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Original Contribution

Cardiovascular Risk Factors, Nonalcoholic Fatty Liver Disease, and Carotid Artery Intima-Media Thickness in an Adolescent Population in Southern Italy

Carmelo Antonio Caserta, Gaspare Maria Pendino, Angela Amante, Carmelo Vacalebre, Maria Teresa Fiorillo, Pasquale Surace, Arianna Messineo, Monica Surace, Saverio Alicante, Rodolfo Cotichini, Massimo Zuin, Francesco Rosmini, Alfonso Mele, and Fabrizio Marcucci*

* Correspondence to Dr. Fabrizio Marcucci, Centro Nazionale di Epidemiologia, Sorveglianza e Promozione della Salute, Istituto Superiore di Sanità, Via Giano della Bella 34, 00162 Roma, Italy (e-mail: fabmarcu@tin.it).

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The objective of this study was to determine, in an adolescent population, the prevalence of nonalcoholic fatty liver disease (NAFLD) and the association of NAFLD and cardiovascular risk factors with carotid artery intima-media thickness (IMT), a marker of subclinical atherosclerosis. The authors conducted a population-based study among 642 randomly selected adolescents aged 11–13 years in Reggio Calabria, southern Italy, between November 2007 and October 2008. Prevalences of overweight and obesity were 30.5% and 13.5%, respectively. The overall prevalence of NAFLD was 12.5%, increasing to 23.0% in overweight/obese adolescents. In univariate analysis, increased IMT was positively associated with the presence of NAFLD, body mass index (BMI), waist circumference, systolic blood pressure (all P's < 0.001), diastolic blood pressure ($P = 0.006$), γ-glutamyl transpeptidase ($P = 0.006$), alanine aminotransferase ($P = 0.007$), and C-reactive protein ($P = 0.008$) and was inversely associated with high density lipoprotein cholesterol ($P < 0.001$). In multivariate analysis, NAFLD ($P = 0.002$), BMI ($P = 0.004$), waist circumference ($P = 0.003$), and systolic blood pressure ($P = 0.005$) retained significant associations. The authors conclude that NAFLD, BMI, waist circumference, and systolic blood pressure are independent markers of increased IMT in a random sample of adolescents.

adolescent; atherosclerosis; blood pressure; body mass index; fatty liver; liver diseases; obesity; sex

Abbreviations: CI, confidence interval; HOMA-IR, homeostasis model assessment of insulin resistance; IMT, intima-media thickness.

Nonalcoholic fatty liver disease (hereafter called fatty liver disease) is a spectrum of clinicopathologic conditions characterized by excessive accumulation of fat in the liver parenchyma of patients who have no history of alcohol abuse. Within this spectrum, steatosis alone is apparently benign, while nonalcoholic steatohepatitis, including liver-cell inflammation and fibrosis, may progress to cirrhosis, liver failure, and hepatocellular carcinoma (1).

Fatty liver disease is strongly associated with obesity, type 2 diabetes, and dyslipidemia, and most patients have evidence of insulin resistance (2, 3). These are typical features of the metabolic syndrome, and in fact, several authors consider fatty liver disease to be the hepatic expression of the metabolic syndrome (4). The constellation of metabolic abnormalities that contribute to the definition of metabolic syndrome are well-documented risk factors for cardiovascular disease (5).

Given that fatty liver disease is associated with the abnormalities of the metabolic syndrome, it would be important to know whether fatty liver disease is an independent risk factor for cardiovascular disease. Several studies have addressed this issue and found evidence that, indeed, fatty liver disease is a significant risk factor for cardiovascular disease (6–13). Most of these studies used as the outcome carotid artery intima-media thickness (IMT) as a noninvasive marker of subclinical atherosclerosis. Increased IMT is associated with the presence and severity of coronary atherosclerosis and cardiovascular disease (14–16).

However, most studies examining the relation of fatty liver disease to IMT have used a case-control design, which is vulnerable to temporal and other biases. Only a very small number of population-based studies have been reported (13, 17). The few studies performed in adolescent populations (7, 18) have all been case-based, rather than population-based, case-control studies.

