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ORIGINAL ARTICLE

Subclinical Hypothyroidism as a Risk Factor for the Development of Cardiovascular Disease in Obese Adolescents With Nonalcoholic Fatty Liver Disease

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Abstract No data are available on the relationship between subclinical hypothyroidism and risk factors for the development of cardiovascular disease in obese adolescents with nonalcoholic fatty liver disease (NAFLD). This study aimed to determine whether an association exists between subclinical hypothyroidism and risk factors for the development of cardiovascular disease in obese adolescents with NAFLD. The study enrolled 111 obese adolescents and 42 lean subjects. The obese subjects were divided into two subgroups based on the presence or absence of fatty liver with high transaminases: a NAFLD group and a non-NAFLD group. Subclinical hypothyroidism was defined as a thyroid-stimulating hormone (TSH) level higher than 4 mIU/l and a normal free-thyroxine level (0.6–1.8 ng/dl). Insulin resistance was calculated by the homeostasis model assessment (HOMA-IR). Left ventricular mass (LVM), LVM index measurements, carotid intima media thickness (IMT), and HOMA-IR values were higher in the NAFLD obese group with TSH levels higher than 4 mIU/l than in the NAFLD obese group with TSH levels lower than 4 mIU/l. Elevated TSH values in the NAFLD obese group were positively correlated with most of the metabolic and cardiovascular risk parameters such as total cholesterol ($r = 0.606$, $p = 0.001$), triglycerides ($r = 0.476$, $p = 0.016$), low-density lipoprotein cholesterol ($r = 0.461$, $p = 0.004$), insulin ($r = 0.607$, $p = 0.001$), HOMA-IR ($r = 0.596$, $p = 0.002$), carotid IMT ($r = 0.894$, $p < 0.0001$), and LVM ($r = 0.563$, $p = 0.003$). The findings demonstrated that the obese adolescents with NAFLD and subclinical hypothyroidism had a more adverse cardiovascular risk profile and a higher carotid IMT and LVM.

Keywords Nonalcoholic fatty liver disease · Subclinical hypothyroidism

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Introduction

Carotid intima media thickness (IMT), left ventricular mass (LVM), and adolescent subclinical hypothyroidism are risk factors for the development of cardiovascular disease in obese adolescents with nonalcoholic fatty liver disease (NAFLD). Obesity in childhood and adolescence has been associated with adverse changes in cardiac geometry, including increased LVM [15]. Abnormal LVM and cardiac geometry are well-established risk factors for cardiovascular events [4].

Due to the current epidemic of pediatric obesity, NAFLD has become the most frequently diagnosed liver disease among children and adolescents [18]. Findings show NAFLD to be strongly associated with obesity, insulin resistance, hypertension, and dyslipidemia, and NAFLD currently is regarded as the liver manifestation of metabolic syndrome [24]. Increasing evidence supports an association between NAFLD and an increased risk of cardiovascular morbidity and mortality [34]. The measurement of carotid artery IMT is increasingly used to evaluate the cardiovascular risk and target

organ damage in children and adolescents with metabolic abnormalities [17].

Recently, increased interest has focused on the association between thyroid dysfunction and obesity. In 7–23 % of obese children, moderately elevated thyroid-stimulating hormone (TSH) levels are manifest in association with normal fT4 or fT3 levels, but the mechanisms underlying such thyroid hormonal changes in obese children are unclear [25]. Patients with primary hypothyroidism have a threefold greater risk for early atherosclerosis, as shown independently for other risk factors such as atherogenic lipid profile, hypertension, and impaired endothelial function. Whether subclinical hypothyroidism has an influence on the same risk factors and is associated with atherosclerosis still is debated [38]. Some studies show an association [6, 10, 31], but others do not [28, 37].

However, no data are available on the relationship between subclinical hypothyroidism and risk factors for the development of cardiovascular disease in obese adolescents with NAFLD. Therefore, we aimed to determine whether an association exists between subclinical hypothyroidism and risk factors for the development of cardiovascular disease in obese adolescents with NAFLD.

Materials and Methods

Study Population

From April 2011 to April 2012, 111 obese adolescents (57 girls and 54 boys) with mean age of 13.19 ± 1.3 years (range 12–17 years) and a mean body mass index (BMI) of 29.89 ± 3.30 kg/m^2 were recruited from obese adolescents admitted to the Pediatric Endocrinology Unit. The obese group was divided into two subgroups: NAFLD patients (25 girls and 33 boys; mean age, 13.13 ± 1.26 years; mean BMI, 30.36 ± 3.08 kg/m^2) with high alanine aminotransferase (ALT) levels (>40 U/L) and ultrasound evidence of fatty changes in the liver (group 1) and non-NAFLD patients (32 girls and 21 boys; mean age, 13.25 ± 1.34 years; mean BMI, 29.33 ± 3.47 kg/m^2) with low ALT levels (<40 U/L) and no ultrasound evidence of fatty changes in the liver (group 2). Lean adolescents (17 girls and 25 boys; mean age, 13.65 ± 1.4 years; range 12–17 years) with a mean BMI of 19.41 ± 2.21 kg/m^2 were enrolled in the study from non-obese healthy adolescents who attended the hospital for minor illnesses such as common cold or conjunctivitis.

