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Insulin sensitivity indices: fasting versus glucose-stimulated indices in pediatric non-alcoholic fatty liver disease

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Abstract. – OBJECTIVE: We aimed to compare insulin sensitivity indices, fasting vs glucose stimulated, in children and adolescents with non-alcoholic fatty liver disease.

PATIENTS AND METHODS: Two hundred-eleven obese children with median age of 11.24 ± 2.65 years were evaluated. After initial clinical and anthropometric examination, B-mode ultrasonography (USG) was performed and all subjects underwent Oral Glucose Tolerance Test (OGTT). Quantitative insulin sensitivity check index (QUICKI), homeostatic model assessment for insulin resistance (Homa-IR), the insulinogenic index (IGI), the Matsuda index, and the oral glucose insulin sensitivity (OGIS) model were used to determine peripheral insulin sensitivity.

RESULTS: 59.24% (68 boys, 57 girls) of obese children had NALFD. The prevalence of FLD in obese adolescents was significantly higher than in prepubertal children (65.8% vs. 51.5%). Fasting glucose, insulin, Homa-IR, QUICKI, and OGIS and Matsuda were significantly different between subjects with and without NALFD. Insulin and glucose indices were not found to be significantly different in the prepubertal group, whereas Homa-IR, QUICKI, Matsuda, and OGIS were significantly different in the pubertal group. Age, waist circumference, and OUICKI were found to be risk factors associated with the presence of NALFD in the logistic-regression analysis.

CONCLUSIONS: Age, waist circumference, and OUICKI were found to be risk factors associated with NALFD. As the value of QUICKI decreases, the probability of having steatosis increases. Although OGTT results gave the information about the glucose tolerance of a subject, indices derived from OGTT were not found to be superior to the traditional surrogates such as Homa-IR or QUICKI.

Key Words:
Fatty liver, Obesity, Children, Puberty, Insulin sensitivity, Indices.

Introduction

Non-alcoholic fatty liver disease (NAFLD) is defined as the presence of steatosis in more than 5% of hepatocytes in the absence of significant alcohol consumption, drug use, or hereditary diseases. NALFD is currently the most common cause of liver disease in youth and its prevalence increases concomitant with the epidemic of xchildhood obesity[1]. The clinical spectrum of NAFLD ranges from simple steatosis to non-alcoholic steatohepatitis, fibrosis, and cirrhosis[2,3]. Obesity is clearly associated with NAFLD, but only a subset of obese children develop the condition. The pathogenesis of NAFLD in overweight and obese individuals is not fully understood. Insulin sensitivity is of prime importance to identify individuals at risk of developing diabetes mellitus (DM), and impaired insulin sensitivity has the most important role in the pathogenesis of NALFD[4-6]. NAFLD is accepted as a hepatic component of the metabolic syndrome[6]. The prevalence of diabetes was reported to be 7.7% among the adult NAFLD patients[7]. The coexistence of NAFLD and DM is clinically important, and may cause the progression of hepatic fibrosis and increases the risk of cardiovascular disorders[7]. We aimed to find the prevalence of impaired glucose tolerance in children and ado-

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lescents with NALFD and to assess Oral Glucose Tolerance Test (OGTT)-derived insulin sensitivity indices, and to compare these indices at fasting state vs OGTT- based between obese children and adolescents with and without NALFD.

Patients and Methods

Two-hundred-eleven obese (99 male, 112 female) children with median age of 11.24 ± 2.65 years, who were evaluated in outpatients clinics, were included in this study. All children have no significant health condition except obesity, and were not affected by any chronic disease. They didn't take any medication and none of them had a history of consumption of alcohol.

The study was approved by the Scientific Ethics Committee of Celal Bayar University. Informed written consent was obtained from the parents of all children who underwent radiological and biochemical investigation.