Here we present results of a study carried out in randomly selected adolescents from Reggio Calabria, a town in southern Italy. The aim of the study was to determine the prevalence of fatty liver disease and its association with carotid artery IMT. To our knowledge, this is the first study of its kind to be performed in a general adolescent population.

MATERIALS AND METHODS

Setting and target population

The study was conducted by the Hepatology Association of Calabria, in collaboration with the Istituto Superiore di Sanità (Italy's Advanced Health Institute). The study protocol was approved by the ethics committees of the participating institutions.

The study was conducted from November 2007 through October 2008 in Reggio Calabria, a town of approximately 185,000 inhabitants in the Calabria region of southern Italy. A sample of 843 adolescents aged 11–13 years was randomly selected from an updated school census list. Written informed consent was obtained from the parents of all adolescents who agreed to participate in the study.

Administration of questionnaire

At enrollment, a standardized, precoded questionnaire on sociodemographic characteristics and medical history was administered by trained health-care professionals. The questionnaire included questions on alcohol consumption—specifically the type and amount of alcohol consumed and the duration of consumption. Based on the alcohol content of the beverages reported, mean daily alcohol intake was calculated and expressed in grams per day. Participants who consumed at least 12.5 g of alcohol per day (half the amount considered the upper normal limit for adults (19)) and those with chronic use of potentially hepatotoxic medications were excluded from the analysis.

The questionnaire also included questions on smoking. Eight subjects answered that they smoked but had not smoked for longer than 1 year; and 5 of them claimed to smoke occasionally, 3 on a regular basis, but not more than 10 cigarettes per day. Given the small number of smokers, their recent initiation of smoking, and their relatively modest use of cigarettes, we considered it unlikely that smoking could represent a cardiovascular risk factor in these subjects. Therefore, we did not consider smoking further in our analysis.

No information about menarche status was collected.

Physical examination

Weight and height were measured without shoes and with light clothing, and body mass index was calculated as body weight (kg) divided by the square of height (m^2). Waist circumference was measured, during expiration, at the narrowest point between the lower rib and the iliac crest. Overweight and obesity were defined according to the International Obesity Task Force criteria (20). Blood pressure was measured 3 times, after the subject had been seated for at least 10 minutes. The last 2 measurements were averaged for analysis.

Biochemistry

Blood samples were collected in the morning after an overnight fast. All blood tests were conducted in the same laboratory. Blood was tested within a few hours of sampling. Fasting levels of glucose, triglycerides, total cholesterol, and high density lipoprotein cholesterol were determined in dry chemistry on a Vitros 950 automatic analyzer (Ortho Clinical Diagnostics, Raritan, New Jersey), and C-reactive protein level was determined with the BN PROSPEC nephelometer (Dade Behring Marburg GmbH, Marburg, Germany). Insulin was measured by microparticle enzyme immunoassay (Abbott Laboratories, Abbott Park, Illinois). Low density lipoprotein cholesterol was calculated using the formula of Friedewald (21). Insulin resistance (homeostasis model assessment of insulin resistance (HOMA-IR)) was calculated as insulin (mU/L) $\times$ [glucose (mg/dL) $\times$ 0.055]/22.5 (22).

Liver ultrasonography

Adolescents underwent abdominal ultrasonography according to a standardized protocol. Real-time ultrasonography was performed using a MyLab 25 ultrasound scanner (Esaote S.p.A., Genoa, Italy), with a 3.5-MHz convex transducer. Technical parameters were optimized on a case-by-case basis. All examinations were carried out and interpreted by the same sonographer. Sonographic findings were categorized into the presence or absence of steatosis. Diagnosis of steatosis was based on the presence of brightness with clear-cut sonographic contrast between the liver and the right renal cortex in the midaxillary line, according to international guidelines (23, 24). In cases of uncertain diagnosis, additional criteria, such as vessel blurring, posterior beam attenuation, and no visualization of the diaphragm were used. No graduation in the severity of steatosis was considered in this study.

Since the sonographer who assessed fatty liver disease could not be blinded to the weight status of the study subjects, she was asked to reexamine 100 anonymous ultrasonograms after 1 month. The initial diagnoses were confirmed.