The study protocols were approved by the institutional review board of the Konya Research Hospital Ethical Committee. Signed informed consent forms were obtained from the parents of the adolescents.

Patients were excluded from the study if they had any systemic disease, including type 1 or 2 diabetes mellitus, were taking medications, or had a condition known to effect insulin action or insulin secretion (e.g., glucocorticoid therapy, Cushing's disease). We also excluded all patients who had any previous diagnosis or treatment for thyroid dysfunction.

Subclinical hypothyroidism was defined as a TSH level exceeding 4.0 mIU/l and a normal free thyroxine level (0.6–1.8 ng/dl). According to data from the National Health and Nutrition Examination Survey (NHANES), in a disease-free population, the TSH values between percentiles 2.5 and 97.5 ranged from 0.45 to 4.17 mIU/l. Based on these data, we chose our cutoffs to be values less than 0.45 mIU/l and greater than 4.0 mIU/l [11].

Anthropometric measurements of all the patients were performed. Height and weight were measured with an empty bladder in postabsorptive conditions. The height was measured to the nearest 0.5 cm on a standard height board, and the weight was determined to the nearest 0.1 kg on a standard physician's beam scale, with the subject dressed only in light underwear without shoes. The BMI was calculated as weight (kg) divided by height (m) squared. Patients with a BMI at the 95th percentile or higher according to reference curves for Turkish adolescents were accepted as obese [23]. Pubertal development stage was assessed by a single pediatric endocrinologist using the Tanner criteria, and the staging for sexual maturation exceeded two for all the patients (Tanner stages 2–4).

After the child had rested for at least 5 min and was in a sitting position, diastolic and systolic pressure (mmHg) measurements were taken using a mercury-gravity manometer and a cuff appropriate for body size.

Laboratory Assessment

Fasting blood samples were obtained by venipuncture in the morning at 8 a.m., after an overnight fast of at least 12 h, to measure serum glucose, insulin levels, and other parameters. Glucose was determined by the glucose oxidase method.

Serum concentrations of total cholesterol, high-density lipoprotein (HDL) cholesterol, and triglycerides were measured using routine enzymatic methods with the (Abbott diagnostics C16000 chemistry analyzer, Illinois, USA). The value of low-density lipoprotein (LDL) cholesterol was calculated using Friedwald's equation.

Serum insulin levels were measured by an immulite immunoassay (Diagnostic Products, Los Angeles, CA, USA). The TSH level (reference range 0.45–4.0 mIU/l) was measured by means of a chemiluminescence immunoassay (ADVIA Centaur XP, Immunoassay System; Siemens Healthcare Diagnostic Inc, Tarrytown, NY, USA). Free thyroxine was determined using a chemiluminescence immunoassay (reference range 0.6–1.8 ng/dl). Standard

liver function tests (ALT, aspartate aminotransferase [AST]) were performed on the same day using an autoanalyzer.

Insulin resistance was estimated using the homeostasis model assessment for insulin resistance (HOMA-IR) as follows: fasting insulin concentration (μU/mL) × fasting glucose concentration (mmol/l)/22.5 [20]

Insulin resistance was defined for adolescents as levels of the HOMA-IR greater than 3.16 [13].

Liver Ultrasonography

All the patients with abnormally high transaminases and an abnormal liver ultrasound were screened for other liver conditions (hepatitis B surface antigen, hepatitis C antibody, prothrombin time, iron, total iron-binding capacity, ferritin, and antinuclear antibodies), which all showed negative results. Liver ultrasound was performed by a trained operator blinded to all clinical and laboratory characteristics of the participants. Scans were performed for all the subjects using a GE Healthcare Logic 7 machine (GE Healthcare Logic; Livonia, MI, USA) equipped with 7.5-MHz probes for younger adolescents and 5-MHz probes for larger or markedly obese adolescents.

The presence of NAFLD was assessed via the scoring system defined by Tominaga et al. [35] according to the hyperechogenicity of liver tissue, the discrepancy between the liver and the diaphragm, and the visibility of vascular structures. The diagnosis of NAFLD usually is made from mild elevations of liver enzymes during a routine blood test and liver ultrasonography of an overweight or obese child. Although liver ultrasonography cannot estimate either fibrosis or inflammation, it has a sensitivity of 89 % and a specificity of 93 % for detecting histologic steatosis [32]. Elevated ALT was defined as more than 40 IU/l in our study [2]. In the current study, all the obese patients with NAFLD had high ALT levels.

