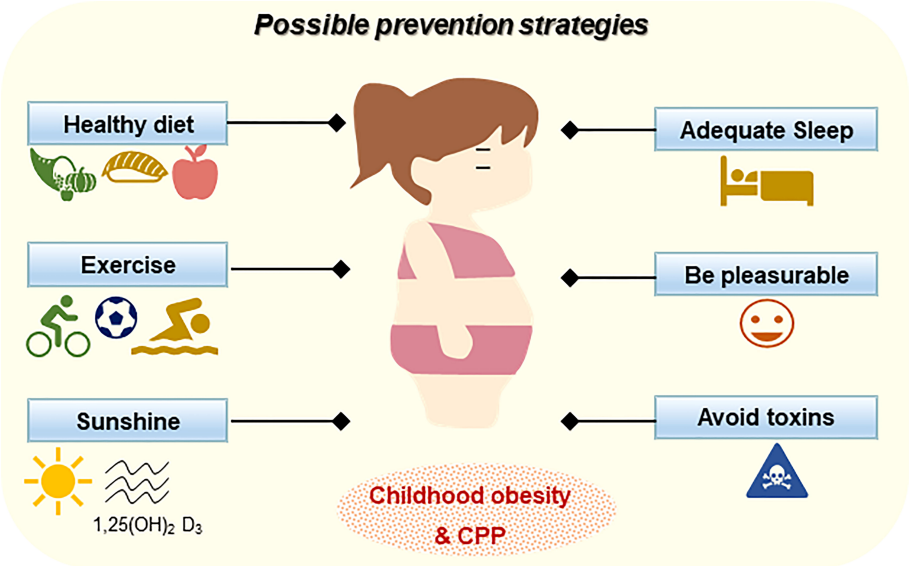

FIGURE 2
The possible prevention strategies for childhood obesity and CPP.

health, as well as a safe environment is not only beneficial for children not only at adolescence, but also for whole life health.

Author contributions

LZ conceptualized the manuscript, LS and ZJ collected the literature and drafted the manuscript, LZ revised the manuscript. All authors contributed to the article and approved the submitted version.

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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.

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References

1. Cantas-Orsdemir S, Garb JL, Allen HF. Prevalence of cranial MRI findings in girls with central precocious puberty: A systematic review and meta-analysis. *J Pediatr Endocrinol Metab* (2018) 31:701–10. doi: 10.1515/jpem-2018-0052

2. Clarkson J, Boon WC, Simpson ER, Herbison AE. Postnatal development of an estradiol-kisspeptin positive feedback mechanism implicated in puberty onset. *Endocrinology* (2009) 150:3214–20. doi: 10.1210/en.2008-1733

3. Kauffman AS, Navarro VM, Kim J, Clifton DK, Steiner RA. Sex differences in the regulation of Kiss1/NKB neurons in juvenile mice: Implications for the timing of puberty. *Am J Physiol Endocrinol Metab* (2009) 297:E1212–21. doi: 10.1152/ajpendo.00461.2009

4. Fava D, Calandrino A, Calevo MG, Allegri AEM, Napoli F, Gastaldi R, et al. Clinical, endocrine and neuroimaging findings in girls with central precocious puberty. *J Clin Endocrinol Metab* (2022) 107:e4132–43. doi: 10.1210/clinem/dgac422

5. He Z, Yuan B. Diagnostic value of combined detection of pelvic ultrasound and serum LH, FSH, and E2 levels in children with idiopathic central precocious puberty. *Evid Based Complement Alternat Med* (2022) 2022:7928344. doi: 10.1155/2022/7928344

6. Huang A, Reinehr T, Roth CL. Connections between obesity and puberty: Invited by Manuel tena-sempere, Cordoba. *Curr Opin Endocr Metab Res* (2020) 14:160–8. doi: 10.1016/j.coemr.2020.08.004

7. Dundar I, Akinci A. Prevalence of type 2 diabetes mellitus, metabolic syndrome, and related morbidities in overweight and obese children. *J Pediatr Endocrinol Metab* (2022) 35:435–41. doi: 10.1515/jpem-2021-0271

8. Perry JR, Day F, Elks CE, Sulem P, Thompson DJ, Ferreira T, et al. Parent-of-origin-specific allelic associations among 106 genomic loci for age at menarche. *Nature* (2014) 514:92–7. doi: 10.1038/nature13545

9. Pai LF, Wang DS, Hsu WF, Huang SW, Chung CH, Chen SJ, et al. New insights into precocious puberty and ADHD: A nationwide cohort study. *Pediatr Res* (2022). doi: 10.1038/s41390-022-02028-5

10. Cesario SK, Hughes LA. Precocious puberty: A comprehensive review of literature. *J Obstet Gynecol Neonatal Nurs* (2007) 36:263–74. doi: 10.1111/j.1552-6909.2007.00145.x

11. Day FR, Elks CE, Murray A, Ong KK, Perry JR. Puberty timing associated with diabetes, cardiovascular disease and also diverse health outcomes in men and women: The UK biobank study. *Sci Rep* (2015) 5:11208. doi: 10.1038/srep11208

12. Canoy D, Beral V, Balkwill A, Wright FL, Kroll ME, Reeves GK, et al. Age at menarche and risks of coronary heart and other vascular diseases in a large UK cohort. *Circulation* (2015) 131:237–44. doi: 10.1161/CIRCULATIONAHA.114.010070

13. Ruscica M, Macchi C, Gandini S, Morlotti B, Erzegovesi S, Bellodi L, et al. Free and bound plasma leptin in anorexia nervosa patients during a refeeding program. *Endocrine* (2016) 51:380–3. doi: 10.1007/s12020-015-0598-6

14. Zhao C, Tang Y, Cheng L. Diagnostic value of LH peak value of the GnRH stimulation test for girls with precocious puberty and its correlation with body mass index. *Comput Math Methods Med* (2022) 2022:4118911. doi: 10.1155/2022/4118911

15. Kaplowitz PB, Backeljauw PF, Allen DB. Toward more targeted and cost-effective gonadotropin-releasing hormone analog treatment in girls with central precocious puberty. *Horm Res Paediatr* (2018) 90:1–7. doi: 10.1159/000491103

16. Livadas S, Chrousos GP. Molecular and environmental mechanisms regulating puberty initiation: An integrated approach. *Front Endocrinol (Lausanne)* (2019) 10:828. doi: 10.3389/fendo.2019.00828

17. Biro FM, Galvez MP, Greenspan LC, Succop PA, Vangeepuram N, Pinney SM, et al. Pubertal assessment method and baseline characteristics in a mixed longitudinal study of girls. *Pediatrics* (2010) 126:e583–90. doi: 10.1542/peds.2009-3079

18. Anyanwu OU, Ibekwe RC, Nwokocha AR, Ibe CB. An assessment of sexual maturation among school girls in abakaliki metropolis, ebonyi state, south-East Nigeria. *Niger Postgrad Med J* (2016) 23:121–6. doi: 10.4103/1117-1936.190348

19. Lian Q, Mao Y, Luo S, Zhang S, Tu X, Zuo X, et al. Puberty timing associated with obesity and central obesity in Chinese han girls. *BMC Pediatr* (2019) 19:1. doi: 10.1186/s12887-018-1376-4

20. Khadgawat R, Marwaha RK, Mehan N, Surana V, Dabas A, Sreenivas V, et al. Age of onset of puberty in apparently healthy school girls from northern India. *Indian Pediatr* (2016) 53:383–7. doi: 10.1007/s13312-016-0857-5

21. Li Y, Ma T, Ma Y, Gao D, Chen L, Chen M, et al. Trajectories, and earlier puberty onset: Results from a longitudinal cohort study. *J Clin Endocrinol Metab* (2022) 107:2462–72. doi: 10.1210/clinem/dgac395