Carotid artery ultrasonography

Carotid artery ultrasonography was performed using a 10-MHz linear transducer on the MyLab 25 scanner (Esaote). The protocol involved scanning of both common carotid arteries. Subjects were examined by 1 trained operator following a standardized protocol (25). Briefly, IMT was assessed at the far wall as the distance between the

Table 1. Characteristics of Study Participants (642 Randomly Selected Adolescents Aged 11–13 Years), Reggio Calabria, Southern Italy, 2007–2008

Characteristic	No. of Subjects[a]	Mean (SD)	Median	90% Range
Intima-media thickness, mm	642	0.397 (0.034)	0.40	0.34–0.46
Body mass index[b]	642	21.3 (4.2)	20.7	15.7–29.5
Waist circumference, cm	641	79.3 (11.6)	78	63–99
Systolic blood pressure, mm Hg	642	105 (11.6)	105	87–124
Diastolic blood pressure, mm Hg	642	65 (7.9)	65	53–80
Alanine aminotransferase, IU/L	572	31 (8.4)	30	22–43
γ-Glutamyl transpeptidase, IU/L	572	14 (5.1)	13	9–23
Total cholesterol, mg/dL	572	158 (26.5)	156	118–205
High density lipoprotein cholesterol, mg/dL	572	51 (11.9)	50	34–73
Low density lipoprotein cholesterol, mg/dL	572	92 (23.9)	89	60–134
Triglycerides, mg/dL	572	74 (35.5)	66	35–140
Glucose, mg/dL	572	83 (6.9)	82	72–94
Insulin, mIU/L	572	9.8 (6.6)	8.4	3.6–19.7
Homeostasis model assessment of insulin resistance	572	2.0 (1.5)	1.6	0.7–4.2
C-reactive protein, mg/L	572	1.13 (2)	0.43	0.08–4.85

Abbreviation: SD, standard deviation.
[a] Of the 642 study subjects, 70 did not provide consent for blood sampling.
[b] Weight (kg)/height $(m)^2$.

leading edge of the first and second echogenic lines. All of the scans were recorded. The best-quality end diastolic image was captured in a longitudinal view that showed the bifurcation and was analyzed off-line. From this image, 3 measurements of the common carotid artery were taken at points 0.5 cm, 1 cm, and 2 cm below the bifurcation, and the average measurement was used. All measurements were carried out by a single sonographer on previously recorded scans. At the time of reading, the sonographer was unaware of the patients' identities and characteristics. Repeated measurements conducted by this sonographer in the same subjects (30 obese, 30 overweight, and 30 normal, randomly selected from the study children) gave an intraobserver coefficient of variation of 3.3%.

Statistical analysis

Data were expressed as percentages, mean values (with standard deviations), or median values (with 90% ranges). Ninety-five percent confidence intervals were calculated for proportions and means. Differences between groups were analyzed with the Wilcoxon rank-sum test for medians, the chi-squared test for proportions, and the *t* test for means. Simple linear models were used to assess the crude association of each predictor variable with IMT. Multiple regression models with IMT as the dependent variable were used for multivariate adjustment of predictor variables that had been shown to have *P* values less than 0.05 in univariate analysis. Different multiple regression models were used for each of the predictor variables that were highly correlated (body mass index and waist circumference). Results were defined as statistically significant when the *P* value (2-sided) was less than 0.05.

RESULTS

Of the 843 randomly selected subjects, 646 (76.6%) agreed to participate. Four subjects were excluded from the analysis because of daily alcohol consumption exceeding the predetermined limit (2 subjects) or chronic use of potentially hepatotoxic medications (2 subjects, 1 taking an antidepressant and the other taking an antiepileptic drug). Of the remaining 642 subjects, 70 did not provide consent for blood sampling. Females represented 50.6% of the study population.