Biochemical Assessments

Children who attended the clinic underwent initial clinical and anthropometric examination, followed by B-mode ultrasonography (US) liver evaluation. After a 12-hour overnight fast, blood samples were drawn to assess triglyceride (TG), aspartate aminotransferase (AST), alanine aminotransferase (ALT), high density cholesterol (HDL-C), low density cholesterol (LDL-C), and total cholesterol (Total-C) levels, and all subjects underwent 1.75 g/kg OGTT examinations to establish their glucose tolerance. The plasma glucose and insulin levels were analyzed before and 30, 60, 90, and 120 minutes after oral glucose loading. Thyroid function tests, diurnal cortisol levels and basal ACTH levels were assessed to exclude hypothyroidism and hypercortisolism. Individuals with suspected FLD using B-Mode had a complete evaluation of iron status (serum iron, transferrin, and ferritin concentrations) and serology to exclude hemochromatosis and viral hepatitis (hepatitis B surface antigen, hepatitis B-C total antibody, antibody against hepatitis A virus immunoglobulin G, and hepatitis C virus antibody immunoglobulin G). Sera was separated from the blood samples that were centrifuged at 1000 g for 10 minutes at +4°C. Serum glucose, TG, AST, ALT, HDL-C, and Total-C levels were assessed by original commercial reagents (Beckman Coulter Ireland Inc., Mervue, Galway, Ireland) on analyzer (UniCelD × C 800 Synchron Clinical System, Fullerton, CA, USA). Serum insulin concentrations were analyzed using the chemiluminescent immunometric assay method on analyzer (Siemens IMMULITE 2000, Siemens Medical Solutions Diagnostics Limited, Llanberis, UK) with original reagents. Intra-assay coefficient of variation (CV) is 5.5% at 7.67 µIU/ml, 4% at 12.5 µIU/ml, 3.3% at 17.2 µIU/ml, and 3.9% at 26.4 µIU/ml concentrations. Inter-assay CV is 7.3% at 7.67 µIU/ml, 4.9% at 12.5 µIU/ml, 4.1% at 17.2 µIU/ml, and 5% at 26.4 µIU/ml concentrations. Fasting insulin and glucose levels were used to calculate the quantitative insulin sensitivity check index (QUICKI): [1/log insulin + log glycemia in mg/dL] and homeostatic model assessment for insulin resistance (Homa-IR) = [fasting insulin (mIU/mL) × fasting glucose (mmol/L)/22.5][8,9]. The insulinogenic index (IGI) was calculated by using fasting and OGTT's insulin and glucose levels at 30 minutes as follows: IGI: [(insulin 30 – insulin 0)/(glucose 30 – glucose 0)]. Matsuda index (Matsuda) = [10,000/√GO × 10 Gmean × Imean] and The Oral Glucose Insulin Sensitivity (OGIS) = f (G_0, G_{90}, G_{120}, I_0, I_{90}, I_{120}, D_O) (http:// www.isib.cnr.it/bioing/ogis/home.html) model was used to determine peripheral insulin sensitivity based on dynamic insulin and glucose responses during the OGTT[10,11].

To estimate insulin resistance, an insulin peak of ≥ 150 µU/ml and/or ≥ 75 µU/ml 120 min after glucose loading was used, and the sum of insulin levels up to 120 min during OGTT was also > 300 µU/mL[12,13]. Impaired glucose tolerance (IGT) was defined as a two-hour post-load plasma glucose level of 140-199 mg/dL, and impaired fasting glucose (IFG) was defined as a plasma glucose level of 100 mg/dL to < 126 mg/dL. Diabetes was defined as a two-hour post-load plasma glucose level of 200 mg/dL[14].

Anthropometric Measurements

Weight was measured to the nearest 0.5 kg using a balance beam scale, and height was measured to the nearest 0.1 cm with a manual height board. The body mass index (BMI; kg/m^2) was used as an index of relative weight. To compare BMI among different ages and in both boys and girls, the BMI standard deviation score (SDS) was calculated and BMI percentile was evaluated for age and gender. Obesity[15] was defined as percentile of BMI for age and sex ≥ 95th. Waist circumference (WC) was measured by a trained

person, to the nearest 0.1 cm at the midpoint between the bottom of the rib cage and the top of the iliac crest with the subjects standing, their weight equally distributed on both feet, their arms at their sides, and head facing straight forward.

Ultrasonographic Evaluation

B-mode sonographic examinations were performed by the same radiologist, by using Siemens Sonoline G 50 (Siemens, Milan, Italy) with 3.5 MHz convex transducers. Using B-mode sonography, the presence or absence and the severity of fatty infiltration was graded using a scale from 0 to 3, indicating absent, mild, moderate and severe hepatosteatosis, respectively, corresponding to increasing degrees of hepatic echogenicity with poorer visualisation of the intrahepatic vessels and diaphragm.