Carotid IMT Measurements

Carotid ultrasound studies were performed by a single radiologist blinded to the clinical and laboratory status of the patients using high-resolution B-mode ultrasonography (GE Healthcare Logic 7) with a high-resolution linear array vascular transducer (14 MHz). An optimal two-dimensional image of the common carotid artery was obtained in which both the near and far wall intima/media complexes were well visualized.

After the subject had rested 10 min and following standard guidelines, the M-mode curser was placed 1 cm proximal to the beginning of the carotid artery bulb during end diastole. Carotid IMT was calculated by taking the mean value of three measurements.

Echocardiographic Examination

All echocardiographic and Doppler assessments were performed by the same expert pediatric cardiologist, who was blinded to the clinical and laboratory results of the study group. ProSound Alpha 7 echocardiography equipment (Aloka, Hitachi-Aloka Medical, Tokyo, Japan) with a 3-MHz phased-array transducer was used for each study subject.

The patients were examined in the left lateral decubitus position, and images were acquired at passive end expiration to minimize global cardiac movement from standard parasternal long-axis and apical planes. The M-mode echocardiographic study of the left ventricle (LV) was performed under two-dimensional control. The ventricular septal and posterior wall thicknesses at end diastole as well as the dimensions of LV end-diastole (LVED) and end-systole were determined from M-mode echocardiography according to the American Society of Echocardiography recommendations. The calculations of LVM were performed using the following equation [7]:

$$0.8\{1.04[(\text{LVED} + \text{LV posterior wall thickness} + \text{interventricularseptal thickness})^3 - \text{LVED}^3]\} + 0.6 \text{ and indexed for height}^{2.7}.$$

Statistical Analysis

Mean and standard deviations were used as descriptive statistics. Obese and normal-weight groups were compared with respect to continuous variables using Student's *t*-test. Bivariate associations of continuous variables were assessed with Pearson correlation coefficients. Normality assumptions were assessed before parametric tests were used. Multivariable stepwise regression analysis was performed to assess independent predictors of TSH levels. A p value lower than 0.05 was used to indicate statistical significance. Statistical analyses were performed with SPSS for Windows, version 15 (SPSS, Chicago, IL, USA).

Results

The characteristics of the study population are shown in Table 1. Gender, age, and BMI were similar in the two groups. The control group (lean) included 42 sex-, age-, and pubertal stage-matched nonobese healthy subjects without liver steatosis.

The NAFLD obese group had significantly higher systolic and diastolic blood pressures than the non-NAFLD and lean groups. The NAFLD obese group also had significantly higher levels of AST, ALT, total cholesterol, LDL cholesterol, triglycerides, fasting insulin, and TSH

Table 1 Characteristics of lean and obese adolescents[a]

	Lean	Obese	
		Non-NAFLD	NAFLD
N (Girl/boy ratio)	42 (17/25)	53 (32/21)	58 (25/33)
Age (years)	13.65 ± 1.4 (13)	13.25 ± 1.34 (13)	13.13 ± 1.26 (13)
BMI (kg/m^2)	19.41 ± 2.21 (19.24)	29.33 ± 3.47 (28.53)[a]	30.36 ± 3.08 (30.35)[b]
Systolic blood pressure (mmHg)	102.9 ± 11.4 (100)	112.6 ± 11.6 (110)[a]	119.3 ± 13.8 (120)[b, c]
Diastolic blood pressure (mmHg)	65.2 ± 8.0 (70)	71.9 ± 9.8 (70)[a]	76.9 ± 9.6 (80)[b, c]
Total cholesterol (mg/dl)	138.2 ± 18.6 (137.5)	170.7 ± 23.8 (168)[a]	190.8 ± 29.1 (186)[b, c]
Triglycerides (mg/dl)	75.2 ± 28.7 (70)	137.8 ± 65.9 (121)[a]	180.8 ± 73.4(170.5)[b, c]
LDL (mg/dl)	75.5 ± 17.3 (70)	100.4 ± 22.3 (101.4)[a]	117.2 ± 28.9 (113.4)[b, c]
HDL (mg/dl)	47.6 ± 8.2 (47)	43.3 ± 7.66 (43)[a]	39.3 ± 6.5 (38.5)[b, c]
Fasting glucose (mg/dl)	76.7 ± 7.5 (75.5)	88.7 ± 9.6 (89)[a]	91.7 ± 6.5 (92)[b]
Fasting insulin (IU/ml)	3.9 ± 1.9 (4.0)	16.2 ± 10.3 (12.5)[a]	22.2 ± 9 (21)[b, c]
Free T4 (ng/dl)	1.25 ± 0.15 (1.27)	1.16 ± 0.13 (1.15)[a]	1.20 ± 0.15 (1.18)
TSH (mIU/l)	1.77 ± 0.63 (1.64)	2.86 ± 1.13 (2.61)[a]	3.56 ± 1.34 (3.46)[b, c]
AST (IU/l)	16.7 ± 2.9 (16)	23.6 ± 5 (24)[a]	41.29 ± 3.3 (40.5)[b, c]
ALT (IU/l)	14.2 ± 5.5 (13)	23.2 ± 8.6 (22)[a]	45.3 ± 3.5 (45)[b, c]
HOMA-IR	0.76 ± 0.29 (0.77)	3.59 ± 2.53 (2.85)[a]	5.14 ± 2.41 (4.66)[b, c]
AST/ALT ratio	1.31 ± 0.41 (1.27)	1.10 ± 0.31 (1)[a]	0.911 ± 0.057 (0.912)[b, c]
Carotid IMT (mm)	0.358 ± 0.011 (0.360)	0.380 ± 0.018 (0.380)[a]	0.440 ± 0.025 (0.445)[b, c]
LVM (g)	117.7 ± 35.2 (118.4)	156.4 ± 45.3 (147.8)[a]	177.9 ± 38.3 (176.5)[b, c]
LVM index (g/m$^{2.7}$)	33.3 ± 10.8 (34.7)	46.1 ± 10.9 (44.3)[a]	49.5 ± 8.6 (48.9)[b]