22. Bleil ME, Appelhans BM, Gregorich SE, Thomas AS, Hiatt RA, Roisman GI, et al. Patterns of early life weight gain and female onset of puberty. *J Endocr Soc* (2021) 5(12):bvab165. doi: 10.1210/jendso/bvab165

23. Bendor CD, Bardugo A, Rotem RS, Derazne E, Gerstein HC, Tzur D, et al. Glucose intolerance in pregnancy and offspring obesity in late adolescence. *Diabetes Care* (2022) 45:1540–8. doi: 10.2337/dc21-2634

24. Nam HK, Lee KH. Small for gestational age and obesity: epidemiology and general risks. *Ann Pediatr Endocrinol Metab* (2018) 23:9–13. doi: 10.6065/apem.2018.23.1.9

25. Verkauskiene R, Petraitiene I, Albertsson Wikland K. Puberty in children born small for gestational age. *Horm Res Paediatr* (2013) 80:69–77. doi: 10.1159/000353759

26. Manfredi-Lozano M, Roa J, Tena-Sempere M. Connecting metabolism and gonadal function: Novel central neuropeptide pathways involved in the metabolic control of puberty and fertility. *Front Neuroendocrinol* (2018) 48:37–49. doi: 10.1016/j.yfrne.2017.07.008

27. Zurita-Cruz JN, Villasis-Keever MA, Manuel-Apolinar L, Damasio-Santana L, Gutierrez-Gonzalez A, Wakida-Kusunoki G, et al. Altered cardiometabolic profile in girls with central precocious puberty and adipokines: A propensity score matching analysis. *Cytokine* (2021) 148:155660. doi: 10.1016/j.cyto.2021.155660

28. Obradovic M, Sudar-Milovanovic E, Soskic S, Essack M, Arya S, Stewart AJ, et al. Leptin and obesity: Role and clinical implication. *Front Endocrinol (Lausanne)* (2021) 12:585887. doi: 10.3389/fendo.2021.585887

29. Kang MJ, Oh YJ, Shim YS, Baek JW, Yang S, Hwang IT. The usefulness of circulating levels of leptin, kisspeptin, and neurokinin b in obese girls with precocious puberty. *Gynecol Endocrinol* (2018) 34:627–30. doi: 10.1080/09513590.2017.1423467

30. Tschop M, Weyer C, Tataranni PA, Devanarayan V, Ravussin E, Heiman ML. Circulating ghrelin levels are decreased in human obesity. *Diabetes* (2001) 50:707–9. doi: 10.2337/diabetes.50.4.707

31. Soriano-Guillen L, Barrios V, Chowen JA, Sanchez I, Vila S, Quero J, et al. Ghrelin levels from fetal life through early adulthood: Relationship with endocrine and metabolic and anthropometric measures. *J Pediatr* (2004) 144:30–5. doi: 10.1016/j.jpeds.2003.08.050

32. Yu Z, Zhan Q, Chen A, Han J, Zheng Y, Gong Y, et al. Intermittent fasting ameliorates di-(2-ethylhexyl) phthalate-induced precocious puberty in female rats: A study of the hypothalamic-pituitary-gonadal axis. *Reprod Biol* (2021) 21:100513. doi: 10.1016/j.repbio.2021.100513

33. A.S.N.R, From the American Association of Neurological Surgeons, C.I.R.A, C.N.S, Cardiovascular Interventional Radiology Society of Europe and E.S.M.I.N.T, et al. Multisociety consensus quality improvement revised consensus statement for endovascular therapy of acute ischemic stroke. *Int J Stroke* (2018) 13:612–32. doi: 10.1177/1747493018778713

34. Bruning JC, Gautam D, Burks DJ, Gillette J, Schubert M, Orban PC, et al. Role of brain insulin receptor in control of body weight and reproduction. *Science* (2000) 289:2122–5. doi: 10.1126/science.289.5487.2122

35. Burcelin R, Thorens B, Glauser M, Gaillard RC, Pralong FP. Gonadotropin-releasing hormone secretion from hypothalamic neurons: stimulation by insulin and potentiation by leptin. *Endocrinology* (2003) 144:4484–91. doi: 10.1210/en.2003-0457

36. Moret M, Stettler R, Rodieux F, Gaillard RC, Waeber G, Wirthner D, et al. Insulin modulation of luteinizing hormone secretion in normal female volunteers and lean polycystic ovary syndrome patients. *Neuroendocrinology* (2009) 89:131–9. doi: 10.1159/000160911

37. Texada MJ, Lassen M, Pedersen LH, Koyama T, Malita A, Rewitz K. Insulin signaling couples growth and early maturation to cholesterol intake in drosophila. *Curr Biol* (2022) 32:1548–1562.e6. doi: 10.1016/j.cub.2022.02.021

38. Dura-Trave T, Gallinas-Victoriano F. Hyper-androgenemia and obesity in early-pubertal girls. *J Endocrinol Invest* (2022) 45:1577–85. doi: 10.1007/s40618-022-01797-4

39. Ouyang L, Yang F. Combined diagnostic value of insulin-like growth factor-1, insulin-like growth factor binding protein-3, and baseline luteinizing hormone levels for central precocious puberty in girls. *J Pediatr Endocrinol Metab* (2022) 35:874–9. doi: 10.1515/jpem-2022-0161

40. Zhu Y, Fang Y, Medina D, Bartke A, Yuan R. Metformin treatment of juvenile mice alters aging-related developmental and metabolic phenotypes. *Mech Ageing Dev* (2022) 201:111597. doi: 10.1016/j.mad.2021.111597

41. Navarro VM. Metabolic regulation of kisspeptin - the link between energy balance and reproduction. *Nat Rev Endocrinol* (2020) 16:407–20. doi: 10.1038/s41574-020-0363-7

42. Gaytan F, Morales C, Leon S, Heras V, Barroso A, Avendano MS, et al. Development and validation of a method for precise dating of female puberty in laboratory rodents: The puberty ovarian maturation score (Pub-score). *Sci Rep* (2017) 7:46381. doi: 10.1038/srep46381

43. Roa J, Barroso A, Ruiz-Pino F, Vazquez MJ, Seoane-Collazo P, Martinez-Sanchez N, et al. Metabolic regulation of female puberty *via* hypothalamic AMPK-kisspeptin signaling. *Proc Natl Acad Sci U.S.A.* (2018) 115:E10758–67. doi: 10.1073/pnas.1802053115

44. Manzano M, Giron MD, Salto R, Vilchez JD, Reche-Perez FJ, Cabrera E, et al. Quality more than quantity: The use of carbohydrates in high-fat diets to tackle obesity in growing rats. *Front Nutr* (2022) 9:809865. doi: 10.3389/fnut.2022.809865

45. Roa J, Garcia-Galiano D, Varela L, Sanchez-Garrido MA, Pineda R, Castellano JM, et al. The mammalian target of rapamycin as novel central regulator of puberty onset *via* modulation of hypothalamic Kiss1 system. *Endocrinology* (2009) 150:5016–26. doi: 10.1210/en.2009-0096

46. Vazquez MJ, Toro CA, Castellano JM, Ruiz-Pino F, Roa J, Beiroa D, et al. SIRT1 mediates obesity- and nutrient-dependent perturbation of pubertal timing by epigenetically controlling Kiss1 expression. *Nat Commun* (2018) 9:4194. doi: 10.1038/s41467-018-06459-9

47. Martinez de Morentin PB, Gonzalez-Garcia I, Martins L, Lage R, Fernandez-Mallo D, Martinez-Sanchez N, et al. Estradiol regulates brown adipose tissue thermogenesis *via* hypothalamic AMPK. *Cell Metab* (2014) 20:41–53. doi: 10.1016/j.cmet.2014.03.031

48. Shao P, Wang Y, Zhang M, Wen X, Zhang J, Xu Z, et al. The interference of DEHP in precocious puberty of females mediated by the hypothalamic IGF-1/PI3K/Akt/mTOR signaling pathway. *Ecotoxicol Environ Saf* (2019) 181:362–9. doi: 10.1016/j.ecoenv.2019.06.017