Characteristics of study participants are shown in Table 1. According to International Obesity Task Force criteria, 30.5% (95% confidence interval (CI): 27.0, 34.3) of the adolescents were overweight and 13.5% (95% CI: 11.0, 16.4) were obese. No difference in the prevalence of overweight was observed between the sexes (31.1% (95% CI: 26.1, 36.5) in boys vs. 29.9% (95% CI: 25.0, 35.2) in girls; $P = 0.74$), while obesity was especially prevalent in boys (17.3% (95% CI: 13.3, 21.9) in boys vs. 9.9% (95% CI: 6.9, 13.7) in girls; $P = 0.006$).

The overall prevalence of fatty liver disease ($n = 642$) was 12.5% (95% CI: 10.0, 15.3), and prevalence was higher in boys than in girls ($P = 0.003$) (Figure 1). There was a graded relation between the prevalence of fatty liver disease and body mass index, with fatty liver disease reaching values well over 40% (43.7%, 95% CI: 33.1, 54.7) in obese

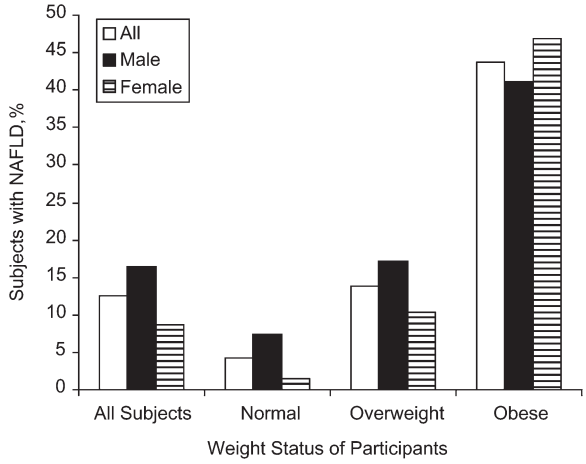

Figure 1. Prevalence of nonalcoholic fatty liver disease (NAFLD) in study participants (642 randomly selected adolescents aged 11–13 years) by sex and body weight, Reggio Calabria, Southern Italy, 2007–2008.

adolescents. Notably, 7.3% (95% CI: 3.8, 12.4) of boys and 1.5% (95% CI: 0.3, 4.4) of girls (overall, 4.2% (95% CI: 2.4, 6.8)) with fatty liver disease had normal weight. Subjects with fatty liver disease had significantly higher median body mass index, waist circumference, alanine aminotransferase, γ-glutamyl transpeptidase, triglycerides, insulin, HOMA-IR, C-reactive protein (all P's < 0.001), systolic blood pressure ($P = 0.01$), and diastolic blood pressure ($P = 0.004$) than subjects without fatty liver disease (Table 2). Moreover, adolescents with fatty liver disease had significantly lower high density lipoprotein cholesterol levels than those without fatty liver disease ($P < 0.001$). There were no significant differences in total cholesterol, low density lipoprotein cholesterol, or glucose concentrations by fatty liver disease status.

The mean IMT in the study population was 0.397 mm (95% CI: 0.395, 0.400), and IMT was higher in males than in females (mean IMT = 0.402 mm (95% CI: 0.399, 0.406) in boys vs. 0.392 mm (95% CI: 0.389, 0396) in girls; $P < 0.001$). IMT was higher in adolescents with fatty liver disease than in those without fatty liver disease (mean IMT = 0.417 mm (95% CI: 0.409, 0.425) vs. 0.395 mm (95% CI: 0.392, 0.397); $P < 0.001$).

Univariate analysis (Table 3) showed that the presence of fatty liver disease, increased body mass index, waist circumference, systolic blood pressure (all P's < 0.001), diastolic blood pressure ($P = 0.006$), γ-glutamyl transpeptidase ($P = 0.006$), alanine aminotransferase ($P = 0.007$), and C-reactive protein ($P = 0.008$) were associated with increased IMT. Only high density lipoprotein cholesterol ($P < 0.001$) was inversely associated with IMT, while total

Table 2. Characteristics of Study Participants According to the Absence or Presence of Nonalcoholic Fatty Liver Disease, Reggio Calabria, Southern Italy, 2007–2008