Data were expressed as the mean ± standard deviation with median given in parentheses

NAFLD nonalcoholic fatty liver disease; *BMI* body mass index; *LDL* low-density lipoprotein; *HDL* high-density lipoprotein; *TSH* thyroid-stimulating hormone; *AST* aspartate transaminase; *ALT* alanine transaminase; *HOMA-IR* homeostasis model assessment for insulin resistance; *IMT* intima-media thickness; *LVM* left ventricular mass

[a] Lean versus obese without NAFLD ($p < 0.05$)

[b] Lean versus obese with NAFLD ($p < 0.05$)

[c] Obese without NAFLD versus obese with NAFLD ($p < 0.05$)

than the non-NAFLD and lean groups, whereas the HDL cholesterol and AST/ALT ratio values were lower in the NAFLD obese group than in the non-NAFLD and lean groups. The non-NAFLD obese group had significantly higher BMI and systolic and diastolic blood pressures than the control group. The levels of AST, ALT, total cholesterol, triglyceride, LDL cholesterol, fasting glucose, insulin, and TSH also were significantly higher in the non-NAFLD obese group than in the control group, whereas the HDL cholesterol levels were lower in the non-NAFLD obese group than in the control group.

The lean group had lower HOMA-IR values than the non-NAFLD and NAFLD obese groups (respectively, 0.76 ± 0.29 vs 3.59 ± 2.53 vs 5.14 ± 2.41). Moreover, the NAFLD obese group compared with the non-NAFLD and lean groups had significantly higher TSH values (3.56 ± 1.34 vs 2.86 ± 1.13 vs 1.77 ± 0.63 mIU/l) and carotid IMT (0.440 ± 0.025 vs 0.380 ± 0.018 vs 0.358 ± 0.011 mm), as well as a higher LVM (177.9 ± 38.3 vs 156.4 ± 45.3 vs 117.7 ± 35.2 g) (Table 1).

The levels of AST, fasting glucose, and insulin also were significantly higher in the NAFLD obese group with a TSH level higher than 4 mIU/l than the NAFLD obese group with a TSH level lower than 4 mIU/l, whereas the HDL cholesterol levels were lower in the NAFLD obese group with a TSH level exceeding 4 mIU/l than in the NAFLD obese group with a TSH level lower than 4 mIU/l. The NAFLD obese group with TSH level higher than 4 mIU/l had significantly higher HOMA-IR values (6.24 ± 2.89 vs. 3.59 ± 2.53), carotid IMT (0.454 ± 0.026 vs 0.430 ± 0.018 mm), LVM (195.4 ± 38.4 vs 164.6 ± 32.9 g), and LVM index (52.5 ± 7.2 vs 47.2 ± 9.0 g/m$^{2.7}$) than the NAFLD obese group with a TSH level lower than 4 mIU/l (Table 2).