49. Srivastava VK, Hiney JK, Dees WL. Manganese-stimulated kisspeptin is mediated by the IGF-1/Akt/Mammalian target of rapamycin pathway in the prepubertal female rat. *Endocrinology* (2016) 157:3233–41. doi: 10.1210/en.2016-1090

50. Cao Y, Hu G, Zhang Q, Ma L, Wang J, Li W, et al. Kisspeptin-10 maintains the activation of the mTOR signaling pathway by inhibiting SIRT6 to promote the synthesis of milk in bovine mammary epithelial cells. *J Agric Food Chem* (2021) 69:4093–100. doi: 10.1021/acs.jafc.0c07613

51. Han Q, Zhang W, Lu C, Wu J, An S, Zhang S. Repression of Kisspeptin1 weakens hydrogen peroxide-caused injury in HTR8 cells *via* adjusting PI3K/AKT/mTOR pathway. *J Biochem Mol Toxicol* (2020) 34:e22461. doi: 10.1002/jbt.22461

52. Gerendai I, Toth IE, Boldogkoi Z, Medveczky I, Halasz B. Neuronal labeling in the rat brain and spinal cord from the ovary using viral transneuronal tracing technique. *Neuroendocrinology* (1998) 68:244–56. doi: 10.1159/000054372

53. Heras V, Castellano JM, Fernandois D, Velasco I, Rodriguez-Vazquez E, Roa J, et al. Central ceramide signaling mediates obesity-induced precocious puberty. *Cell Metab* (2020) 32:951–966.e8. doi: 10.1016/j.cmet.2020.10.001

54. Brito VN, Spinola-Castro AM, Kochi C, Kopacek C, Silva PC, Guerra-Junior G. Central precocious puberty: Revisiting the diagnosis and therapeutic management. *Arch Endocrinol Metab* (2016) 60:163–72. doi: 10.1590/2359-3997000000144

55. Glab E, Wikiera B, Bieniasz J, Barg E. The influence of GnRH analog therapy on growth in central precocious puberty. *Adv Clin Exp Med* (2016) 25:27–32. doi: 10.17219/acem/31433

56. Arrigo T, De Luca F, Antoniazzi F, Galluzzi F, Segni M, Rosano M, et al. Reduction of baseline body mass index under gonadotropin-suppressive therapy in girls with idiopathic precocious puberty. *Eur J Endocrinol* (2004) 150:533–7. doi: 10.1530/eje.0.1500533

57. Schug TT, Janesick A, Blumberg B, Heindel JJ. Endocrine disrupting chemicals and disease susceptibility. *J Steroid Biochem Mol Biol* (2011) 127:204–15. doi: 10.1016/j.jsbmb.2011.08.007

58. Faienza MF, Brunetti G, Acquafredda A, Delvecchio M, Lonero A, Gaeta A, et al. Bone health, and risk of polycystic ovary syndrome in girls with idiopathic central precocious puberty treated with gonadotropin-releasing hormone analogues. *Horm Res Paediatr* (2017) 87:162–9. doi: 10.1159/000456546

59. Loochi SA, Demol S, Nagelberg N, Lebenthal Y, Phillip M, Yackobovitch-Gavan M. Gonadotropin releasing hormone analogue therapy in girls with idiopathic precocious puberty/early-fast puberty: dynamics in adiposity indices, eating habits and quality of life. *J Pediatr Endocrinol Metab* (2021) 34:373–83. doi: 10.1515/jpem-2020-0471

60. Wijarn P, Poomthavorn P, Khlairit P, Pongratanakul S, Chailurkit L, Mahachoklertwattana P. Short-term effects of gonadotropin-releasing hormone analogue treatment on leptin, ghrelin and peptide YY in girls with central precocious puberty. *J Pediatr Endocrinol Metab* (2021) 34:479–84. doi: 10.1515/jpem-2020-0470

61. Ramos CO, Latronico AC, Cukier P, Macedo DB, Bessa DS, Cunha-Silva M, et al. Long-term outcomes of patients with central precocious puberty due to hypothalamic hamartoma after GnRHa treatment: Anthropometric, metabolic, and reproductive aspects. *Neuroendocrinology* (2018) 106:203–10. doi: 10.1159/000477584

62. Mizuno G, Yamada H, Munetsuna E, Ando Y, Teshigawara A, Ito M, et al. High-fructose corn syrup intake has stronger effects on the transcription level of hepatic lipid metabolism-related genes, *via* DNA methylation modification, in childhood and adolescence than in other generations. *Life Sci* (2022) 301:120638. doi: 10.1016/j.lfs.2022.120638

63. Liu G, Guo J, Zhang X, Lu Y, Miao J, Xue H. Obesity is a risk factor for central precocious puberty: A case-control study. *BMC Pediatr* (2021) 21:509. doi: 10.1186/s12887-021-02936-1

64. Liu S, Zhu X, Wang Y, Yan S, Li D, Cui W. The association between vitamin d levels and precocious puberty: A meta-analysis. *J Pediatr Endocrinol Metab* (2020) 33:427–9. doi: 10.1515/jpem-2019-0388

65. Dura-Trave T, Gallinas-Victoriano F. Vitamin d status and parathyroid hormone assessment in girls with central precocious puberty. *J Endocrinol Invest* (2022) 45(11):2069–75. doi: 10.21203/rs.3.rs-499316/v1

66. Acinikli KY, Erbas IM, Besci O, Demir K, Abaci A, Bober E. Has the frequency of precocious puberty and rapidly progressive early puberty increased in girls during the COVID-19 pandemic? *J Clin Res Pediatr Endocrinol* (2022) 14:302–7. doi: 10.4274/jcrpe.galenos.2022.2022-12-11

67. Imamura S, Hur SP, Takeuchi Y, Badruzzaman M, Mahardini A, Rizky D, et al. Effect of short- and long-term melatonin treatments on the reproductive activity of the tropical damselfish chrysiptera cyanea. *Fish Physiol Biochem* (2022) 48:253–62. doi: 10.1007/s10695-022-01051-x

68. Zhou F, Jin Z, Zhu L, Huang F, Ye A, Hou C. A preliminary study on the relationship between environmental endocrine disruptors and precocious puberty in girls. *J Pediatr Endocrinol Metab* (2022) 35:989–97. doi: 10.1515/jpem-2021-0691

69. Pivonello C, Muscogiuri G, Nardone A, Garifalos F, Provvisiero DP, Verde N, et al. Bisphenol a: An emerging threat to female fertility. *Reprod Biol Endocrinol* (2020) 18:22. doi: 10.1186/s12958-019-0558-8

70. Bigambo FM, Sun H, Yan W, Wu D, Xia Y, Wang X, et al. Association between phenols exposure and earlier puberty in children: A systematic review and meta-analysis. *Environ Res* (2020) 190:110056. doi: 10.1016/j.envres.2020.110056

71. Hu Y, Li J, Yuan T, Yu T, Chen Y, Kong H, et al. Exposure to antibiotics and precocious puberty in children: A school-based cross-sectional study in China. *Environ Res* (2022) 212:113365. doi: 10.1016/j.envres.2022.113365

72. Sakali AK, Bargiota A, Fatouros IG, Jamurtas A, Macut D, Mastorakos G, et al. Effects on puberty of nutrition-mediated endocrine disruptors employed in agriculture. *Nutrients* (2021) 13(11):4184. doi: 10.3390/nu13114184

73. Hu B, Mo Z, Jiang J, Liang J, Wei M, Zhu X, et al. The pheromone affects reproductive physiology and behavior by regulating hormone in juvenile mice. *Growth Factors* (2022) 40:13–25. 10.1080/08977194.2022.2053527

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Clinical, genetic, and epidemiological survey of Polish children and adolescents with severe obesity: A study protocol of the Polish–German study project on severe early-onset obesity