Statistical Analysis

Statistical Package for Social Sciences for Windows statistical software version 18.0 (SPSS Inc., Chicago, IL, USA) was used for all calculations. Data distribution was analyzed using the Kolmogorov-Smirnov test. Data showing normal distribution were analyzed with independent *t*-test, and Mann-Whitney U-test was used to evaluate for the data showing abnormal distribution, and the data were reported as mean (± SD). Pearson's correlation coefficients were used to quantify univariate associations among variables and multiple regression analysis was carried out to test the joint effects of different variables, on hepatosteatosis according to B-mode US. Due to the nature of QUICKI scores in the study, we used the percentage of OUICKI in the multiple regression analysis. In all analyses, a p-value ≤ 0.05 was considered significant.

Results

The obese population included 112 girls and 99 boys. 97 of them were in the prebubertal stage; the remaining 114 were in puberty. Anthropometric and biochemical characteristics of obese populations were shown in Table I.

We determined that 59.2% (125) of obese children and adolescents had FLD based on B-mode US. Among the children and adolescents with liver steatosis, 68 (68.7%) were male, 57 (50.9%) were female, whereas the numbers of boys were lower than girls in the study populations ($p < 0.01$). The prevalence of FLD in obese adolescents was significantly higher than prepubertal children (65.8% *vs.* 51.5%) ($p < 0.05$).

Obese children were divided into two groups: obese children with fatty liver and without fatty liver. Age, height, height z score, weight, weight z score, BMI, BMI z score, TG, ALT, AST levels, and waist circumference of obese children with

Table I. Anthropometric and metabolic characteristics of obese children.

Anthropometric and biochemical characteristics	Obese patients (n = 211)	Obese patients without fatty liver (n = 86)	Obese patients with fatty liver (n = 125)	*p*-value
Age (years)	11.24 ± 2.65	10.79 ± 2.65	11.49 ± 2.63	< 0.05
Gender (male/female)	99/112	31/55	68/57	< 0.01
Prepubertal/pubertal % (n:)	97/114	47/39	50/75	< 0.05
Height (cm)	148.96 ± 13.77	145.63 ± 13.22	150.81 ± 14.12	< 0.01
Weight (kg)	63.09 ± 19.20	55.98 ± 15.65	67.81 ± 20.72	< 0.01
Weight z score	3.52 ± 1.62	3.05 ± 1.50	3.86 ± 1.66	< 0.01
BMI (kg/m^2)	27.88 ± 4.75	25.87 ± 3.48	29.31 ± 5.22	< 0.01
BMI z score	2.79 ± 0.64	2.57 ± 0.59	2.95 ± 0.66	< 0.01
Waist (cm)	90.59 ± 12.81	85.30 ± 9.97	94.23 ± 13.32	< 0.01
Triglyceride (mg/dl)	113.83 ± 69.80	99.11 ± 65.18	122.14 ± 67.18	< 0.05
Cholesterol (mg/dl)	165.54 ± 30.66	164.26 ± 30.17	166.08 ± 31.17	0.386
HDL-chol (mg/dl)	39.641 ± 0.21	41.28 ± 10.12	38.13 ± 10.03	0.051
LDL-chol (mg/dl)	103.27 ± 26.27	104.82 ± 26.25	102.24 ± 28.53	0.780
AST (U/L)	31.19 ± 14.44	26.70 ± 5.85	33.04 ± 11.74	< 0.001
ALT (U/L)	31.42 ± 23.92	23.56 ± 29.31	35.33 ± 20.99	< 0.001

BMI: body mass index; ALT: alanine aminotransferase; AST: aspartate aminotransferase; HDL-C: high-density lipoprotein cholesterol; LDL-chol: Low-density lipoprotein cholesterol; BP: blood pressure; HOMA-IR: homeostasis model assessment of insulin resistance. All data are expressed as mean ± SD. $p < 0.05$ is statistically significant.

Table II. Insulin indices in obese children and adolescents with and without fatty liver.