Characteristic	No NAFLD			NAFLD			*P* Value[a]
	No.	Median	90% Range	No.	Median	90% Range	
Body mass index[b]	562	20.3	15.5–28.1	80	25.6	17.3–31.6	<0.001
Waist circumference, cm	561	76.5	62.5–97	80	90.3	69–104.3	<0.001
Systolic blood pressure, mm Hg	562	105	87–124	80	109	90–125.5	0.01
Diastolic blood pressure, mm Hg	562	64	53–79	80	67	52.5–82	0.004
Alanine aminotransferase, IU/L	498	29	21–41	74	36	26–74	<0.001
γ-Glutamyl transpeptidase, IU/L	498	12	9–19	74	17	10–33	<0.001
Total cholesterol, mg/dL	498	156	119–205	74	154.5	115–217	0.73
High density lipoprotein cholesterol, mg/dL	498	51	34–74	74	45	31–63	<0.001
Low density lipoprotein cholesterol, mg/dL	498	88.5	60–133	74	89.5	56–146	0.23
Triglycerides, mg/dL	498	65.5	34–127	74	80	36–181	<0.001
Glucose, mg/dL	498	82	72–94	74	83	72–99	0.08
Insulin, mIU/L	498	7.7	3.4–17.7	74	12.6	4.6–33.2	<0.001
Homeostasis model assessment of insulin resistance	498	1.5	0.7–3.7	74	2.6	0.8–7.5	<0.001
C-reactive protein, mg/L	498	0.38	0.08–3.30	74	1.57	0.08–10.10	<0.001

Abbreviation: NAFLD, nonalcoholic fatty liver disease.
[a] Derived using the Wilcoxon rank-sum test.
[b] Weight (kg)/height $(m)^2$.

Table 3. Crude Association Between Characteristics of Study Participants and Intima-Media Thickness, Reggio Calabria, Southern Italy, 2007–2008

Characteristic	No. of Subjects	β[a]	95% Confidence Interval	*P* Value
Presence of nonalcoholic fatty liver disease (vs. absence)	642	0.0221	0.0143, 0.0299	<0.001
Body mass index[b]	642	0.0019	0.0013, 0.0025	<0.001
Waist circumference, cm	641	0.0007	0.0005, 0.0009	<0.001
Systolic blood pressure, mm Hg	642	0.0006	0.0003, 0.0008	<0.001
Diastolic blood pressure, mm Hg	642	0.0005	0.0001, 0.0008	0.006
Alanine aminotransferase, IU/L	572	0.0005	0.0001, 0.0008	0.007
γ-Glutamyl transpeptidase, IU/L	572	0.0008	0.0002, 0.0013	0.006
Total cholesterol, mg/dL	572	−0.0001	−0.0002, 0.0000	0.158
High density lipoprotein cholesterol, mg/dL	572	−0.0004	−0.0006, −0.0002	<0.001
Low density lipoprotein cholesterol, mg/dL	572	−0.00002	−0.00014, 0.00010	0.722
Triglycerides, mg/dL	572	0.00005	−0.00003, 0.00013	0.214
Glucose, mg/dL	572	0.0003	−0.0001, 0.0007	0.149
Insulin, mIU/L	572	0.0003	−0.0001, 0.0007	0.149
Homeostasis model assessment of insulin resistance	572	0.0013	−0.0005, 0.0032	0.164
C-reactive protein, mg/L	572	0.0019	0.0005, 0.0033	0.008

[a] Regression coefficients represent, for the dichotomous variable (nonalcoholic fatty liver disease), the average difference in intima-media thickness (mm) as compared with the reference group and, for continuous variables, the change in intima-media thickness (mm) per unit increase in the characteristic.
[b] Weight (kg)/height (m)^2.

cholesterol, low density lipoprotein cholesterol, triglycerides, glucose, insulin, and HOMA-IR were not associated with increased IMT.