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Severe early-onset obesity (SEOO) in children is a common feature of monogenic obesity. Nowadays, mutations in at least 50 genes are known to be related to monogenic obesity, and many others are tested. Part of them is involved in the leptin–proopiomelanocortin pathway. The aim of the project is to establish the Polish database of severely obese children and adolescents and to evaluate the prevalence of monogenic forms of obesity in this cohort, with a special focus on leptin–proopiomelanocortin pathway abnormalities. The secondary project aim is to identify new population-specific mutations in obesity-related genes in severely obese Polish children and adolescents. This is a prospective multi-center clinical study performed in four Polish centers. The estimated sample size is 500 patients aged 1–18 years, with severe obesity, hyperphagia, and food-seeking behaviors. In each patient, the medical history regarding the obesity duration in the patient and obesity and its complication existence in the family will be taken. Next, the questionnaire regarding the symptom characteristic of specific mutations, which we are going to test, will

be performed. Hyperphagia will be assessed on the basis of age-specific questionnaires. The physical examination with anthropometric measurement, basic biochemical and hormonal tests, and leptin and biologically active leptin measurements will be performed. Finally, genetic analysis will be performed using next-generation sequencing with sequencing libraries prepared to include obesity-related genes. The genotyping findings will be confirmed with the use of classic sequencing (Sanger's method). In the future, the pathogenicity of new mutations in obesity-related genes identified in our cohort is planned to be confirmed by functional testing *in vitro*. Nowadays, there are no data regarding the prevalence of severe obesity or monogenic obesity in Polish children. This project has the potential to improve understanding of obesity etiology and may contribute to implementing attribute mutation-specific treatment. Moreover, it may lead to a finding of new, population-specific mutations related to SEOO.

KEYWORDS

monogenic obesity, leptin, severe early onset obesity, children, adolescents

1 Introduction

The obesity epidemic has become an extremely important medical and socioeconomic issue in many countries, as excessive body weight is the leading cause of increased morbidity and mortality. The risk of obesity and its consequences in adulthood are even higher if the problem starts in childhood. In some countries, efforts have been undertaken to prevent and treat childhood obesity leading to the stabilization of the number of cases. However, the proportion of severely obese adults and children is still growing, and this group is at high risk of morbidity and mortality.

It is well known that the most common reason for excessive body weight is an imbalance between calorie input and output, and it is strongly related to lifestyle. Reduced daily physical activity with increased calorie consumption enhanced by polygenic background is the leading cause of obesity. However, there is a group of patients in whom a monogenic cause of obesity can be identified. This group is usually characterized by severe obesity with early onset, before the age of 6 years (severe early-onset obesity (SEOO)) (1). Kohlsdorf et al. revealed in a retrospective cohort study that patients with monogenic obesity due to leptin or leptin receptor deficiency showed an enormous increase in body mass index (BMI) during the first 2 years of life. Their BMI was >25 kg/m^2 at the age of 2 years and >30 kg/m^2 at the age of 5 years (2). These BMI values may be useful in distinguishing the monogenetic form of obesity from common obesity based on trajectories of BMI during early childhood. Nevertheless, in patients with some mutations, for example, in the melanocortin-4 receptor gene (*MC4R*), the BMI trajectories are similar to those of patients with simple obesity (3). Skinner et al. (4) showed that 7% of girls and 8.7% of boys in the USA suffer from severe obesity defined as a BMI above 120% of the 95th percentile. In a Spanish cohort, 2% of preschool children were severely obese (5). In subjects with SEOO, depending on the country, population studied, ethnicity, and the number of genes tested, the prevalence of monogenic obesity could vary between 3% and 10% of cases (6–8). Akinci et al. in a cohort of 105 SEOO children screened for 41 known-obesity-related genes found the genetic background in 10.4% of them (6). Slovenian national study has revealed that 1.4% of participants had known disease-causing heterozygous variants (DCVs) in the genes of the leptin–melanocortin signaling pathway and that 4.1% of participants were carriers of rare variants of unknown clinical significance (VUS) (9). Additionally, population-specific mutations in obesity-related genes exist. In a US cohort of African American and Latino children with SEOO, three out of eight discovered mutations in MC4R gene were new and not previously reported (7). A systematic review of the genetics of monogenic obesity in the 22 Arab countries in 13 studies has revealed carried 14 variants in five genes related to monogenic obesity; all of these variants were pathogenic, homozygous, and carried by members of consanguineous families (10). Unfortunately, there are no data about the prevalence of severe obesity in Polish children as well as about the prevalence of monogenic obesity in Polish SEOO children.

The aim of our project was to establish the Polish database of severely obese children and adolescents and to evaluate the prevalence of monogenic forms of obesity in this cohort, with a special focus on leptin–proopiomelanocortin pathway

abnormalities. The secondary project aim was to identify new population-specific mutations in obesity-related genes in severely obese Polish children and adolescents. This is the first research assessing the prevalence of monogenic obesity in these parts of Europe in the population of such homogeneity. In Poland, there are a few minorities of different origins who migrated from other parts of the world (Middle East, North Africa, etc.), and the majority of Polish citizens are Caucasians with a low incidence of consanguinity.

2 Study objectives

1. To establish a Polish database of children with severe obesity
2. To characterize clinically and biochemically patients with severe obesity
3. To assess leptin and bio-active leptin in children with severe obesity
4. To evaluate the prevalence of monogenic obesity among Polish children with severe obesity, especially in those with early-onset obesity
5. To identify new mutations in obesity-related genes specific to the population of Polish children with severe obesity

3 Study design

This is a prospective multi-center clinical study performed in four Polish centers. The sample size targeted is 500 patients aged 1–18 years, with severe obesity of an early origin, hyperphagia, and food-seeking behaviors.

4 Materials

4.1 Patients

4.1.1 Sample size

Five hundred children aged 1–18 years, with severe obesity of an early origin, will be recruited, using the following inclusion and exclusion criteria. The patients will be recruited from four Polish centers of pediatric endocrinology involved in childhood obesity management (Zabrze, Cracow, Rzeszów, and Szczecin) from inpatient and outpatient departments. The child population in Poland was estimated to be about 7,000,000 (11); for a 5% error threshold and a test power of 0.95, the appropriate sample size was determined at 384 participants. Funds earmarked for this national project are sufficient for conducting a study on about half a thousand children. During the last 2 years, a widespread promotion campaign for this project has been carried out, among both the medical community and patients in Poland. Qualification criteria for the project were presented at medical conferences organized by Polish medical societies. Broad-based information campaign was held in the main Polish mass media through media presentations (interviews and radio and television studios). Through this action, children from the entire area of Poland will be enrolled.

4.1.2 Inclusion criteria

1. Age 1–18 years
2. The presence of severe obesity will be defined as a BMI > 25 kg/m^2 in a child below the age of 2 years, a BMI > 30 kg/m^2 in children aged 2–6 years, a BMI > 35 kg/m^2 in children aged 6–14 years, and BMI > 40 kg/m^2 in children aged >14 years (2)
3. Hyperphagia and food-seeking behaviors
4. Written informed consent of the patient's parent/guardian and patient above the age of 13 years to participate in the study

4.1.3 Exclusion criteria

1. Lack of written informed written consent from patients' parent/guardian or patient above the age of 13 years
2. Secondary cause of obesity: previously diagnosed genetic syndrome coexisting with obesity, treatment with medicine with known effect on weight gain (glucocorticoids, valproic acid, risperidone, and others), Cushing's syndrome, and other secondary causes of obesity (note: patients with other endocrine diseases, especially deficiencies, will not be excluded, as they could be the presentation of monogenic obesity)

5 Data collection

A single visit of the patient in the study center will be needed. In each patient, the study procedures mentioned below will be performed:

1. Taking the medical history of the patient and the patient's family (Appendix 1: Case Report Form (CRF)) regarding the obesity duration in the patient and obesity and its complication existence in the family
2. Questionnaire regarding symptoms characteristic of the specific mutation (Appendix 2 of CRF)

3. Hyperphagia assessment using the Polish version of questionnaires:

a. Children's Eating Behavior Questionnaire (CEBQ) by Wardle for children below the age of 8 years (Appendix 3a of CRF) (12)—answered by parent/guardian

b. Three-Factor Eating Questionnaire (TFEQ): for children above 8 years of age (Appendix 3b of CRF) (13)—answered by the patient.