In the NAFLD obese group with subclinical hypothyroidism, high TSH values were positively correlated with most of the metabolic parameters such as total cholesterol ($r = 0.606$, $p = 0.001$), triglycerides ($r = 0.476$, $p = 0.016$), LDL cholesterol ($r = 0.461$, $p = 0.004$), insulin ($r = 0.607$, $p = 0.001$), AST ($r = 0.467$, $p = 0.019$), HOMA-IR

Table 2 Characteristics obese adolescents with nonalcoholic fatty liver disease (NAFLD) according to serum thyroid-stimulating hormone (TSH) levels

	Obese with NAFLD		
	TSH (<4 mIU/l)	TSH (>4 mIU/l)	*p* Value
N	33	25	
Age (years)	12.97 ± 1.15 (12.5)	13.35 ± 1.38 (13)	0.259
BMI (kg/m^2)	30.26 ± 3.02 (30.26)	30.49 ± 3.22 (30.40)	0.777
Systolic blood pressure (mmHg)	118.9 ± 12.2 (120)	119.8 ± 15.97 (120)	0.817
Diastolic blood pressure (mmHg)	76.2 ± 8.2 (80)	77.8 ± 11.3 (80)	0.541
Total cholesterol (mg/dl)	186.7 ± 16 (184)	191.4 ± 29.3 (186)	0.222
Triglycerides (mg/dl)	177.4 ± 85.3 (164)	176.3 ± 73.1(172)	0.684
LDL (mg/dl)	113.7 ± 18.3 (113)	118.2 ± 29.1 (117.6)	0.293
HDL (mg/dl)	40.9 ± 6.3 (40)	39.9 ± 7.1 (35)	0.031
Fasting glucose (mg/dl)	89.8 ± 5.9 (91)	94.2 ± 6.5 (99)	0.010
Fasting insulin (IU/ml)	19.1 ± 6 (17.1)	26.3 ± 10.6 (25)	0.002
Free T4 (ng/dl)	1.21 ± 0.16 (1.26)	1.17 ± 0.14 (1.11)	0.344
TSH (mIU/l)	2.56 ± 0.72 (2.85)	4.89 ± 0.59 (4.94)	<0.0001
AST (IU/l)	39.7 ± 3.1 (39)	43.2 ± 2.5 (44)	<0.0001
ALT (IU/l)	44.8 ± 4 (43.8)	45.9 ± 2.8 (46)	0.267
HOMA-IR	3.59 ± 2.53 (2.85)	6.24 ± 2.89 (6.11)	0.002
AST/ALT ratio	0.888 ± 0.05 (0.902)	0.940 ± 0.037 (0.955)	<0.0001
Carotid IMT (mm)	0.430 ± 0.018 (0.430)	0.454 ± 0.026 (0.455)	<0.0001
LVM (g)	164.6 ± 32.9 (158.2)	195.4 ± 38.4 (200.4)	0.002
LVM index (g/m$^{2.7}$)	47.2 ± 9.0 (46.4)	52.5 ± 7.2 (51.1)	0.019

Data are expressed as the mean ± standard deviation, with the median given in parentheses

BMI body mass index; *LDL* low-density lipoprotein; *HDL* high-density lipoprotein; *AST* aspartate transaminase; *ALT* alanine transaminase; *HOMA-IR* homeostasis model assessment for insulin resistance; *IMT* intima-media thickness; *LVM* left ventricular mass

($r = 0.596, p = 0.002$), carotid IMT ($r = 0.894, p < 0.0001$), and LVM ($r = 0.563, p = 0.003$) (Table 3; (Figs. 1, 2, 3).

In the multivariate stepwise regression analysis, the TSH level was positively correlated with increased carotid IMT ($\beta = 0.243, p = 0.001$) in the NAFLD obese group with TSH exceeding 4 mIU/l even after adjustment for age, sex, BMI, systolic and diastolic blood pressures, total cholesterol, triglycerides, LDL cholesterol, HDL cholesterol, fasting glucose and insulin, AST/ALT ratio, and HOMA-IR as cofactors, with the total variance explained as 79.8 % (Table 4, 5).

Discussion

This study demonstrated that a significant relationship exists between subclinical hypothyroidism and cardiac geometry in obese adolescents with NAFLD and obesity-related cardiovascular risk factors. We suggest that the presence of subclinical hypothyroidism may have an additional increasing impact on LVM and carotid IMT in these patients with NAFLD.

Some studies support an association between overt or subclinical thyroid disease and the development of primary NAFLD [5, 16]. Liangpunsakul and Chalasani [16] reported the prevalence of hypothyroidism in 174 patients with nonalcoholic steatohepatitis (NASH) to be double that seen in 442 matched control subjects. In a large population study involving 10,292 outpatient adults with a wide range of age and thyroid function tests (TSH and fT4), serum ALT and gamma-glutamyl transferase concentrations increased steadily across the increasing TSH categories. Likewise, a negative and graded relationship was found between serum ALT and gamma-glutamyl transferase concentrations and fT4 categories [33].