Anthropometric and biochemical characteristics	Obese patients without fatty liver (n = 86)	Obese patients with fatty liver (n = 125)	*p*-value
FBG (mg/dl)	81.02 ± 0.89	82.88 ± 0.87	< 0.05
Insulin levels (uU/ml)	13.71 ± 1.11	19.08±1.04	< 0.001
Homa-IR	2.75 ± 2.03	3.91 ± 2.46	< 0.001
QUICKI	0.34 ± 0.00	0.32 ± 0.00	< 0.001
I at 30 min OGTT	77.37 ± 55.70	87.06 ± 55.76	0.780
BG at 30 min OGTT	133.83 ± 22.91	135.31 ± 26.49	0.95
IGI	1.40 ± 1.36	1.37 ± 1.14	0.542
Matsuda	5.01 ± 0.32	3.90 ± 0.24	< 0.01
OGIS	469.11 ± 67.34	445.80 ± 80.10	< 0.05
I at 120 min OGTT	74.69 ± 6.93	86.32 ± 6.85	0.379
BG at 120 min OGTT	118.27 ± 2.53	117.44 ± 2.16	0.953
AUC insulin	132.05 ± 77.57	156.75 ± 103.92	0.130

FBG: fasting blood sugar; Homa-IR: homeostasis model assessment of insulin resistance; QUICKI: the quantitative insulin sensitivity check index; OGTT: Oral Glucose Tolerance Test; BG: blood sugar; I: Insulin; IGI: the insulinogenic index; OGIS: Oral Glucose Insulin Sensitivity; AUC: Area under the curve. Data are expressed as mean ± SD. $p < 0.05$ is significant.

fatty liver were significantly higher in children and adolescents with liver steatosis ($p < 0.05$, Table I). Cholesterol, LDL, and HDL levels were not different among children with and without fatty liver ($p > 0.05$). Insulin and glucose indices derived from fasting state and OGTT-based were presented in Table II. The differences between obese children and adolescents with and without NALFD for fasting state glucose, insulin, and insulogenic indices (Homa-IR and QUICKI) were significant ($p < 0.001$). OGTT-based indices such as Insulinogenic index (IGI), insulin and blood glucose level at 120 minutes were not significantly different ($p > 0.05$). On the other hand, OGIS and Matsuda index were significantly lower in the obese group with NALFD ($p < 0.05$, $p < 0.01$ respectively).

When the study group was divided into prepubertal and pubertal children, insulin and glucose indices at fasting state, and OGTT-based were no longer significantly different in the prepubertal group (Table III) ($p > 0.05$). Whereas in the pubertal group, Homa-IR, QUICKI, Matsuda, and OGIS were significantly different between obese

Table III. Insulin indices in obese prepubertal children with and without fatty liver.

Anthropometric and biochemical characteristics	Obese patients without fatty liver (n = 47)	Obese patients with fatty liver (n = 50)	*p*-value
FBG (mg/dl)	82.04 ± 6.99	84.30 ± 9.15	0.071
Insulin levels (uU/ml)	11.23 ± 6.18	13.68 ± 9.02	0.189
Homa-IR	2.25 ± 1.25	2.89 ± 2.18	0.141
QUICKI	0.34 ± 0.03	0.33 ± 0.02	0.141
I at 30 min OGTT	69.62 ± 48.96	80.68 ± 68.46	0.908
BG at 30 min OGTT	130.02 ± 27.17	140.76 ± 24.88	0.142
IGI	1.37 ± 1.28	1.18 ± 0.91	0.795
Matsuda	5.49 ± 2.86	5.21 ± 2.99	0.626
OGIS	479.63 ± 67.08	473.95 ± 65.42	0.769
I at 120 min OGTT	60.00 ± 40.28	67.51 ± 65.55	0.795
BG at 120 min OGTT	111.78 ± 15.42	112.28 ± 17.09	0.828
AUC insulin b	115.99 ± 62.33	124.74 ± 89.72	0.660

FBG: fasting blood sugar; Homa-IR: homeostasis model assessment of insulin resistance; QUICKI: the quantitative insulin sensitivity check index; OGTT: Oral Glucose Tolerance Test; BG: blood sugar; I: Insulin; IGI: the insulinogenic index; OGIS: Oral Glucose Insulin Sensitivity; AUC: Area under the curve. Data are expressed as mean ± SD. $p < 0.05$ is significant.