In order to avoid bias, the questionnaires will be performed prior to the contact with an investigator.

Each patient or guardian will be asked to fulfill a standard 9-day diary of physical activity and a diary of the diet.

4. Physical examination with anthropometric measurements (Appendix 4 of CRF). Body weight will be measured to the nearest 0.1 kg on a calibrated balance beam scale, and body height was measured to the nearest 0.1 cm. Waist circumference will be measured at the level of the midpoint between the lowest rib and iliac crest, and head circumference will be measured with the band to the nearest 0.1 cm. In all children, the pubertal stage (development of breast, genitalia, and pubic hair) will be documented according to the classification by Tanner. Blood pressure will be measured with calibrated automatic blood pressure monitor with the cuff size appropriate to the arm size in children sitting for at least 15 min before the examination.

5. Biochemical and hormonal tests (Appendix 4 of CRF) which are usually performed on every patient with obesity during routine management of the subject (at the first visit at the outpatient/inpatient department and then usually repeated once a year). They are performed in each center by using commercial, widely available methods. Oral glucose tolerance test (OGTT) results will be available from obese children above the age of 10 years or younger in whom puberty has begun (14).

6. Leptin and biologically active leptin assessment (15). Additional fasting blood taking (2–3 ml of blood) will required to obtain 1 ml of serum. Measurements of total leptin and biologically active leptin will be performed in the central laboratory, using ELISA.

7. Genetic analysis—next-generation sequencing. Sequencing libraries prepared to include 11 target genes (*LEP*, *LEPR*, *MC4R*, *SIM1*, *KSR1*, *POMC*, *PCSK1*, *NTRK2*, *MRAP2*, *SH2B1*, and *BDNF*), in addition to further analysis of other, less frequent, obesity-related genes (*UCP1*, *UCP3*, *CARTPT*, *DYRK1B*, *NROB2*, *PCSK2*, *PPARG*, *PPP1R3A*, *PPARGC1A*, *CCK*, *SLC2A4*, *TUB*, *ADCY3*, *SREBF1*, *ADRB2*, *ADRB3*, *AGRP*, *MC3R*, *ENPP1*, *PPARGC1B*, *PYY*, *SDC3*, *ADIPOQ*, *NAMPT*, *CFD*, *RETN*, *NPY*, *ADD1*, *PTPN1*, *IRS-1*, *GHRL*, *NEGR1*, *GIPR*, *TMEM18*, *FTO*, and *SLC22A1*) will be performed. The full names of the 11 genes, their location, type of inheritance, and phenotype characteristics of mutation are presented in Table 1. The possible function and clinical phenotype of the obesity-related genes that we examined are presented in Table 2.

Genetic testing needed an additional 1 ml of blood taking (EDTA). The biological samples will be immediately sent to the Department of Medical Genetics for centralized analyses. For assessment of the presence of small gene variants, 100 ng of genomic DNA of every sample will be enzymatically fragmented (as determined at Sure Select XT and XT Low Input Enzymatic Fragmentation Protocol, Agilent Technologies, Santa Clara, CA, USA) and used for exome library preparation according to the SureSelectXT HS Target Enrichment System for Illumina Paired-End Multiplexed Sequencing Library protocol (Agilent Technologies). Next, the libraries will be tagged, pooled, and prepared for sequencing. Finally, the sequencing of the libraries will be performed by an external genotyping service provider (Macrogen Europe, Netherlands) with the use of the NovaSeq 6000 genomic sequencer. The genotyping findings will be confirmed with the use of classic sequencing (Sanger's method) in the Department of Medical Genetics.

The raw genotyping data will be processed with the use of the DRAGEN Enrichment software (Illumina, San Diego, CA, USA) in order to obtain a table of all genetic variants in every patient. However, the final data analysis was restricted only to the *a priori*-selected set of genes of interest.

All data from the medical history, physical examination, and biochemical tests will be used to characterize the study group clinically and biochemically. The assessment of the prevalence of metabolic disturbances (lipid and carbohydrate disorders), metabolic-associated fatty liver disease, arterial hypertension, and metabolic syndrome will be performed, and their correlation with BMI z-score, bio-impedance data, and leptin and bioactive leptin level will be performed. To assess the leptin SDS, the Blum et al. formula will be used (16). The eating disorders will be analyzed on the basis of CEBQ and TFEQ. In children with an established mutation in genes related to obesity, a detailed clinical and biochemical data analysis will be performed, and further, necessary biochemical and hormonal tests will be performed.

In the future, the pathogenicity of new mutations in obesity-related genes identified in our cohort is planned to be confirmed by functional testing *in vitro*. Moreover, the DNA samples from the parents of the patient with a confirmed mutation will be analyzed.

6 Data management

The patients' data will be anonymized and stored in each center (paper CRF). All data from paper CRF will be electronically documented in a password-secured database. An

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TABLE 1 Most common genes associated with severe early-onset obesity.

Gene name	Inheritance	Type of mutation	Chromosome Location	Additional symptoms
Melanocortin 4 receptor (MC4R)	AR	Homozygous or compound heterozygous	18q21.32	Somatomegaly, increased head circumference, hyperinsulinemia
	AD	Heterozygous		
Leptin (LEP)	AR	Homozygous or compound heterozygous	7q32.1	Hypogonadotropic hypogonadism, hypothyroidism, growth hormone deficiency, immune deficiency
Leptin receptor (LEPR)	AR	Homozygous or compound heterozygous	1p31.3	Hypogonadotropic hypogonadism, hypothyroidism, growth hormone deficiency, immune deficiency
Single-minded homolog 1 (SIM1)	AD	Heterozygous, chromosomal rearrangement	6q16.3	Autism, behavioral problems
Kinase suppressor of Ras 2 (KSR2)	AD	Heterozygous	12q24.22-q24.23	Insulin resistance, low heart rate, reduced basal metabolic rate
Proopiomelanocortin (POMC)	AR	Homozygous or compound heterozygous	2p23.3	ACTH/adrenal insufficiency, abnormal pigmentation
	AD	Heterozygous		
Protein convertase subtilisin/kexin 1 (PCSK1)	AR	Homozygous or compound heterozygous	5q15	Neonatal diarrhea, hypoglycemia, hypothyroidism, adrenal insufficiency, diabetes insipidus
	AD	Heterozygous		
Neurotrophic receptor tyrosine kinase 2 (NTRK2)	AD	Heterozygous	9q21.33	Developmental delay, hyperactivity
Melanocortin 2 receptor accessory protein 2 (MRAP2)	AD	Heterozygous	6q14.2	none
SH2B adaptor protein 1 (SH2B1)	AD	Heterozygous	16p11.2	Severe insulin resistance, behavioral difficulties, developmental delay
Brain-derived neurotrophic factor (BDNF)	AD	Heterozygous	11p14.1	Developmental delay, hyperactivity

Note. AR, autosomal recessive; AD, autosomal dominant; ACTH, adrenocorticotropic hormone.

electronic platform will be developed for data storage and statistical analysis. The statistical analysis of the results will be conducted with software dedicated to analyzing medical data.

7 Discussion

Nowadays, mutations in at least 50 genes are known to be related to monogenic obesity, and many others are tested (1, 8, 17–19). Many of them are involved in the leptin–proopiomelanocortin pathway. In our project, we are going to examine Polish SEOO children for the presence of mutations in 11 genes, most frequently related to monogenic severe obesity with early onset (Table 1) (17, 20, 21). In addition, we performed further analysis of other, less frequent, obesity-related genes (6, 18).