In a recent study, Carulli et al. [5] found that euthyroid patients with biopsy-proven NASH have high (although still in the upper normal range) TSH values compared with those who have simple steatosis. Although the association between thyroid hormonal derangements and fatty liver is established, which one causes the other still is unclear [25]. In our multivariate regression analysis, all the metabolic variables showed TSH levels positively correlated with carotid IMT and LVM in the NAFLD obese group with a TSH level higher than 4 mIU/l.

Thyroid hormones may have a negative effect on the major metabolic pathways, and thyroid dysfunction may

Table 3 Relationship between carotid intima-media thickness (IMT) and cardiovascular risk factors in obese adolescents with nonalcoholic fatty liver disease (NAFLD)

	Obese with NAFLD	
	r	*p* Value
Age (years)	0.300	0.022
BMI (kg/m^2)	0.319	0.018
Systolic blood pressure (mmHg)	0.445	<0.0001
Diastolic blood pressure (mmHg)	0.505	<0.0001
Total cholesterol (mg/dl)	0.583	<0.0001
Triglycerides (mg/dl)	0.120	0.371
LDL (mg/dl)	0.551	<0.0001
HDL (mg/dl)	−0.328	0.012
Fasting glucose (mg/dl)	0.546	<0.0001
Fasting insulin (IU/ml)	0.724	<0.0001
Free T4	−0.256	0.052
TSH	0.598	<0.0001
AST (IU/l)	0.551	<0.0001
ALT (IU/l)	0.220	0.01
HOMA-IR	0.722	<0.0001
AST/ALT ratio	0.495	<0.0001
LVM (g)	0.686	<0.0001
LVM index (g/m$^{2.7}$)	0.469	<0.0001

BMI body mass index; *LDL* low-density lipoprotein; *HDL* high-density lipoprotein; *TSH* thyroid-stimulating hormone; *AST* aspartate transaminase; *ALT* alanine transaminase; *HOMA-IR* homeostasis model assessment for insulin resistance; *LVM* left ventricular mass

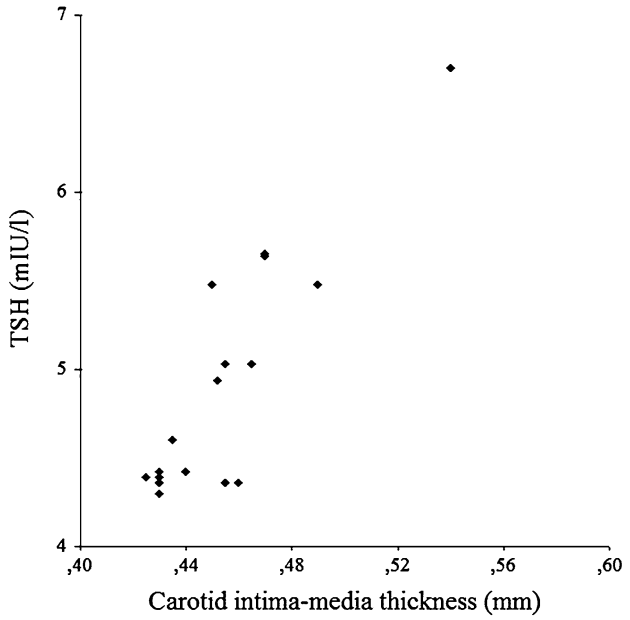

Fig. 1 Relationship between thyroid-stimulating hormone (TSH) level and carotid intima media thickness in obese patients with nonalcoholic fatty liver disease (NAFLD) whose TSH levels are higher than 4 mIU/l

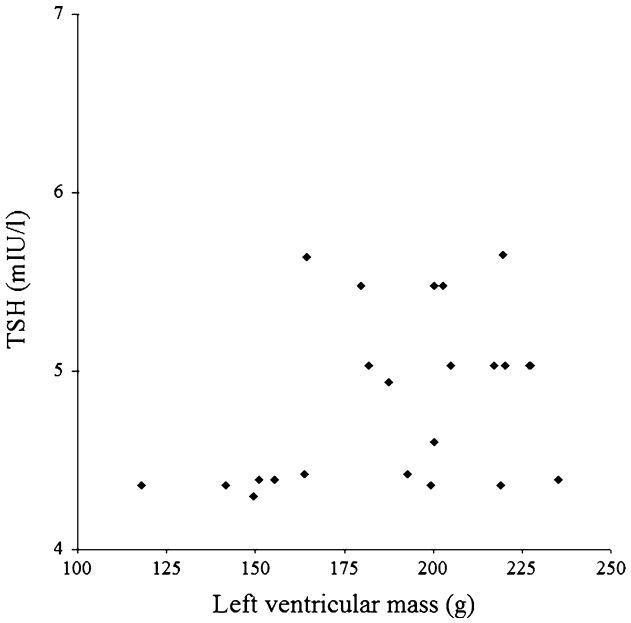

Fig. 2 Relationship between thyroid-stimulating hormone (TSH) level and left ventricular mass in obese patients with nonalcoholic fatty liver disease (NAFLD) whose TSH levels are higher than 4 mIU/l