Table IV. Insulin indices in obese pubertal adolescants with and without fatty liver.

Anthropometric and biochemical characteristics	Obese patients without fatty liver (n = 39)	Obese patients with fatty liver (n = 75)	*p*-value
FBG (mg/dl)	79.53 ± 8.49	82.61 ± 9.29	0.574
Insulin levels (uU/ml)	16.28 ± 12.36	22.41 ± 10.95	< 0.001
G0/I0	7.61 ± 6.83	4.98 ±3.18	< 0.001
HOMA-IR	3.28 ± 2.74	4.55 ± 2.37	< 0.001
QUICKI	0.33 ± 0.03	0.31 ± 0.02	< 0.001
I at 30 min OGTT	84.75 ± 63.19	95.60 ± 75.18	0.969
BG at 30 min OGTT	134.94 ± 25.05	131.28 ± 27.15	0.332
IGI	1.44 ± 1.47	1.48 ± 1.29	0.498
Matsuda	4.50 ± 2.68	3.09 ± 1.77	< 0.01
OGIS	454.38 ± 65.89	427.22 ± 83.75	< 0.05
I at 120 min OGTT	93.17 ± 76.73	97.64 ± 74.48	0.635
BG at 120 min OGTT	125.40 ± 27.33	120.75 ± 25.55	0.631
AUC insulin	148.94 ± 91.69	182.46 ± 109.17	0.176

FBG: fasting blood sugar; Homa-IR: homeostasis model assessment of insulin resistance; QUICKI: the quantitative insulin sensitivity check index, OGTT: Oral Glucose Tolerance Test, BG: blood sugar; I: Insulin; IGI: the insulinogenic index; OGIS: Oral Glucose Insulin Sensitivity; AUC: Area under the curve. Data are expressed as mean ± SD. $p < 0.05$ is significant.

adolescants with NALFD and without NALFD. But the other indices were not significantly different (Table IV).

According to the OGTT results, out of 211 obese subjects, 8 (3.7%) were diagnosed as having impaired fasting glucose and 29 (13.74%) of them showed abnormal glucose tolerance test, and none of them were diabetic. The differences in the prevalance of IGTT in obese children and adolescents with and without NALFD was not significant (14.4 *vs.* 12.7%, *p*:0.931) but the prevalence was higher in obese adolescents than prepubertal obese children (21.9 *vs.* 5.1%). Among the114 pubertal children, 69 (60.5%) exhibited insulin resistance; on the other hand, in 97 prepubertal children, only 28 (28.9%) exhibited insulin resistance ($p < 0.001$). But no significant differences in insulin resistance were observed between children and adolescents with and without NALFD in each group (*p*: 0.485).

Correlations of Anthropometric Measurements and the Indices (Table V)

In bivariate analysis, there were significantly strong correlations between Matsuda and fasting indices (Homa-IR, QUICKI), but OGIS was weakly correlated with fasting indices (Homa-IR and QUICKI). Age and waist circumference were low positively correlated with Homa-IR and low negatively with Matsuda and QUICKI. In adolescents,

Table V. Pearson correlation coefficients regarding the associations between anthropometric/biochemical parameters and insulin indices in obese children and adolescents with NAFLD.

	HOMA-IR(r)	QUICKI(r)	Matsuda(r)
Ages	0.421**	-0.416**	-0.398**
BMI z score	0.225**	-0.227**	-0.245**
Waist	0.490**	-0.486**	-0.488**
TG	0.189*	-0.187**	-0.206**
HDL-C	-0.312*	0.312**	0.247**
HOMA-IR(r)	1	-0.560	-0.865**
QUICKI(r)	-0.998**	1	0.868**
Matsuda(r)	-0.865**	0.868**	1
OGIS	-0.474**	0.473	0.666**

BMI: body mass index; TG: triglyceride; HDL-C: high-density lipoprotein cholesterol; Homa-IR: homeostasis model assessment of insulin resistance; QUICKI: the quantitative insulin sensitivity check index; OGIS: Oral Glucose Insulin Sensitivity. **Correlation is significant at the 0.01; *Correlation is significant at the 0.05.