In all patients with those mutations, both hyperphagia, characterized by extreme food-seeking behavior, and early onset of obesity are typical symptoms. In most patients, there are also other diverse symptoms that are characteristic of each mutation (Table 1). Mutations in the *MC4R* are the most common form of inherited SEOO with a prevalence of 0.5%–5.8% in different populations (22). Among the obese Czech child population, similar to the Polish one, the prevalence of *MC4R* homozygous and heterozygous mutations is 2.4% (23). Other mutations are less frequent. It is suggested that leptin deficiency or leptin receptor defects, inherited in an autosomal recessive pattern, could be found even in up to 3% of patients with SEOO (24). So far, 100 patients with SEOO caused by leptin gene mutation were identified. The prevalence of leptin receptor gene (*LEPR*) mutations can reach even 2%–3% in certain populations (24). Mutations in proopiomelanocortin (*POMC*) and proprotein convertase subtilisin/kexin type 1 (*PCSK1*) genes were identified in 10 patients for each gene (8).

The establishment Polish database of children with severe obesity and assessment of bio-active leptin in children with severe obesity were partly met between 2015 and 2019 when the Polish–German consortium was established to implement the "Early-onset Obesity and Leptin—German-Polish Study (EOL-GPS)" project. Fifty SEOO children were recruited from three Polish medical centers (Katowice, Rzeszów, and Szczecin) and

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TABLE 2 The possible function and clinical phenotype of the obesity-related genes.

Possible function	Gene	Clinical phenotype
Leptin–melanocortin pathway	*LEP, LEPR, MC4R, POMC, PCSK1, SH2B1, PCSK2, CARTPT, MRAP2, SIM1, BDNF, NPY, AGRP, MC3R, IRS-1, SDC3*	Given in Table 1
Thermogenesis	*UCP1, UCP3, UCP2, PPARG PPARGC1A,*	Obesity, type 2 diabetes
Glucose oxidation	*KSR2*	Hyperphagia in childhood, low heart rate, reduced basal metabolic rate severe insulin resistance
Fatty acid oxidation	*KSR2, PPARGC1B*	Severe insulin resistance
Glycogen synthesis	*PPP1R3A*	Severe insulin resistance, lipodystrophy, type 2 diabetes, carotid intima media thickness
Adipogenic differentiation	*DYRK1B, PPARG*	Abdominal obesity, metabolic syndrome
Mesolimbic pathway—regulates appetite, gastric emptying	*CCK, PYY, GHRL*	
Incretin regulation (insulin secretion, gastric motility, nutrient absorption, food intake)	*GIPR*	
Glucose transport	*SLC2A4*	Severe insulin resistance, type 2 diabetes
Regulation of catecholamine function (lipolysis, energy expenditure)	*ADRB2, ADRB3*	Type 2 diabetes, lower resting metabolic rate, abdominal obesity, insulin resistance
Regulation Insulin Receptor	*NAMPT, IRS-1*	
Regulation Complement System (regulation of insulin production, glucose uptake, and triglyceride synthesis)	*CFD*	
Adipokine genes	*ADIPOQ, LEP, LEPR, RETN*	
Renin-angiotensin-aldosterone system (RAAS)	*ADD1*	
Regulation of insulin and leptin signaling	*PTPN1*	
Unclear/pleiotropic regulation of obesity	*NEGR1, TMEM18, FTO, SLC22A1, NR0B2, ADCY3*	

one German center (Ulm). Clinical profile, leptin, and biologically active leptin were determined. In this cohort with SEOO, we identified no new cases of children with leptin deficiency or bio-inactive leptin. However, relative leptin deficiency in children with SEOO could be suspected (25). In order to further explore this important topic, the Polish–German consortium decided to continue its cooperation. The current study is the first to focus on monogenic obesity in Polish SEOO children, to establish the prevalence of the most common monogenic lesions as a cause of severe obesity, and to identify new mutations in obesity-related genes specific to the Polish population. In the future, the pathogenicity of new mutations in obesity-related genes identified in our cohort is planned to be confirmed by functional testing *in vitro*.

Based on the rarity of monogenic gene variants as the cause of obesity, the question has to be posed whether there is a justification to search for them. Aside from purely scientific reasons—finding a new, population-specific mutation related to SEOO—the justification is given by the fact that during the past years, mutation-specific treatments have been developed. Patients with leptin deficiency as well as patients with biologically inactive leptin can be treated by administration of recombinant human leptin (metreleptin) (24–27). MC4R agonist, setmelanotide, is now approved for the treatment in patients with POMC, LEPR, and proprotein convertase subtilisin/kexin type 1 (PCSK1) deficiencies (28–31). It is also known that patients with some mutations can be successfully treated with well-known drugs; e.g., glucagon-like peptide 1 (GLP-1) agonist is effective in weight reduction in patients with *MC4R* mutations, and obesity related to Kinase Suppressor of Ras-2 (*KSR2*) mutation is well treated with metformin (21, 28, 30). Identification of monogenic background is also important in patients' qualification for bariatric surgery. Cooiman et al. (17) showed that patients with *MC4R* mutations achieved superior weight loss after primary Roux-en-Y gastric bypass compared with sleeve gastrectomy.

There are no data regarding the prevalence of severe obesity or monogenic obesity in Polish children. However, looking at the data available for the Czech Republic where 2.2% of children are severely obese, we can estimate that at least the same percentage of Polish children also suffer from severe obesity (32). This figure would be equivalent to an estimated 150,000 Polish children. Most probably because of the genetic diversity of our population and the lack of ethnic minorities with high consanguinity in our country, the prevalence of monogenic obesity is expected to be rather low. If we assume that it is at the level of 3%, it means that about 4,500 children in Poland can be obese due to a mutation in a single gene. In some of them, more than lifestyle intervention and diet can be offered, and specific treatment can be used.

8 Study limitation

The study is a cross-sectional analysis. Our sample size is limited; therefore, all cases of monogenic obesity in the Polish population will not be detected. The active recruitment is taking place only in four centers in Poland. Despite the vast informational campaign, there is the risk of the underrepresentation of patients from the regions located at larger distances from the recruiting centers. Moreover, we realized that we are going to analyze a limited group of the selected genes, not covering all possible reasons for monogenic obesity. For this reason, we decided to bank the same genetic material from every patient for future analysis, if it will be possible to perform.

Ethics statement

The study will be conducted according to the Declaration of Helsinki on "Ethical Principles for Medical Research in Humans" (9 July 2018). The study was approved by the local ethics committees (No. PCN/CBN/0022/KB1/137/I/21/22, KBETUJ 1072.6120.69.2022, KB-006/12/2022). At each participating institution, informed written consent will be obtained from every patient's parents/guardians and every patient above the age of 13 years.

Author contributions

AZ, AM, EP, MR, MWa, EM-T, MWo, BF, and SB: conceived the project. AZ, MB-M, and MM: wrote the original draft. MWo, EP, and AM: responsible for conducting the literature review. EM-T and MR: participated in revising the work for important intellectual content. All authors contributed to the article and approved the submitted version.

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Conflict of interest

Author BF is employed by Mediagnost GmbH.