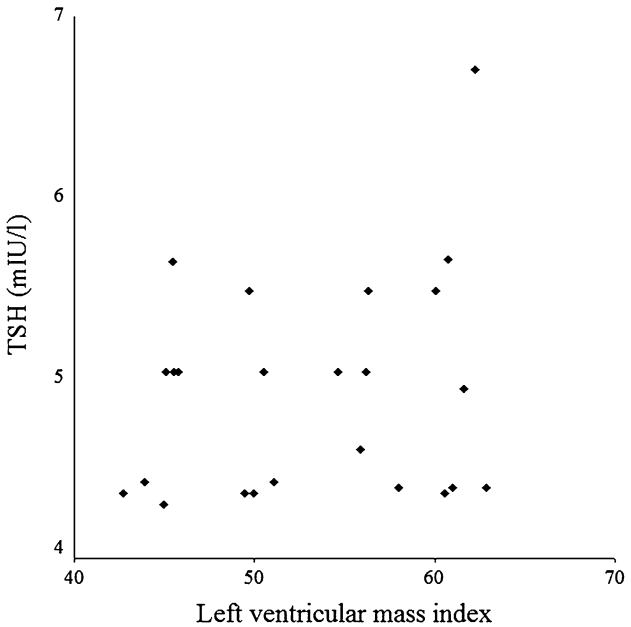

Fig. 3 Relationship between thyroid-stimulating hormone (TSH) level and left ventricular mass index in obese patients with nonalcoholic fatty liver disease (NAFLD) whose TSH levels are higher than 4 mIU/l

play a role in the pathogenesis of NAFLD. Patients with hypothyroidism not only have evidence of hyperlipidemia, but also exhibit decreased fatty acid oxidation and hepatic output of triglycerides. Otherwise, hypothyroidism is associated with insulin resistance, which seems to be a

Table 4 Relationship between left ventricular mass (LVM) and cardiovascular risk factors in obese adolescents with nonalcoholic fatty liver disease (NAFLD)

	Obese with NAFLD	
	r	*p* Value
Age (years)	0.319	0.015
BMI (kg/m^2)	0.326	0.013
Systolic blood pressure (mmHg)	0.457	<0.0001
Diastolic blood pressure (mmHg)	0.486	<0.0001
Total cholesterol (mg/dl)	0.454	<0.0001
Triglycerides (mg/dl)	0.075	0.578
LDL (mg/dl)	0.441	0.001
HDL (mg/dl)	−0.261	0.047
Fasting glucose (mg/dl)	0.445	<0.0001
Fasting insulin (IU/ml)	0.709	<0.0001
Free T4	−0.241	0.069
TSH	0.476	<0.0001
AST (IU/l)	0.387	0.003
ALT (IU/l)	0.124	0.353
HOMA-IR	0.695	<0.0001
AST/ALT ratio	0.377	0.004
LVM index (g/m$^{2.7}$)	0.719	<0.0001

BMI body mass index; *LDL* low-density lipoprotein; *HDL* high-density lipoprotein; *TSH* thyroid-stimulating hormone; *AST* aspartate transaminase; *ALT* alanine transaminase; *HOMA-IR* homeostasis model assessment for insulin resistance

Table 5 Multiple regression analysis of determinants of carotid intima-media thickness (IMT) and left ventricular mass (LVM) in the obese adolescents with nonalcoholic fatty liver disease (NAFLD) and subclinical hypothyroidism

	R^2	R^2 Change	β	95 % CI	*p* Value
Carotid IMT versus variable					
TSH level	0.791	0.800	0.894	0.4–10.3	0.001
LVM versus variable					
BMI	0.566	0.113	0.368	0.58–8.1	0.026
HOMA-IR			0.523	2.6–11.1	0.003

CI confidence interval; *TSH* thyroid-stimulating hormone; *BMI* body mass index; *HOMA-IR* homeostasis model assessment for insulin resistance

hallmark of NASH. Finally, hypothyroidism is associated with lipid peroxidation, one of the leading candidates for cellular injury in patients with NASH [25].

In obese children, a relationship between TSH levels, triglycerides, total cholesterol, and LDL cholesterol have been reported in some but not all studies [25]. Paoli-Valeri et al. [26] compared the lipid profile between 2- to 9-year-old Spanish children with subclinical hypothyroidism (TSH > 4.65 mIU/L and normal T4) and healthy control subjects. The mean HDL cholesterol was significantly lower in the children with subclinical hypothyroidism.