Homa-IR and QUICKI correlated significantly with age ($r = 0.243$, $p < 0.009$ and $r = -0.293$, $p < 0.010$, respectively), but they weren't significantly correlated in prepubertal children. TG was correlated with Homa-IR and QUICKI, HDL was correlated with Homa-IR and QUICKI, but these correlations were negligible.

Risk Factors Associated with NALFD

Risk factors associated with the presence of NALFD were analyzed in the logistic-regression analysis. Anthropometric measurements (age, BMI, BMI z score, weight, weight z score, waist circumference) and insulogenic indices (Homa-IR, OUICKI, Matsuda, OGIS, IGI) were included in different models. Age and waist circumference among the anthropometric measurements, and OUICKI among the insulogenic indices predicted NALFD in obese children and adolescents (Table VI).

Discussion

Even though the diagnostic sensitivity of abdominal USG reaches up to 80% in moderate-severe hepatosteatosis, we thought that fatty infiltration of the liver parenchyma was underestimated in the present study[16]. The study assessed insulin sensitivity and insulin resistance indices based on fasting and OGTT in a group of obese children and adolescents with and without NALFD. Overall, Homa-IR as a surrogate index of insulin resistance was significantly higher, and insulin sensitivity indices such as Matsuda, OGIS, and QUICKI were significantly lower in the obese group with NALFD. When the study population was grouped into pubertal and prepubertal group, the differences in insulin sensitivity and resistance indices lost their significance in the prepubertal group, but they remained significantly different in the pubertal group. Logistic regression analysis demonstrated that the QUICKI, age and waist circumference, were significant independent factors for NALFD, and the decrease in 1% of QUICKI increases the risk of having NALFD about 17% in the obese children and adolescents.

The true prevalence of NAFLD is unknown due to differences the disease's definitions and modalities used for diagnosis[17]. Most studies of the prevalence of fatty liver in children have been restricted to the use of indirect measures to predict a histological outcome[18]. Schwimmer et al[5] found the prevalence of biopsy proven fatty liver for children and adolescents age 2 to 19 years as 9.6% and the highest rate of fatty liver was seen in obese children. Its prevalence may reach 12-80% in overweight and obese children[20]. In the present study prevalence of hepatosteatosis was 59.24% and its prevalence was higher in pubertal children. The higher occurrence in pubertal obese children may be explained by the longer duration of obesity and the influence of sex hormones and growth hormones[21]. There is conflicting evidence on gender as a risk factor for NAFLD. The pediatric fatty liver disease has shown a male predominance on the basis of pathology, and biopsy-proven NAFLD has a ratio of 2.2:1 in boys to girls[22,23]. In our study population, the occurrence of NALFD was significantly higher in obese boys.

QUICKI, age, and waist circumference were found to be the independent risk factors of liver steatosis in obese children and adolescents in the present study. It has been revealed that the pattern of obesity plays a role in NAFLD development and progression. Visceral adiposity, which may be related to a state of insulin resistance, represents a more influential component than BMI in predicting fatyy liver[24]. Waist circumference is a surrogate measure of central obesity and a known predictor of early and late metabolic complications of childhood obesity[25].

Table VI. Associations between liver steatosis and other main parameters in multiple logistic regression analysis.

Variables	β parameter estimation	p	OR	95% C.I. for EXP(B)	
				Lower	Upper
Age	-0.168	$p < 0.05$	0.845	0.722	0.990
WC	0.084	$p < 0.001$	1.087	1.047	1.130
OUICKI (%)	-0.180	$p < 0.001$	0.835	0.758	0.920

WC: Waist Circumference; QUICKI: the quantitative insulin sensitivity check index; $p < 0.05$ is significant.