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

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References

1. AbouHashem N, Zaied RE, Al-Shafai K, Nofal M, Syed N, Al-Shafai M. The spectrum of genetic variants associated with the development of monogenic obesity in Qatar. *Obes Facts.* (2022) 15(3):357–65. doi: 10.1159/000521851

2. Kohlsdorf K, Nunziata A, Funcke JB, Brandt S, von Schnurbein J, Vollbach H, et al. "Frühkindlicher BMI-verlauf bei monogener adipositas". *Medizinische Genetik* (2017) 294:360–4. doi: 10.1007/s11825-017-0167-x"

3. Kohlsdorf K, Nunziata A, Funcke JB, Brandt S, von Schnurbein J, Vollbach H, et al. Early childhood BMI trajectories in monogenicobesity due toleptin, leptinreceptor, and melanocortin 4 receptordeficiency. *Int J Obes (Lond).* (2018) 42(9):1602–9. doi: 10.1038/s41366-018-0049-6

4. Skinner AC, Ravanbakht SN, Skelton JA, Perrin EM, Armstrong SC. Prevalence of obesity and severe obesity in US children, 1999-2016. *Pediatrics* (2018) 141:e20173459. doi: 10.1542/peds.2017-3459

5. Cadenas-Sanchez C, Intemann T, Labayen I, Artero EG, Alvarez-Bueno C, Sanchis-Moysi J, et al. Prevalence of severe/morbid obesity and other weight status and anthropometric reference standards in Spanish preschool children: The PREFIT project. *Pediatr Res* (2020) 87:501–10. doi: 10.1038/s41390-019-0325-8

6. Akinci A, Türkkahraman D, Tekedereli İ, Özer L, Evren B, Şahin İ, et al. Novel mutations in obesity-related genes in Turkish children with non-syndromic early onset severe obesity: A multicentre study. *J Clin Res Pediatr Endocrinol* (2019) 11:341–9. doi: 10.4274/jcrpe.galenos.2019.2019.0021

7. De Rosa MC, Chesi A, McCormack S, Zhou J, Weaver B, McDonald M, et al. Characterization of rare variants in MC4R in African American and Latino children with severe early-onset obesity. *J Clin Endocrinol Metab* (2019) 104:2961–70. doi: 10.1210/jc.2018-02657

8. Thaker VV. Genetic and epigenetic causes of obesity. *Adolesc Med State Art Rev* (2017) 28:379–405. doi: 10.1542/9781581109405-genetic

9. Šket R, Kotnik P, Bizjan BJ, Kocen V, Mlinarič M, Tesovnik T, et al. Heterozygous genetic variants in autosomal recessive genes of the leptin-melanocortin signalling pathway are associated with the development of childhood obesity. *Front Endocrinol (Lausanne).* (2022) 13:832911. doi: 10.3389/fendo.2022.832911

10. AbouHashem N, Al-Shafai K, Al-Shafai M. The genetic elucidation of monogenic obesity in the Arab world: a systematic review. *J Pediatr Endocrinol Metab* (2022) 35(6):699–707. doi: 10.1515/jpem-2021-0710

11. *Statistic polands* (2022). Available at: https://bdl.stat.gov.pl/bdl/dane/podgrup/tablica.

12. Wardle J, Guthrie CA, Sanderson S, Rapoport L. Development of the children's eating behaviour questionnaire. *J Child Psychol Psychiatry* (2001) 42:963–70. doi: 10.1111/1469-7610.00792

13. Dzielska A, Mazur J, Małkowska-Szkutnik A, Kołoło H. Adaptation of the tree-factor eating questionnaire (TFEQ-13) for school-aged adolescents in a population study. *ProblHigEpidemiol* (2009) 90:362–9.

14. Available at: https://cukrzyca.info.pl/aktualnosc/zalecenia-kliniczne-dot-postepowania-u-chorych-z-cukrzyca-2021-stanowisko-ptd.

15. Wabitsch M, Prizdum L, Ranke M, von Schnurbein J, Moss A, Brandt S, et al. Measurement of immunofunctional leptin to detect and monitor patients with functional leptin deficiency. *Eur J Endocrinol* (2017) 176:315–22. doi: 10.1530/EJE-16-0821

16. Blum WF, Englaro P, Hanitsch S, Juul A, Hertel NT, Müller J, et al. Plasma leptin levels in healthy children and adolescents: dependence on body mass index, body fat mass, gender, pubertal stage, and testosterone. *J ClinEndocrinolMetab.* (1997) 82(9):2904–10. doi: 10.1210/jcem.82.9.4251

17. Cooiman MI, Kleinendorst L, Aarts EO, Janssen IMC, van Amstel HKP, Blakemore AI, et al. Genetic obesity and bariatric surgery outcome in 1014 patients with morbid obesity. *Obes Surg* (2020) 30:470–7. doi: 10.1007/s11695-019-04184-w

18. Paolacci S, Pompucci G, Paolini B, Del Ciondolo I, Miggiano GAD, Aquilanti B, et al. Mendelian non-syndromic obesity. *Acta BioMed* (2019) 90:87–9. doi: 10.23750/abm.v90i10-S.8766

19. Durmaz A, Aykut A, Atik T, Özen S, AyyıldızEmecen D, Ata A, et al. A new cause of obesity syndrome associated with a mutation in the carboxypeptidase gene detected in three siblings with obesity, intellectual disability and hypogonadotropic hypogonadism. *J Clin Res Pediatr Endocrinol* (2021) 13(1):52–60. doi: 10.4274/jcrpe.galenos.2020.2020.0101

20. Von Schnurbein J, Wabitsch M. Monogene adipositas. pathophysiologie – diagnostik – therapieoptionen. *Medizinische Genetik* (2017) 4:348–59. doi: 10.1007/s11825-017-0157-z

21. Pearce LR, Atanassova N, Banton MC, Bottomley B, van der Klaauw AA, Revelli JP, et al. KSR2 mutations are associated with obesity, insulin resistance, and impaired cellular fuel oxidation. *Cell* (2013) 155:765–77. doi: 10.1016/j.cell.2013.09.058

22. El-Sayed Moustafa JS, Froguel P. From obesity genetics to the future of personalized obesity therapy. *Nat Rev Endocrinol* (2013) 9:402–13. doi: 10.1038/nrendo.2013.57

23. Hainerová I, Larsen LH, Holst B, Finková M, Hainer V, Lebl J, et al. Melanocortin 4 receptor mutations in obese Czech children: studies of prevalence, phenotype development, weight reduction response, and functional analysis. *J Clin Endocrinol Metab* (2007) 92:3689–96. doi: 10.1210/jc.2007-0352

24. Farooqi IS, O'Rahilly S. 20 years of leptin: human disorders of leptin action. *J Endocrinol* (2014) 223:T63–70. doi: 10.1530/JOE-14-0480

25. Zachurzok A, Ranke MB, Flehmig B, Jakubek-Kipa K, Marcinkiewicz K, Mazur A, et al. Relative leptin deficiency in children with severe early-onset obesity (SEOO) - results of the early-onset obesity and leptin - German-polish study (EOL-GPS). *JPediatrEndocrinolMetab* (2020) 33:255–63. doi: 10.1515/jpem-2019-0469

26. Wabitsch M, Funcke JB, Lennerz B, Kuhnle-Krahl U, Lahr G, Debatin KM, et al. Biologically inactive leptin and early-onset extreme obesity. *N Eng J Med* (2015) 372:48–54. doi: 10.1056/NEJMoa1406653

27. Wabitsch M, Funcke JB, von Schnurbein J, Denzer F, Lahr G, Mazen I, et al. Severe early-onset obesity due to bioinactive leptin caused by a p.N103K mutation in the leptin gene. *J ClinEndocrinolMetab* (2015) 100:3227–30. doi: 10.1210/jc.2015-2263

28. Iepsen EW, Zhang J, Thomsen HS, Hansen EL, Hollensted M, Madsbad S, et al. Patients with obesity caused by melanocortin-4 receptor mutations can be treated with a glucagon-like peptide-1 receptor agonist. *CellMetab* (2018) 28:23–32.e3. doi: 10.1016/j.cmet.2018.05.008

29. Markham A. Setmelanotide: First approval. *Drugs.* (2021) 81(3):397–403. doi: 10.1007/s40265-021-01470-9

30. Available at: https://www.rhythmtx.com/our-pipeline/.

31. Poitou C, Mosbah H, Clément K. MECHANISMS IN ENDOCRINOLOGY: Update on treatments for patients with genetic obesity. *Eur J Endocrinol* (2020) 183 (5):R149–66. doi: 10.1530/EJE-20-0363

32. Spinelli A, Buoncristiano M, Kovacs VA, Yngve A, Spiroski I, Obreja G, et al. Prevalence of severe obesity among primary school children in 21 European countries. *Obes Facts* (2019) 12:244–58. doi: 10.1159/000500436

Appendix 1

Medical history of patient and patient's family.