After adjustment for confounders in the multivariate analysis (Table 4), only fatty liver disease (difference in IMT with the presence of fatty liver disease = 0.0147 mm (95% CI: 0.0054, 0.0240)), body mass index (difference per unit increase = 0.0011 mm (95% CI: 0.0003, 0.0019)), waist circumference (difference per 1-cm increase = 0.0004 mm (95% CI: 0.0001, 0.0007)), and systolic blood pressure

Table 4. Multivariate Analysis of Relations Between Selected Clinical Characteristics of Participants and Intima-Media Thickness ($n = 572$), Reggio Calabria, Southern Italy, 2007–2008

Characteristic	β[a]	95% Confidence Interval	*P* Value
Presence of nonalcoholic fatty liver disease (vs. absence)	0.0147	0.0054, 0.0240	0.002
Body mass index[b,c]	0.0011	0.0003, 0.0019	0.004
Waist circumference, cm[c]	0.0004	0.0001, 0.0007	0.003
Alanine aminotransferase, IU/L	0.0000	−0.0004, 0.0004	0.836
γ-Glutamyl transpeptidase, IU/L	−0.0001	−0.0008, 0.0005	0.662
High density lipoprotein cholesterol	−0.0001	−0.0004, 0.0001	0.263
C-reactive protein, mg/dL	−0.0001	−0.0016, 0.0014	0.868
Systolic blood pressure, mm Hg	0.0004	0.0001, 0.0007	0.005
Diastolic blood pressure, mm Hg	−0.0001	−0.0005, 0.0003	0.613

[a] Regression coefficients represent the average estimated increase in intima-media thickness (mm) per unit increase in the characteristic or the presence (vs. absence) of the characteristic. Results were simultaneously and reciprocally adjusted for all other variables shown in the table (except waist circumference) and for sex.
[b] Weight (kg)/height (m)^2.
[c] Body mass index and waist circumference were added independently to the model because of the high correlation between these 2 variables.

(difference per 1-mm Hg increase = 0.0004 mm (95% CI: 0.0001, 0.0007)) remained significantly associated with IMT.

DISCUSSION

The prevalence of overweight and obesity among adolescents in our study population (44.1%, 95% CI: 40.2, 48.0) was very high. It was roughly similar to the prevalence reported for other pediatric populations in southern Italy (26, 27) and in the United States, where the levels of childhood overweight and obesity are among the highest recorded in the world (28).

The present data on fatty liver disease are among the few that have been reported for an adolescent population in the age range of 12–13 years. We found an overall prevalence of ultrasonographically diagnosed "bright liver" of 12.5% (95% CI: 10.0, 15.3), which could not be attributed to excessive alcohol consumption or intake of medications potentially steatogenic to the liver.

Not surprisingly, the prevalence of fatty liver disease increased progressively from normal-weight subjects to overweight subjects to obese subjects, affecting almost 1 in 2 adolescents within the latter category. In fact, fatty liver disease is strongly associated with overweight/obesity (29–32) and other markers of the metabolic syndrome (6, 33–35) and is now considered by many investigators to be the hepatic expression of the metabolic syndrome (4, 36, 37).

In a previous study, the prevalence of fatty liver disease was determined ultrasonographically in a population of 810 northern Japanese children (38). The overall prevalence of fatty liver disease was 2.6% and was higher for boys (3.4%) than for girls (1.8%), although the difference was not statistically significant. Fatty liver disease was found in children as young as 6 years of age. Zhou et al. (39) published a report on the prevalence of fatty liver disease in a general population in southern China. The overall study population of 3,543 subjects included 379 children and youths aged 7–18 years. The prevalence of fatty liver disease in this age group, determined by real-time ultrasonography, was 1.3% and was 3 times higher for boys (1.9%) than for girls (0.6%). Imhof et al. (40) studied a population-based sample of 378 adolescents aged 12–20 years who were randomly selected from the general population of a small town in southern Germany. The prevalence of fatty liver disease in these study participants was approximately 5%, almost all of it found among overweight/obese subjects.

Thus, the prevalence of fatty liver disease in our study appears to be the highest reported so far for an adolescent population. This is not unexpected in view of the very high prevalence of overweight and obesity in our study population and the close association of these conditions with fatty liver disease. It should also be considered that the prevalence of fatty liver disease that we found is a likely underestimation of the true, histologically defined prevalence of fatty liver disease. In fact, mild forms of fatty liver disease cannot be visualized by echography, because this method does not reliably detect steatosis affecting less than 33% of hepatocytes (23).