In our study, the NAFLD obese group had significantly higher levels of total cholesterol, LDL cholesterol, and triglycerides than the non-NAFLD and lean groups. Furthermore, the HDL cholesterol levels were lower in the NAFLD obese group with TSH levels higher than 4 mIU/l than in the NAFLD obese group with TSH levels lower than 4 mIU/l. The findings showed TSH values positively correlated with most of the metabolic parameters such as total cholesterol, triglycerides, LDL cholesterol in the NAFLD obese group with TSH levels higher than 4 mIU/l.

An increased risk of hypertension also has been reported in some studies of patients with subclinical hypothyroidism. Because it is in overt disease, increased peripheral vascular resistance, increased arterial stiffness, and endothelial dysfunction can contribute to systemic hypertension in subclinical hypothyroidism [25]. In our study, the NAFLD obese group had significantly higher systolic and diastolic blood pressures than those in the non-NAFLD and lean groups.

Thyroid hormones are important determinants of glucose homeostasis. Increases in plasma thyroid hormone levels impair the ability of insulin to suppress hepatic glucose production and to increase glucose uptake in muscle [25]. Maratou et al. [19] showed that patients with subclinical hypothyroidism had insulin resistance.

Studies of children are limited. In euthyroid children without a history of hypo- or hyperthyroidism, increasing levels of TSH and decreasing levels of free T4 are reported to be associated with elevated markers of insulin resistance [25]. A recent study of obese children showed that during weight loss, independent of changes in body weight or body fat and lean tissue decreases in elevated serum TSH predict decreases in fasting insulin and insulin resistance [1].

In our study, the NAFLD obese group had significantly higher HOMA-IR values than the non-NAFLD and lean groups. Moreover, the HOMA-IR values were significantly higher in the NAFLD obese group with TSH levels higher than 4 mIU/l than in the NAFLD obese group with TSH levels lower than 4 mIU/l. The TSH values were positively correlated with HOMA-IR in the NAFLD obese group with TSH levels higher than 4 mIU/l. These findings could justify the increased risk for insulin resistance–associated disorders such as the cardiovascular disease observed in NAFLD patients with subclinical hypothyroidism.

In the current study, we found marked abnormalities in LV structure including increased LVM and LVM index in the NAFLD obese adolescents with TSH levels higher than 4 mIU/l. Monzani et al. [21] noted significantly higher values for LVM in 20 patients with subclinical hypothyroidism than in control subjects, similar to the findings of

our study. However, other studies have reported no significant associations between subclinical hypothyroidism and LVM [3, 27, 39]. Rodondi et al. [29] observed an increase in LVM over 5 years in the subgroup of subclinical hypothyroidism with TSH levels of 10.0 mU/l or higher. However, an epidemiologic study performed by Iqbal et al. [12] that included more than 2,000 subjects could not find any significant association between serum TSH and LVM index after adjustment for age, BMI, and systolic blood pressure, which is in accordance with the report by Dörr et al. [8]. In their study of 1,510 individuals, the adjusted LVM index was almost identical in subjects with elevated, normal, and decreased serum TSH levels, whereas those with overt hyperthyroidism had an increased LVM index.

Carotid IMT is a reliable index of subclinical atherosclerosis, and epidemiologic studies have demonstrated a significant association between carotid IMT and cardiovascular disease [30]. No study has assessed whether subclinical hypothyroidism is associated with carotid atherosclerosis in children or adolescents with subclinical hypothyroidism, but this association is shown in adult studies. Valentina et al. [36] showed that subclinical hypothyroidism is associated with an increase in carotid IMT, independently of classical risk factors for atherosclerosis.

Kim et al. [14] studied to determine whether subclinical hypothyroidism is associated with an increase in the IMT of the common carotid artery and whether thyroid hormone replacement can reverse this change in the carotid IMT. Recently, in a double-blind placebo-controlled study, Monzani et al. [22] demonstrated that early carotid IMT alterations also were present in patients with subclinical hypothyroidism, mainly appearing during the fourth decade of life and onward.

Furthermore, several factors may contribute to cardiovascular risk in subclinical hypothyroidism including BMI, fat distribution, lipid profile, and vascular dysfunction [9]. A population-based survey concluded that subclinical hypothyroidism was associated with aortic atherosclerosis and myocardial infarction in adults, independently of serum cholesterol levels [10].

Conclusion

The current study confirms that obese adolescents who have NAFLD with subclinical hypothyroidism demonstrate a more adverse cardiovascular risk profile and higher LVM. Increased LVM and carotid IMT are significant predictors of cardiovascular-related death. Addition of carotid IMT measurements to the echocardiographic examination may assist in risk stratification of NAFLD obese adolescents with subclinical hypothyroidism.

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