Patient code						**Date**
Date of birth						
Birth weight [g]						
Birth length [cm]						
Head circumference at birth [cm]						
Length of pregnancy[weeks]						
Apgar score						
The age when obesity occurred						
Mother's age						
Mother's height [cm]						
Mother's weight [kg]						
Father's age						
Father's height [cm]						
Father's weight [kg]						
Sibling's age						
Sibling's height [cm]						
Sibling's weight [kg]						
Body weight available from med. data [kg] **(date/value)**						
Length/height available from med. data [cm] **(date/value)**						
Family history	Obesity	Arterial hypertension	Diabetes mellitus (type 2/ during pregnancy)	Lipids disturbances (hypercholesterolemia/ hypertriglyceridemia)	Cardiovascular diseases (coronary disease, stroke, MI)	Others (endocrine disorders, neurological, suggesting monogenic obesity)
Who						

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Appendix 2 Questionnaire regarding the symptoms characteristic for specific mutation.

Patientcode…… **Date**……

	Yes	No	Not known	Comment
Facial dysmorphia				
Somatomegaly				
Frequent, severe infections (immunodeficiency)				
Endocrine disorders				
Growth hormone deficiency				
Hypothyroidism				
Adrenal insufficiency				
Hypogonadism				
Diabetes insipidus				
Neurological disorders				
Intellectual disability/ developmental delay				
Behavioral problems – ADHD				
Autism				
Memory problems				
Impaired pain sensation				
Skin (pale) and hair (red) abnormal pigmentation				
Acanthosis nigricans/ hyperinsulinemia				
Hypoglycemic episodes				
Neonatal diarrhoea				

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Appendix 3A

Patient code	Date				
	Never	Rarely	Some-times	Often	Always
My child loves food	☐	☐	☐	☐	☐
My child eats more when worried	☐	☐	☐	☐	☐
My child has a big appetite	☐	☐	☐	☐	☐
My child finishes his/her meal quickly	☐	☐	☐	☐	☐
My child is interested in food	☐	☐	☐	☐	☐
My child is always asking for a drink	☐	☐	☐	☐	☐
My child refuses new foods at first	☐	☐	☐	☐	☐
My child eats slowly	☐	☐	☐	☐	☐
My child eats less when angry	☐	☐	☐	☐	☐
My child enjoys tasting new foods	☐	☐	☐	☐	☐
My child eats less when s/he is tired	☐	☐	☐	☐	☐
My child is always asking for food	☐	☐	☐	☐	☐
My child eats more when annoyed	☐	☐	☐	☐	☐
If allowed to, my child would eat too much	☐	☐	☐	☐	☐
My child eats more when anxious	☐	☐	☐	☐	☐
My child enjoys a wide variety of foods	☐	☐	☐	☐	☐
My child leaves food on his/her plate at the end of a meal	☐	☐	☐	☐	☐
My child takes more than 30 minutes to finish a meal	☐	☐	☐	☐	☐

	Never	Rarely	Some-times	Often	Always
Given the choice, my child would eat most of the time	☐	☐	☐	☐	☐
My child looks forward to mealtimes	☐	☐	☐	☐	☐
My child gets full before his/her meal is finished	☐	☐	☐	☐	☐
My child enjoys eating	☐	☐	☐	☐	☐
My child eats more when she is happy	☐	☐	☐	☐	☐
My child is difficult to please with meals	☐	☐	☐	☐	☐
My child eats less when upset	☐	☐	☐	☐	☐
My child gets full up easily	☐	☐	☐	☐	☐
My child eats more when s/he has nothing else to do	☐	☐	☐	☐	☐
Even if my child is full up s/he finds room to eat his/her favourite food	☐	☐	☐	☐	☐
If given the chance, my child would drink continuously throughout the day	☐	☐	☐	☐	☐
My child cannot eat a meal if s/he has had a snack just before	☐	☐	☐	☐	☐
If given the chance, my child would always be having a drink	☐	☐	☐	☐	☐
My child is interested in tasting food s/he hasn't tasted before	☐	☐	☐	☐	☐
My child decides that s/he doesn't like a food, even without tasting it	☐	☐	☐	☐	☐
If given the chance, my child would always have food in his/her mouth	☐	☐	☐	☐	☐
My child eats more and more slowly during the course of a meal	☐	☐	☐	☐	☐

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Appendix 3b

Three-Factor Eating Questionnaire (TFEQ).

Patient code	**Date**			
	Definitely yes	**Rather yes**	**Rather not**	**Definitely not**
I take small portions on purpose, in order to control my body mass				
I eat when I feel nervous				
The company of someone eating makes me so hungry I have to eat too				
I overeat when I feel sad				
When I see something tasty I get so hungry I immediately have to eat				
I often feel so hungry that I could eat endlessly				
I am always hungry, therefore I cannot stop eating until I empty my plate				
Food comforts me when I feel lonely				
I consciously control the amount of my meals not to gain weight				
I abstain from some food because they make me gain weight				
I am always so hungry I can eat anytime				
Can you consciously eat less than you would like to?				
How much do you restrain eating? Check the 1-8 scale (1 - I never restrain eating/ 8 - I always restrain eating)		2. 3. 4. 5. 6. 7. 8		

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Appendix 4

Data from physical examination, biochemical and hormonal tests results.

Patient code	**Date of investigation**
Physical examination	
Height [cm]	
Weight [kg]	
Waist circumference [cm]	
Head circumference [cm]	
Blood pressure [mmHg]	
Pubertal stage [Tanner scale]	
Body composition (by bioimpedance) – not obligatory	yes/no (if yes please attach the result)

Fasting blood results	**value**	**unit**
Glucose		
Insulin		
Total cholesterol		
HDL cholesterol		
LDL cholesterol		
Triglycerides		
ALT		
TSH		
FT4		
Others (esp. US of the abdomen – liver echogenicity, OGTT, Cortisol)		

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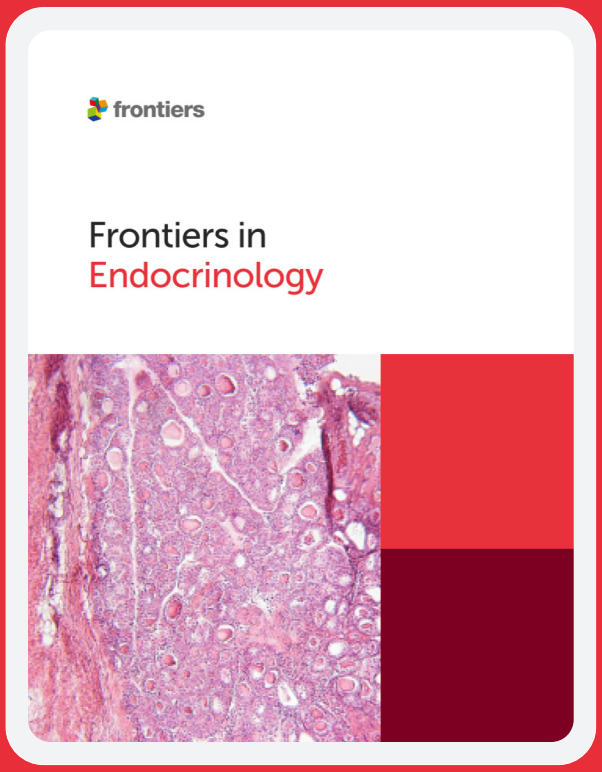

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