Another main objective of the present study was to identify increased carotid artery IMT as an early marker of atherosclerosis and to verify whether cardiovascular risk factors or components of the metabolic syndrome and fatty liver disease are associated with increased IMT in our sample. Increased IMT has been associated with cardiovascular risk factors as well as the presence and severity of coronary atherosclerosis and cardiovascular disease (16, 41, 42). It has also been reported in children with diabetes, hypertension, and obesity (43, 44). In particular, several studies comparing obese and nonobese children have found significant differences in IMT (45, 46).

We found that increased body mass index or waist circumference, systolic blood pressure, and the presence of fatty liver disease were independently associated with increased IMT, allowing us to consider them as markers of increased IMT and cardiovascular risk. To our knowledge, this is the first report of an independent association between fatty liver disease and increased IMT in a population-based study of adolescent subjects.

The difference in IMT between subjects with and without fatty liver disease was narrow (median value of 0.417 mm (95% CI: 0.0.409, 0.425) vs. 0.395 mm (95% CI: 0.392, 0397); $P < 0.001$). Nevertheless, our results show that fatty liver disease is a highly significant marker of increased IMT. As regards a possible link between fatty liver disease and increased IMT and cardiovascular risk, it has been suggested that fatty liver disease might contribute to accelerated atherosclerosis through increased oxidative stress, chronic subclinical inflammation, and decreased liver production of cytokines with antiatherogenic properties (8).

Regarding the other factors that were independently associated with increased IMT, several articles in the literature have reported associations of body mass index (47, 48) and/or systolic blood pressure (49–51) with increased IMT, in adults as well as adolescents. Reducing systolic blood pressure has even been shown to result in regression of carotid IMT and a decrease in left ventricular mass among persons with type 2 diabetes (52). On the other hand, it is surprising that components of the metabolic syndrome (5), such as dyslipidemia, elevated fasting glucose levels, and insulin resistance—which are established risk factors for cardiovascular disease—did not show a significant association with increased IMT in our study.

There are different possible explanations for this lack of association. Some risk factors may only affect IMT upon prolonged cumulative exposure, which may not have occurred in our participants, given their young ages (51). Moreover, as other investigators have already discussed (53), changes in carotid IMT may not reflect the effects of all cardiovascular risk factors. That is, not all cardiovascular risk factors may cause increased IMT; rather, they may affect later stages of the atherosclerotic process, such as plaque formation.

Our study presents some limitations that should be considered: its cross-sectional nature, which precluded the possibility of establishing temporal relations, and the lack of biopsy-proven fatty liver disease, which made it impossible to evaluate the association of IMT with severity of liver disease. In addition, we did not test for hepatitis B virus

or hepatitis C virus, since vaccination against the former is mandatory in Italy and the prevalence of the latter was expected to be extremely low in the age group included in our study. Indeed, in a previous study from our group performed in a geographic area close to Reggio Calabria (54), we could not identify a single adolescent with hepatitis C virus infection. Among the strengths of this study was the relatively large sample size (appropriate for studying a continuous outcome such as IMT), its novelty, and the consideration of important confounding factors.

Notwithstanding its limitations, our study showed a high prevalence of fatty liver disease in a sample of adolescents from southern Italy and identified fatty liver disease, body mass index, and systolic blood pressure as independent markers for increased IMT.

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Author affiliations: Madonna della Consolazione Hospital, Reggio Calabria, Italy (Carmelo Antonio Caserta, Gaspare Maria Pendino, Angela Amante); Provincial Health Authority No. 5, Reggio Calabria, Italy (Pasquale Surace, Carmelo Vacalebre, Maria Teresa Fiorillo); Hepatology Association of Calabria, Reggio Calabria, Italy (Arianna Messineo, Monica Surace, Saverio Alicante); Department of Clinical Epidemiology and Guidelines, Advanced Health Institute, Rome, Italy (Rodolfo Cotichini, Francesco Rosmini, Alfonso Mele, Fabrizio Marcucci); and Department of Internal Medicine, School of Medicine, University of Milan, Milan, Italy (Massimo Zuin).

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