Lin et al[26] showed that in obese children and adolescents, for every 5 cm increase in waist circumference, there was an odds ratio of 1.391 for predicting ultrasonographic liver steatosis and they found no significant differences between subjects with and without liver steatosis for BMI. Papendrou et al[27] found 35 out of 82 children (42.6%) had fatty liver based on ultrasonography, and divided them into three groups on the basis of the degree of steatosis. They found that BMI and WC indicate a statistically linear correlation with the degree of steatosis, and logistic regression analysis of factors associated with NAFLD revealed that Homa-IR and n-3 fatty acids consumption were the most significant factors. Kelishadi et al[28] found WC and ApoB/ApoA-I ratio had the highest odds ratio in increasing the risk of insulin resistance and NAFLD. In a pediatric study[29], every 1 cm increase in WC was associated with a 1.97-fold increased risk of NAFLD in boys and a 2.08-fold increased risk in girls. Sartoria et al[30] found BMI-Z score, ALT, uric acid, glucose and insulin during OGTT were independent predictors of NAFLD in Italian obese children.

The key factor in the pathogenesis of NAFLD is insulin resistance, and the associations between insulin resistance and NALFD have been found by previous studies[30-35]. Some investigators found the HOMA-IR was associated with liver steatosis in the pubertal group unlike in the prepubertal children, but in the other studies[36,37], HOMA-IR was associated with liver steatosis in prepubertal children.

In the present work, IR was found in 53.6% of obese patients with NALFD and in 44.18% of obese patients without NALFD. The mean of insulin and the indices were not significantly different in prepubertal children, but the differences of these indices were significant in the pubertal group. Pubertal insulin resistance was supported by previous cross-sectional researches and there is an approximately 25-30% reduction in insulin sensitivity as a result of transient increase in growth hormone levels during puberty[38,39]. Although our findings were consistent with the literature, the small size of the study group or sensitivity of the method to detect IR and hepatosteatosis may affect the interpretations of the results.

Homa-IR, OUICKI, Matsuda, OGIS, and IGI were included in the logistic regresion analysis model to predict NALFD in the present paper and only QUICKI was found to be the independent risk factor of liver steatosis in obese children and adolescents. The reduced insulin sensitivity that leads to lipolysis in adipose tissue gives rise to an increased flux of free fatty acids to the liver[6,40]. This effect combined with the increased hepatic lipogenesis as a result of high insulin level is responsible for the accumulation of triglycerides within the hepatocytes and the development of steatosis[41]. Homa-IR and QUICKI were highly correlated, but Homa-IR is not subject to change linearly along wide ranges of insulin sensitivity, so QUICKI might have been found to be the independent risk factor of liver steatosis, and a more accurate index of insulin sensitivity[42-44].

Obesity and NAFLD are closely associated with impaired glucose metabolism. Ectopic fat deposition both in thel iver and pancreas may play a role in the pathogenesis of DM, but the pathophysiological mechanisms and clinical importance of fatty pancreas are not as clear as those of the liver[45-48].

In the present study, prevalence of impaired glucose tolerance was higher in obese adolescents than prepubertal obese children (21.9 *vs.* 5.1%), but the prevalence was not different in children and adolescents with and without NALFD . The prevalence of IGT was consistent with the literature in prepubertal obese children[48].

Even though surrogate indices of insulin sensitivity derived from OGTT incorporate both peripheral and hepatic insulin sensitivity, they didn't add any extra information regarding the risk of obesity complications except states of glucose tolerance, in which we didn't find significant differences between our study populations with and without NALFD, and OGTT-derived indices weren't found to be superior to the fasting indices (e.g., Homa-IR and QUICKI), so it would be better to select fasting surrogates that are simpler to obtain, as opposed to the dynamic surrogates if diabetes or state of glucose tolerance of patients are not investigated closely.

The present work is associated with several limitations. First, obese children and adolescents didn't undergo liver biopsy, which is the gold standard, to diagnose NALFD. Second, control subjects who were lean and healthy were not evaluated for insulin resistance, sensitivity indices, and hepatosteatosis. Third, we couldn't perform the hyperinsulinemic euglycemic clamp-measured insulin sensitivity to confirm the correlation of these indices for obese children and adolescents with and without NALFD in our study population.

Conclusions

We observed a high prevalence of NALFD in obese children and adolescents. Age, waist circumference, and OUICKI were found to be risk factors associated with NALFD. As the value of QUICKI decreases, the probability of having steatosis increases. Although OGTT results gave the information about the glucose tolerance of a subject, indices derived from OGTT were not found to be superior to the traditional surrogates such as Homa-IR or QUICKI.

Conflict of Interest

The Authors declare that there are no conflicts of interest.

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